

DNA Technologies and Production Markers

Alison Van Eenennaam, PhD

Cooperative Extension Specialist Animal Biotechnology and Genomics University of California, Davis, USA

Email: alvaneenennaam@ucdavis.edu

http://animalscience.ucdavis.edu/animalbiotech





Overview



- Background information on genetic improvement and DNA marker technology
- Improvements in genetic gain possible from including DNA information into genetic predictions
- Genetic tests that are on the market for production traits
- DNA testing for recessive genetic defects



Traditional animal breeding on the basis of phenotype (appearance) can be very successful

















However breeding based on objective performance recording has been spectacularly successful.....



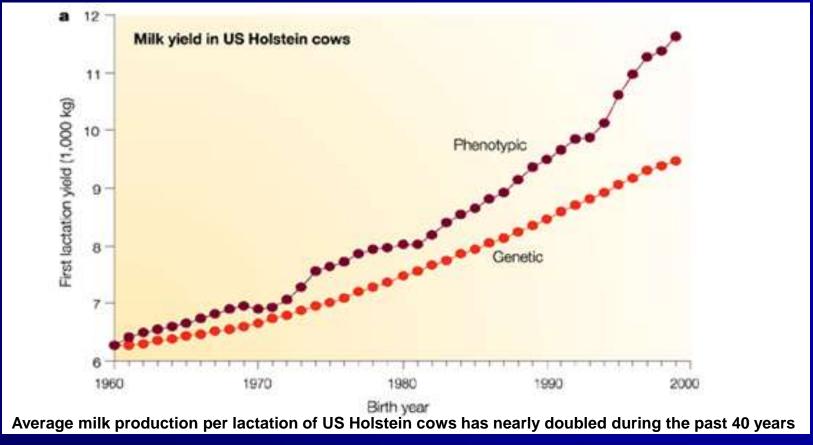


Image from: Dekkers, J.C.M, and F. Hospital, 2002. The use of molecular genetics in the improvement of agricultural populations. *Nature* Reviews Genetics 3: 22-32



Should you use this bull?









In the absence of any other information selection can only be based on appearance or reputation...









But what you would really like to know is the future performance of his unborn calves !!





What if I told you these calves belonged to the bull?









What if I told you these were his daughters' calves?





What if I told you this was a typical steak from his progeny?







Animal breeders use records of an animals own performance and that of its relatives to predict an animal's genetic merit or estimated breeding value

1/2 x EBV = Expected Progeny Difference (EPD)









An animal's **breeding value** is an estimate of its genetic merit, half of which will be passed on to its progeny (EPD). While we will never know the exact breeding value, for performance traits it is possible to make good estimates based on performance records of the animal and its relatives. These are called Estimated Breeding Values

Calculating EBVs

In the calculation of EBVs, the performance of individual animals within a contemporary group is directly compared to the average of other animals in that group. A contemporary group consists of animals of the same sex and age class within a herd, run under the same management conditions and treated equally. Indirect comparisons are made between animals reared in different contemporary groups, through the use of pedigree links between the groups.





Challenge for breeders is to accurately identify those individuals that have the best true breeding values

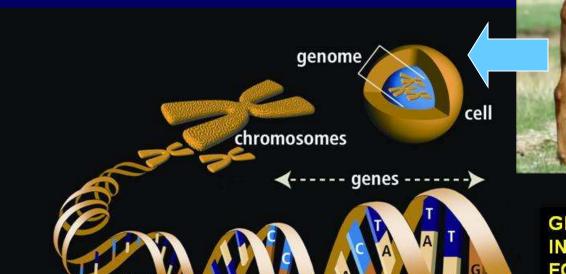


- Records more are better
- Would ideally like records before selection
- Need to know genetic relationship between what you re measuring (e.g. an ultrasound scan) and the trait you get paid for (e.g. marbling)
- Perhaps DNA information can help improve the accuracy of breeding values if they have been shown to work in Wagyu!









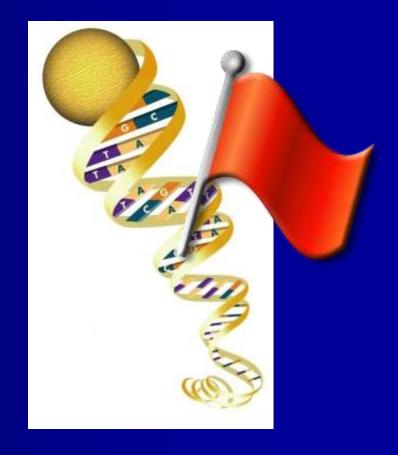
GENES CONTAIN INSTRUCTIONS FOR MAKING PROTEINS

PROTEINS ARE THE BUILDING BLOCKS OF LIFE AND COLLECTIVELY ACT TO DETERMINE PHENOTYPE

proteins ...

SNP (Single Nucleotide Polymorphism)

A DNA sequence variation that varies sufficiently between individuals that its inheritance can be tracked through families

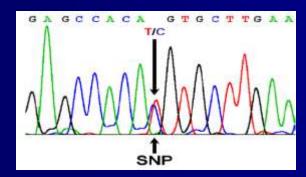


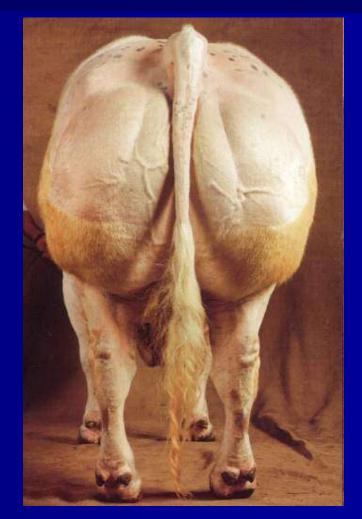


What is a Genetic Marker?



A DNA sequence variation that has been associated with a given trait in one or more populations





Wagyu 10/27/2011



What is needed for develop a DNA test?



TRAINING

- Population
- Phenotypes
- Genotypes

Training = Need a population where genetic markers have been associated with production traits in BREED of interest

Prediction = the results of training can then be used to predict the genetic merit of new animals based on their DNA genotype alone



There are DNA-markers for simple traits



DNA test result is highly predicitive

- Coat color
- Polled/horned staus
- Certain genetic diseases (e.g. "curly calf")
- SCD (Prescribe Genomics, \$90 per test)
- "Exon 5" (Prescribe Genomics, \$100/test)



Wagyu-specific DNA-makers for simple traits



SCD - Stearoyl-CoA desaturase (AA, VA, VV)

- "the percentage of beef that is "not delicious" has increased as a result of increased fat of a high melting temperature
- ~ 18 % AA, 74% VA, 8% VV

Exon 5 – growth hormone (AA,AB,BB,BC,CC)

- Only found in Wagyu,
- Purportedly associated with marbling
- No more information on frequency

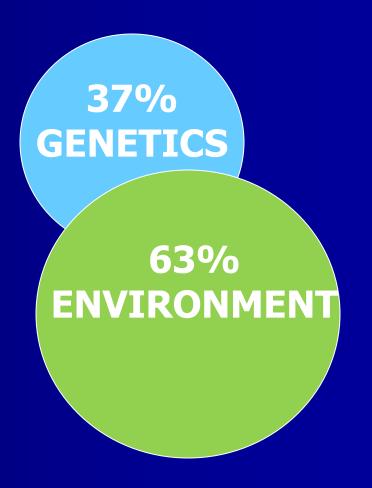


SIMPLE TRAITS
e.g. Coat Color
Double muscling

COMPLEX TRAITS e.g. Marbling ($h^2 = 0.37$)



100 %
GENETICS









Merial, Quantum sign leptin test pact.(Business Report)

Publication: Feedstuffs

Publication Date: 04-AUG-03

Ads by Google

B-Bridge International ELISA Assays for Metabolic Studies Adipocytokines, esRAGE, HGF

Need College Scholarship? Find Update, Fast, Easy & Free! 2.4 million Scholarships. Apply Now

Antique Sheep Bid on Antique Sheep. Shop Victoriously!

<u>Public Art Projects</u> Custom designed fiberglass animals for Public Art Fundraising events.

How to access the full article: Free access to all articles is available courtesy of your local library. To access the full article click the "See the full article" button below. You will need your US library barcode or password.

COPYRIGHT 2003 Miller Publishing Company, Inc.

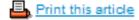
SASKATOON, SASK., and DULUTH, GA. -- Quantum Genetics Inc. and Merial Ltd. announced July 23 that they have entered into a global marketing agreement to provide Merial with exclusive rights to market Quantum's new patent-pending DNA test to determine an animal's leptin genotype.

The leptin protein has been demonstrated...

COPYRIGHT 2003 Miller Publishing Company, Inc.

InfoTrac OneFile®











Wagyu 10/27/2011

Igenity L was a single T/C SNP test for Leptin



Advanced technology. Advanced knowledge.

What if there was a test that could tell you - in advance - an animal's genetic potential for energy utilization or carcass quality? You'd have the advantage of knowing an animal's potential now, instead of discovering it later through success or failure in the milk string or when the animal goes to market.

Researchers have discovered the specific gene that carries the code for the production of a protein called leptin. Leptin is associated with an animal's potential for appetite and energy utilization, among other things.

- · For dairy cattle this translates directly into maximum dry matter intake (DMI) and peak milk production.
- · For beef cattle it relates to days on feed and carcass quality.

The IGENITY" L Test identifies leptin genotype (L-tt", L-ct" or L-cc"). Now you have another important resource to help you breed, feed, sort, manage and market cattle at an optimum level.

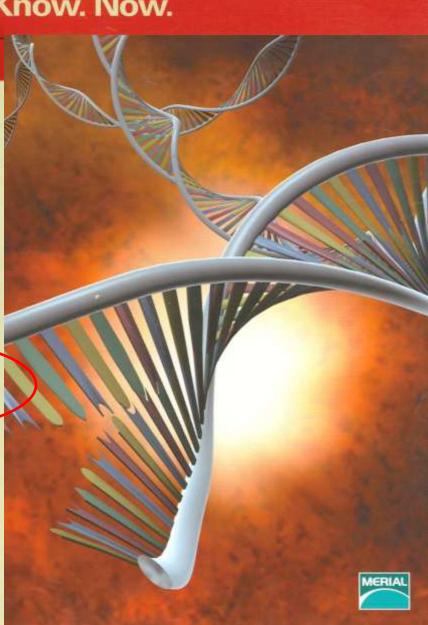
Igenity™ L. Know. Now.

- · Leptin is involved in the regulation of feed intake, energy balance, milk production, milk components, marbling scores, puberty, fertility and immune functions.
- · Knowing an animal's leptin genotype empowers you to make more informed, strategic management decisions regarding breeding and nutrition.
- The IGENITY L Test identifies an animal's leptin genotype at the DNA level, with 100% accuracy.

Market your cattle with the IGENITY L logo system:



L-ct.



BOVIGEN....



MARBLING

GeneSTAR™ Marbling is a DNA genetic marker test offered by

Bovigen Solutions,™ L.L.C. for a major gene (Thyroglobulin) associated with marbling. This marker provides beef producers the opportunity to determine an animal's potential for improved marbling more accurately and at an earlier age.

Marbling	Resuits ("Choice)	Difference
AND DESCRIPTION OF THE PERSON NAMED IN COLUMN TWO IS NOT THE PERSON NAMED IN COLUMN TWO IS NAMED IN COLUM		

	0	太	太太	*************
Yearling Fed	58%	62%	74%	16%
Calf Fed	34%	41%	53%	19%
NBCEC Trial	47%	54%	64%	17%

In independent trials on over 1500 head, 2-STAR animals produced 16-19% more choice carcasses than 0-STAR animals. Equally important was the effect that the proportion of carcasses qualifying for premium Quality Grades doubled (21% vs. 10%) in 2-STAR carcasses versus 0-STAR.

Trial was a study conducted by an independent third party.

- National Beef Cattle Evaluation Consortium.

LEGEND

The results of the GeneSTAR** tests are reported as:

2-STARS = two copies of the desired gene

1-STAR = one copy of each form of the gene

O O-STARS = no copies of the desired gene

"DNA technology, although in its infancy in the beef cattle industry, can help us by identifying genes that effect marbling and tenderness. Tenderness seems to be one that interests many who produce and have concerns for improving end product value for our consumers. The GeneSTAR** testing at Summitterst has identified a number of cattle homozygous for marbling and tenderness genes. Having done that, we added market value to our cattle and new customers, even from other breeds of cattle, with tenderness concerns."

- Henry Bergfeld, Summittrest Farms

HOW DO I USE THIS IN MY BREEDING PROGRAM?

Using 2-STAR sires is the quickest way to influence the presence of these genes in a herd. In addition, selecting 2-STAR females will more rapidly increase the frequency of the positive forms of these genes. Overall, selecting 2-STAR animals with appropriate EPDs and good structural and breeding soundness is the recommended way forward.

1	Mat	ing	Desi	gn
_	_		_	

Sire is	Dam is	0	of Proge	ny ****
OOO	<u>。</u> 本本	100% 50%	50% 100%	
蒸 蒸	<u>。</u> 杰杰	50% 25%	50% 50% 50%	25% 50%
本本 本本 本本	.Q. 杰.本		100% 50%	50% 100%



Which would you rather have???



 A bull that is 'homozygous' for a positive genetic variant with a low-accuracy marbling EPD of +3, or



 Or an unrelated bull carrying no copies of that genetic variant with a low-accuracy marbling EPD of +3





Both are important!!

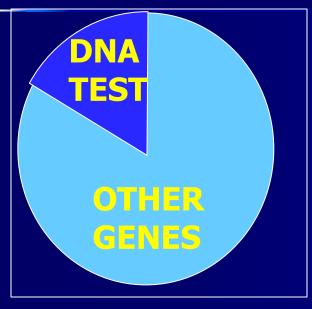


- The 'homozygous' bull is a source of favorable alleles (genetic variant) of the gene. Can eventually be used to create homozygous calves
- The other bull contributes favorable unmarked alleles of other genes, which will improve the frequency of other desirable alleles for the trait.
- Breeding the marker-associated form of the gene into the bull that has no copies should improve the trait by combining all of the good forms of the genes together in one animal



DNA TEST FOR MARBLING EXPLAINS SOME % (r²) OF GENETIC VARIATION





37% GENETICS

63% ENVIRONMENT

EPD estimates all genes



Recap of early product offerings



- Single gene tests reported as actual results of genotyping (** or AT)
- Great deal of explanation of what the gene was and how it had its effect
- DNA-test billed as 100% accurate
- The need for third-party validation of commercial tests becomes evident
- A lot of emphasis was put on a single SNP



First multi-gene test arrives



TENDERNESS

GeneSTAR Tenderness was the first multi-gene single trait DNA test commercially available to the beef industry. It combines test results for several markers and genes for the same trait. The test is based, in part, on two genes involved in the post-mortem tenderization process: Calpastatin and Calpain. Calpain is an enzyme which weakens muscle fibers thereby making the fibers more tender. Calpastatin is an enzyme which inhibits the post-mortem tenderization process by inhibiting the effects of Calpain.

GeneSTAR Tenderness Senetic Progeny Difference

T1	12	T3	GPD Lbs. Shear Force
	杰杰	**	-2.2 -1.8 -1.4
杰	*	#.# #	-1,8 -1,4 -1,0
	0	北.北 北	-1.5 -1.1 -0.7
	本 本	±.± ±	-1.8 -1.4 -1.0
*	杰	**	-1.5 -1.1 -0.7
	0	**	-1.1 -0.7 -0.3
	态态	2. 12. 12. 12. 12. 12. 12. 12. 12. 12. 1	-1.5 -1.1 -0.7
0	*	2. A.	-1.2 -0.8 -0.3
	<u>*</u>	## ##	-0.8 -0.4 -0.0

 Values derived from Independent Validation Population

MATING DESIGN

As an added tool to GPDs, the Mating Design table shows the predictable results which can be obtained through planned breeding decisions utilizing DNA test results from Bovigen.

SIRE	DAM	% o	ny	
IS	IS	**	*	0
女女	女女	100%		
食食	*	50%	50%	
会会	0		100%	
*	**	50%	50%	
*	*	25%	50%	25%
*	0		50%	50%
0	女女		100%	
0	*		50%	50%
0	0			100%

This Mating Design illustrates Mendelian Heredity and the probability of results from mating a 0,1 or 2 STAR Sire and Dam.

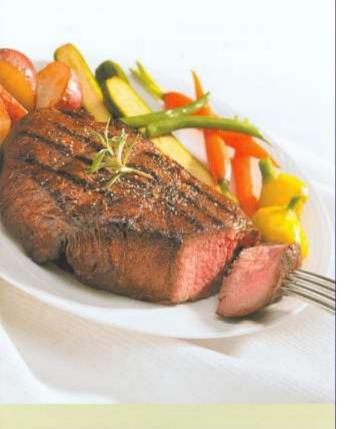
DNA tests should be considered additional information to be used in conjunction with available performance data for each animal. When comparing two bulls with similar EPDs, GeneSTAR* GPD* results can be used to further evaluate an animal's true genetic potential.

BOVIGEN COMPANY PROFILE

Bovigen, as is an innovative-driven genomics company developing a portfolio of world class DNA technological products. Our mission is to promote excellence in the beef industry through

IGENITY® TenderGENE™

A practical, powerful tenderness selection tool.



What do you bring to the table?



IGENITY® TenderGENE.™

The inside track to tender beef.

IGENITY® TenderGENE" profiles the tenderness potential of an animal and is:

- A powerful and comprehensive tenderness selection tool
- Fully validated by the National Beef Cattle Evaluation Consortium (NBCEC)
- Informative in all breed types, including Bos indicus

IGENITY TenderGENE analyzes multiple markers associated with the calpain and calpastatin genes. Both of these genes are important for profiling an animal's potential for tenderness:

- Calpain enzymes weaken muscle fibers during the post-mortem aging process.
- Calpastatin interacts with the calpain enzymes to impact overall tenderness.

Together they have a significant effect on tenderness as measured by Warner-Bratzler Shear Force (WBSF). In fact, in independently validated research, the calpain and calpastatin markers included in IGENITY TenderGENE are associated with improving tenderness as much as 2.3 lb.

The benefits of IGENITY TenderGENE are proven.

- IGENITY TenderGENE includes markers discovered by researchers at the U.S. Meat Animal Research Center (MARC) and the University of Guelph.
- IGENITY TenderGENE is a powerful tenderness test that is fully validated by the NBCEC.
- Validation work involved cattle from the NCBA National Carcass Merit Project.
- Research included more than 1,200 animals, representing all breed types.
- Because of the favorable effect on WBSF of up to 2.3 lb, selecting for tenderness using IGENITY TenderGENE can help ensure a quality eating experience for consumers.

Learn how this inside information can give you a competitive edge. Call 1-877-IGENITY.









	D (Market	NA Tes	t Resu	lts		
7	FE1	FE2	FE3	FE4	Total	GPD
Feed Efficiency	. 文. 支	*	*	大大	6	-3.2

In this example, the results describe an animal for Feed Efficiency. The genes identified for Feed Efficiency thus far, identify cattle with lower Net Feed Intake (NFI). Cattle with lower NFI will eat less without sacrificing ADG or any other performance trait. The animal in this example will consume 3.2 lbs less feed per day and still have the same ADG and Marbling potential as the rest of his contemporary group. Simply put, the more STARS for Feed Efficiency, the less feed consumed without sacrificing any performance!!!

Ge	M DNA	Test R	esults		
	T1	T2	T3	Total	GPD
Tenderness	***	*	***	5	2

In this example, the results describe an animal for its Tenderness potential. Nationwide, tenderness is measured by Warner-Bratzler Shear Force, or the pounds needed to cut a core sample from a ribeye. The animal in this example will produce a ribeye that is 2.0 pounds more tender than an animal without the STARS for tenderness.

The GeneSTAR Tenderness panel continues to lead the industry in identification of markers for one of the most valuable traits for producing a quality eating experience. GeneSTAR animals that are homozygous for all the Tenderness markers (T1, T2, T3) show a difference in Tenderness that virtually eliminates the "unsatisfactory eating experience" that plagues nearly 25% of all carcasses compared to an animal devoid of these genes.

	GeneSt R	DNA Tes	st Result	S	era.	
- II	QG1	QG2	QG3	QG4	Total	GPD
Quality Grade	***	*	支支		6	27.42

In this example, the results describe an animal for Quality Grade. The genes identified thus far for Quality Grade work across all breeds and identify animals with a greater chance of grading Choice or higher. The animal in this example has a 27.42% greater chance of grading Choice or higher than an animal with out the STARS for Quality Grade.

1.877.BEEF.DNA www.bovigen.com 250 Plauche St. Harahan, LA 70123



FEATURES

- Contains 128 DNA markers where each marker is highly associated with expression of marbling score
- Measures the cumulative effects of all 128 markers associated with marbling
- Results are expressed as the Molecular Genetic Value (MGV) which can be utilized to rank animals by their genetic potential
- Animals can be tested at any age
- Validated in Angus (validation) in other breeds is underway)

BENEFITS

- The most powerful and comprehensive DNA selection tool currently available for marbling
- Accounts for a significant. proportion of total observed genetic variation for marbling
- Results are easy to utilize and incorporate into any existing breeding program
- Can be used to make early. selection and breeding decisions
- Provides accurate and reliable results for ranking and/or selection of animals



TRU-MARBLING

One in a series of break-through products that will advance breeding practices in the cattle industry, Tru-Marbling" is a powerful and comprehensive DNA selection tool that can determine the genetic potential of animals to express marbling. In a collaborative research program between Cargill and MMI Genomics, an innovative scientific approach was used on over 4000 feedlot animals to identify the majority of regions throughout the bovine genome that have an effect on this economically important trait.

Tru-Marbling " is a DNA-based genetic test that contains a panel of 128 unique DNA markers. each one highly associated with the expression for marbling score and quality grade. By measuring the cumulative effects for each of these 128 markers. Tru-Morbling" accounts for a significant proportion of the total genetic variation for this complex metabolic trait—the first DNA-based product to do so!

Tru-Marbling" is an advanced and revolutionary tool that will allow cattle producers to make early breeding decisions that increase the accuracy of selection and decrease the age at which animals can be selected.

The results? Rapid improvement of marbling within herds and the ability to determine the "Ini" genetic potential of animals.

PROVEN RESULTS

Tru-Marbling** has been validated in both commercial cross-bred feeder cattle populations and in Angus cattle.

The validation in Angus was conducted using samples from the National Carcass Merit Project. representing Angus sires bred to Angus-based commercial cows. While this is a small population of animals, the data indicate that Tru-Marbling Maccounts for 70% of the genetic variation observed in this population.

No. of samples: Heritability*: No. of markers: Phenotypic variation explained (R1)++: As a percent of Heritability

Explains 70% of 414 the genetic 0.36 128 variation in 0.25 70% marbling with

* Angus National Cattle Evaluation, Spring 2007
** estimated from a model that included contemporary group

128 markers Tru-Marbling" has also been validated against comme This study utilized 89 marbling markers that are a subset of the 128 markers used in the purebred Angus population. The data clearly demonstrates the relationship between quality grade and average Molecular Genetic Value (MGV).

A STATE OF THE PARTY OF THE PAR	Water I	STEERS	Town.	H	EIFERS	
Quality Grade	Number	Seftetal	Arg. MGV	Number	% of total	Avg. MGV
Prime	,	0.15	14	5	0.1%	14
High Chaice	73	1,1%	20	78	1.3%	12
Medium Choice	475	5.8%	34	423	7.0%	9
Low Chalice	2006	24.3%	7.	1526	25.2%	0
High Select	3980	48.3%	- 6	2835	46.9%	-9
Low Select	1344	16.3%	34-	915	15.1%	-20
No Rell	337	4.1%	-20	268	4,4%	-23
Totals	#255	100.0%		6950	100.0%	

14,305 head tested from 6 commercial feedlots

Unlike other products, Tru-Matisling ** MGVs account for a highly significant amount of variation for marbling

TRU-TENDERNESS

One in a series of break-through products that will advance breeding practices in the cattle industry, Tru-Tenderness* is a powerful and comprehensive DNA selection tool that can determine the genetic potential of animals to produce tender meat. In a collaborative research program between Cargill and MMI Genomics, an innovative scientific approach was used on over 4000 feedlot animals to identify the majority of regions throughout the bovine genome that have an effect on this valuable consumer trait.

Tru-Tendemess" is a DNA-based generic test that contains a panel of 11 unique DNA markets. each one highly associated with expression for tender meet. By measuring the cumulative effects for each of these 11 markers, Tru-Tenderness* accounts for a substantial proportion of the total genetic variation for this complex metabolic trait.

Since tenderness can only be measured in harvested cattle it in difficult, time consuming and expensive to make genetic progress for this trait using traditional genetic improvement tools. In-Tendemens" changes this paradigm by allowing producers to accurately assess the genetic potential of their breeding stock to produce tender meat, in addition, Try Tindemess* also shortens the interval for making genetic progress because it can be used to test animals of any age.

The Tendemeis" is an advanced and revolutionary tool that will allow cattle producers to make early breeding decisions that increase the accuracy of selection and decrease the age in which animals can be selected.

The results? Rapid improvement of tendemess within heads and the ability to determine the "Tru" genetic potential of animals.

PROVEN RESULTS

Tru-Tenderness* has been validated in Angus using samples from the National Carcass Merit Project, representing Angus sires bred to Angus-based commercial cows. While this is a small population of animals, the data indicate that Tru-Tenderness** accounts for 100% of the genetic variation observed in this population as measured by Warner-Bratzler shear force.

No. of samples: Heritability*: 0.35 No. of markers: 11 Phenotypic variation explained (R2)**: 0.38 As a percent of Heritability 100% * as estimated in Minick et al. 2004, Can. J. Anim. Sci. 84:59

" eximated from a model that included corresposity gra-

Explains 100% of the genetic variation in tenderness



Results reported on 1-10 scale



The IGENITY profile.

Comprehensive.

Comprehensive. Practical. Powerful.

It's easy to understand an IGENITY profile.

One of the greatest values of the IGENITY profile is that all results are integrated and provided in one single profile, similar to the report shown here.

	IGENITY Profile												
Animal ID	M/F	Breed	Sample Barcode #	Tenderness	Red/Black Coat Color	Heifer Pregnancy Rate	Yield Grade	Ribeye Area	Hot Carcass Weight	Quality Grade	Stayability	BVD-PI	Polled
701	М		nv011507_01	10	ED/ED	9	8	8	8	8	8	POS	Homozygous
702	F		nv011507_02	6	ED/ED	3	4	6	5	7	6	NEG	
704	F	٠	nv011507_04	10	ED/E	7	3	8	6	6	6	POS	
705	F	-	nv011507_05	6	ED/D	3	4	6	5	5	3	NEG	

Results reported as a MGV

MOLECULAR GENETIC VALUE (MGV) REPORT

NAME Legends of the West - Angus Farm

13457 Trujillo Creek Road

Aguilar, CO

81020

ADDRESS

REPORT DATE 08/23/2006
ORIGINAL REPORT 08/23/2006

CASE ID MT-0059302S
CUSTOMER LWAF - Legends of the West Angus Farm

CONTACT Wes Johnson, Foreman

TRU-MARBLING and TRU-TENDERNESS REPORT

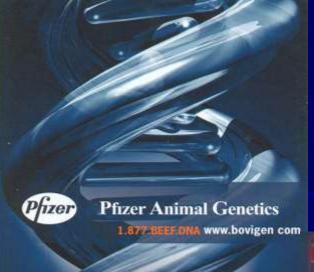
BREED	NAME	REG#	TAG/ TATTOO	SEX	BORN	SAMPLE ID	MARBLING MGV	TENDERNESS MGV
Angus	AF Paul Bunyan	19352178	AZ-105	М	06/14/2004	0539812G	-28.61	0.92
Angus	AF Casey Jones	19352211	AZ-112	М	05/29/2004	0539813G	-19.41	-3.98
Angus	AF Alamo Rising	19352385	AZ-146	М	06/19/2005	0539814G	42.79	-0.75
Angus	AF Geronimo	19352448	AZ-175	М	12/20/2004	0539815G	70.15	-4.40

All four animals represented in the above test result have Tru-MarblingTM and Tru-TendernessTM MGVs. All MGVs within a trait have the same accuracy because every animal has the same number of markers in the prediction of genetic potential.

Paul Bunyan and Casey Jones are predicted to grade in the low Select and No Roll USDA grades because of their large negative marbling MGVs. Alamo Rising and Geronimo are predicted to grade in the USDA grades of high Choice and Prime because of their high MGVs for marbling. MGVs near 0 are expected to grade in the high Select USDA grading category.

Paul Bunyan is predicted to produce tough meat with the larger MGV for *Tru-Tenderness*™. Casey Jones and Geronimo are predicted to produce very tender meat. Alamo Rising is predicted to produce acceptable tenderness, bordering on slightly tough.

These results can be used to rank bulls for their genetic potential for marbling and tenderness.



Pfizer Animal Genetics aquired Bovigen — results reported as GPD

dem Shift

Feed Efficiency

GeneSTAR Feed Efficiency is based on Net Feed Intoke (NFI). NFI is the difference between an animal's actual feed intake and the amount of feed an animal is expected to eat based on its size composition and growth rate.

Coives that eat less than expected will have a negative NFI
Negative NFI = Superior Feed Efficiency
Coives that eat more than expected will have a positive NFI
Positive NFI = Inferior Feed Efficiency

GeneSTAR GPD Feed Efficiency

Expected Liss consumed per day

White TITE	GPD	1231	GPD	TTT GPD
4020	3.94	CERE	-8.00	DECEMBER 437
AKES.	3.74	KERK	100	9 2 2 2 4 2 H
1111	1.02	1222	4.0	2 2 2 2 4.58
2023	132	KAYE	4.18	93111 -131
2234	-13	KENT	-100	ATT 421
2270	1.29	SERVE	-246	0.010 -230
2282	1.00	1261	310	93332 420
PARK	-1.26	2241	-286	2222-447
5235	-5.84	5258	-2.86	5000 -LAN
2122	EM	SILE	-2.66	2222 -145
4121	-0.80	22.40	-2.33	STATE -140
ATTE	24	1111	4.8	141
REAL PROPERTY.	429	22.12	2.2	E100 14
23.86	3.97	5 0 6 6	15.88	E112 -1-11
47.56	2.89	STREET	-1.76	2011 E - 1.36
2783	110	1382	COM.	100000 1-20
SHAR	231	13.02	-1.24	DECEMBER 18.34
2000	-2.11	5100	1.50	22.22 4.0
2121	2.11	1222	-1.82	FEET 4.82
2022	-126	1821	-1.3	11111 47
2022	-1.67	2000	-1.06	E1722 -0.48
2413	-0.87	2014	-1.27	DESCRIPTION OF THE PERSON NAMED IN COLUMN
2810	-08	50.00	-1.89	SEE 2.15
2 1 1 1	CAR	2000	-0.40	E1110 4.16
ZAKE	188	1 4 6 3	-1.03	2000 4H
22.65	-1.41	1,000	-0.81	BARR 422
	400		-0.59	TOTAL 14

Sensitif

Tenderness

GeneSTAR Tenderness was the first multi-gene single trait DNA test commercially available to the beef industry. It combines test results for several markers and genes for the same trait. The test is based, in part, on two genes involved in the post-mortem tenderization process: Calipastatin and Calgain. Calipain is an enzyme which weakers muscle fibers thereby making the fibers more tender. Calipastatin is an enzyme which inhibits the post-mortem tenderization process by inhibiting the effects of Calpain.

GeneSTAR GPD

T1	T2	T3	GPO Lbs. Shear Force
	tal tal	1	12
<u> </u>	*	++	-1,8 -1,8 -1,8
	0	1	-13 -13 67
	杰杰	4	-1.8 -1.4 -1.0
*	<u>*</u>	益益	-1.5 -1.3 -0.7
(A-1990)	0	2.4 4.	-1.1 -0.7 -0.3
	**	1 A	-1.5 -1.1 -0.7
.0.	盏	**	-1.3 -0.8 -0.3
	<u>*</u>	44	-0.8 -0.4 -0.0

 Values derived from Independent Validation Papulation

Results are reported as D2 STARS per marker for use in metag decisions along with the SPD which reflects the "true" value of the exempts unique combination of markers.

Lean Stilet

Quality Grade

GeneSTAR Quality Grade is a DNA genetic marker panel test offered by Pfizer Animal Genetics which identifies the presence of multiple major genes associated with quality grade. These markers provide beef producers with a quicker and more accurate test to determine an animals potential for improved quality grade. GenesTAR Quality Grade includes TGS, which was the first gene marker for a production trait in beef cattle. This panel of markers is the only marbling or quality grade test to have passed an independent validation by the National Beef Cattle Evaluation Consortium.

GeneSTAR GPD Quality Grade

Improvement in Percent

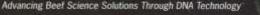
Others on		GCAbeller Terath USS		States are	
12.1.4	Schke	1226	%-Charter	5 F. S. W	N-Diau
2222	23.00	1222	24.87	4055	23.26
2225	29,00	122	DAIY.	4222	18.28
2229	23.86	12.17	16.57	4222	33.28
2212	28.66	101100	23.37	4212	19.57
2211	23.56	62.63	18.07	9233	13.08
2210	18.66	1210	13.37	8518	6.06
2224	25.46	1000	18,17	4381	12.88
2151	16.46	1200	13.77	4531	7.88
2224	13.46	1299	8.17	42.48	2.60
6122	12.40	F. K.R.E.	22.23	01.22	21.84
2333	37.42	VEE 0	11.11	4121	16.66
2000	22.42	1120	10.11	4122	11.86
2334	27.22	0.00	21.83	41.11	25.64
2111	23:22		1633	4421	11.64
2119	17.22	2004	11.93	2115	5.64
2102	22.02	1000	16.23	****	11.44
2100	12.02	2.4.4.4	11.23	2121	6.66
2224	12.02	1000	6.73		1.66
7077	20.86	1020	25.66	****	20.26
1221	25.56	1000	20.69	****	15.46
4434	30.16	****	15.89		15.45
XXXX	25.78		35.49	****	15.21
XAX	24.79		15.40		19.79
2000	15.78	1010	10.89		1.20
1011	35.58	1000	1529	0.002	16.66
ZHEK	75.50	1221	10.29	9885	3.00
	10.58	1211	3.29		2.26

+ ligiture derived from Independent Validation Population









Preserve Sample Identification Repeatable Genetic Analysis: Genetic Markers for Animal Quality

Genetic Markers for Boef Quality

& Management

& Management

DNA Traceability

Parentage



Recap of next generation of products



- Multigenic marker panels start to become the norm
- Number of traits and markers grow exponentially
- No longer any emphasis on which genes the markers are associated with, or how those genes function
- No independent validation of many tests
- Multiple different reporting systems that do not allow interchange (1-10, MGV, GPD, Number of Stars) or interpretation relative to EPDs
- DNA information still being presented separately from EPDs

Lead Today with 50K

- 1. Birth weight
- 2. Weaning weight
- 3. Weaning maternal (milk)
- 4. Calving ease direct
- 5. Calving ease maternal
- 6. Marbling
- 7. Backfat thickness
- 8. Ribeye area
- 9. Carcass weight
- 10. Tenderness
- 11. Postweaning average daily gain
- 12. Daily feed intake
- 13. Feed efficiency (net feed intake)





50K SNP chip assays 50,000 SNPs spread throughout genome



The American Angus Association® through its subsidiary, Angus Genetics Inc.* (AGI), has a vision to provide Angus breeders with the most advanced solutions to their genetic selection and

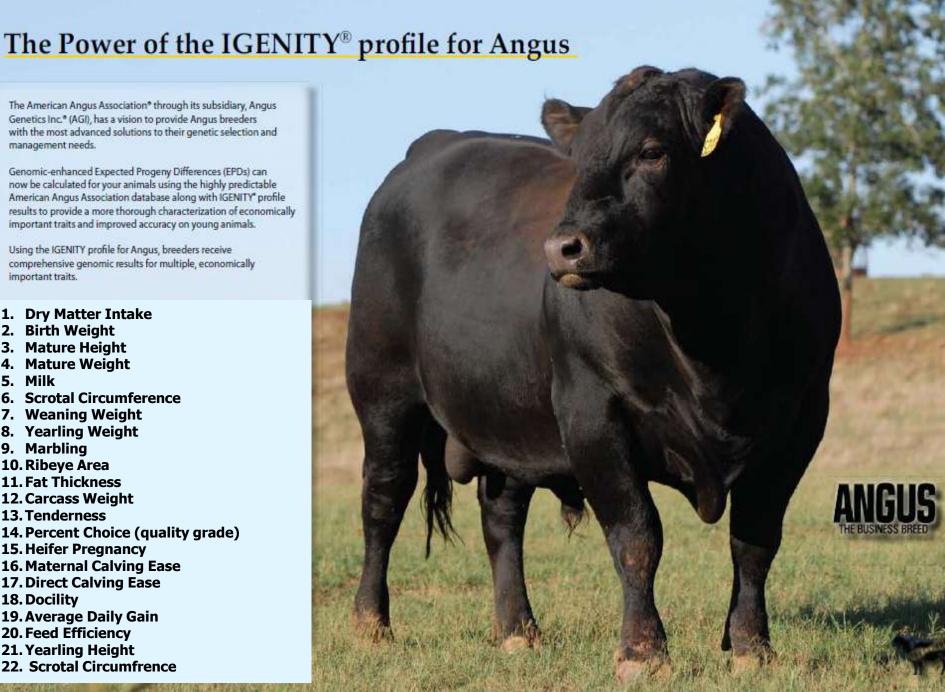
Genomic-enhanced Expected Progeny Differences (EPDs) can now be calculated for your animals using the highly predictable American Angus Association database along with IGENITY* profile results to provide a more thorough characterization of economically important traits and improved accuracy on young animals.

Using the IGENITY profile for Angus, breeders receive comprehensive genomic results for multiple, economically important traits.

- 1. Dry Matter Intake
- 2. Birth Weight

management needs.

- 3. Mature Height
- 4. Mature Weight
- 5. Milk
- 6. Scrotal Circumference
- 7. Weaning Weight
- 8. Yearling Weight
- 9. Marbling
- 10. Ribeye Area
- 11. Fat Thickness
- 12. Carcass Weight
- 13. Tenderness
- 14. Percent Choice (quality grade)
- 15. Heifer Pregnancy
- 16. Maternal Calving Ease
- 17. Direct Calving Ease
- 18. Docility
- 19. Average Daily Gain
- 20. Feed Efficiency
- 21. Yearling Height
- 22. Scrotal Circumfrence



Black Angus Sire

GAR Predestined



Reg. No.: 13395344 Calved: 8/16/1999 Tattoo: 5899 Semen: \$25 Certificates: \$20 Spring 2010 EPD

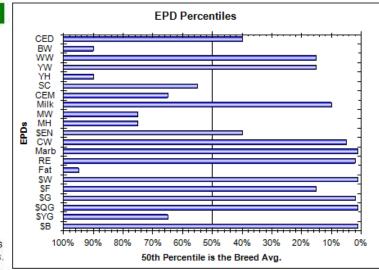
G A R Predestined:

From start to finish-conception to carcass-no other bull in the beef business today adds as much real value to cattle as Predestined. Ranking as the #1 bull for \$B in the breed-our customers tell us that their Predestined-sired cattle return the most dollars to their pockets--they know that \$B works. Unlike any other 036 son, Predestined tones down size, adds depth of flank, superior feet and legs and a pleasant disposition to his offspring. His conception rate is high and he's been a standout in timed-Al programs. His progeny look good-his bulls are thick and his heifers are fancy--and they always display additional shape and capacity. He ended 2006 as our top-seller and rightfully so--Predestined's many talents for creating value are for real.

	Production							Mate	rnal		
								MkH MkD			
+7 .84	+4.1 .97	+53 .96	+99 .94	+0 .96	+.31 .95	+6 .80	+28 .85	345 1135	+13 .81	+.2 .81	+5.24

Carcass				Usnd			\$Value	s			
CW Acc	Marb Acc	RE Acc	Fat Acc	Grp Prog	UGrp UProg	Wean	Feedlot	Grid	\$QG	\$YG	Beef
+26 .82	+1.07 .84	+.59 .82	+.046 .81	47 261	4269 11990	37.39	37.08	38.21	35.04	3.17	69.78

8	QG1	na	QG2	па	QG3	па	QG4	na	QG GPD	
lleS:	Т1		T2	0	Т3	0	100	je.	T GPD	-0.35
	FE1	na	FE2	na	FE3	па	FE4	na	FE GPD	e e



Current Sires Percent Breakdown

Registration #	Tendemess	Fal Thickness	Yield Grade	Ribeye Area	Carcass Weight	Percent Choice	Marbling
13395344	3	6	6	4	2	8	9

As of 03/22/2010

G A R Prede	G A R Predestined 13395344											13395344			
	CED	BW	WW	YW	ADG	DMI	NFI	CEM	MA	CW	FAT	REA	MS	TND	\$B/\$MVPFL
EPD	7	4.1	53	99	-	-	-	6	28	26	0.046	0.59	1.07	-	69.78
ACC	0.84	0.97	0.96	0.94	-	-	-	0.8	0.85	0.82	0.81	0.82	0.84	-	-
EPD % Rank	30	85	15	15	-		-	55	10	4	90	2	1	-	1
MVP	13	1.0	37	-	0.45	0.97	0.04	8	33	55	0.07	0.92	1.52	-0.43	243
MVP % Rank	3	70	10	-	30	90	90	4	1	1	90	1	1	80	1



American Angus Association performs weekly evaluations with genomic data



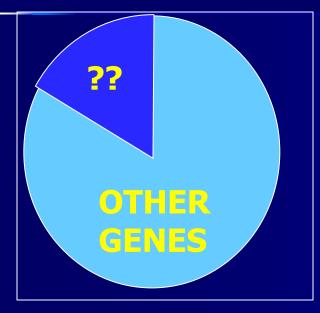
	Igenity	Pfizer
Calving ease (CED)	\checkmark	\checkmark
Growth (BW WW YW Milk)	\checkmark	\checkmark
Residual Average Daily Gain (RADG)	\checkmark	\checkmark
Docility (DOC)	\checkmark	
Carcass (CWT MARB RIB FAT)	✓	✓

http://www.angus.org/AGI/GenomicChoice070811.pdf



DNA TEST FOR MARBLING EXPLAINS SOME % (r²) OF GENETIC VARIATION





37%
GENETICS

63% ENVIRONMENT

EPD estimates all genes





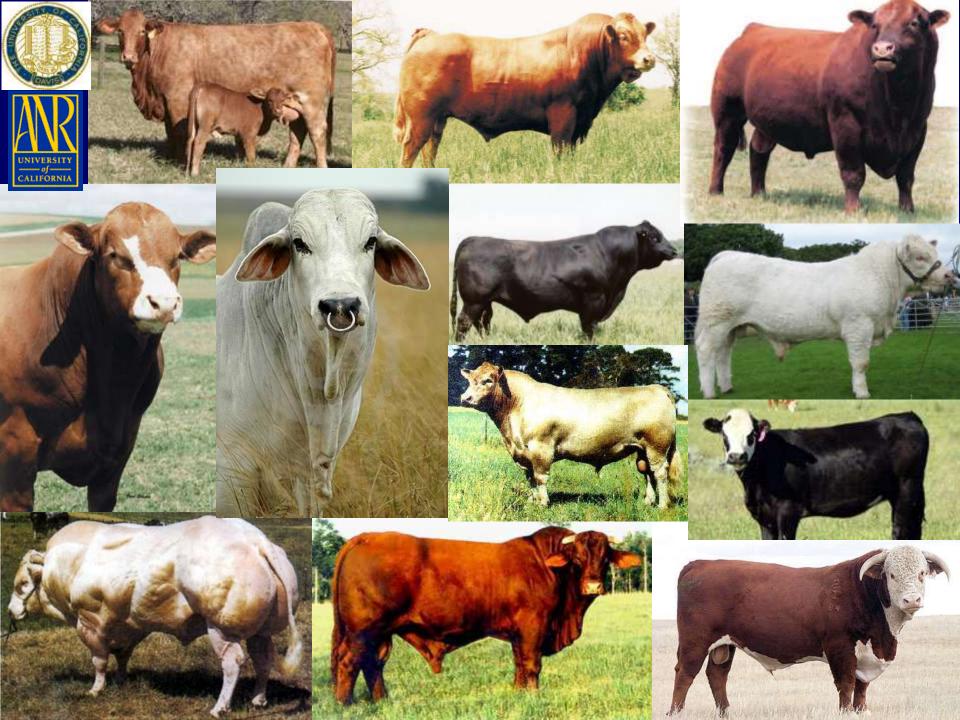


	AGI h ²	IGENITY	Y[®] Angus	Pfizer	HD 50K
Trait		Pro	ofile	for A	Ingus
		Included	% Genetic variation (r ²)	Included	% Genetic variation (r ²)
Average Daily Gain		X		X	30 ²
Net/residual Feed Intake				X	12 ²
Dry matter intake	0.31	X	20 ¹	X	42 ¹
Feed Efficiency		X		X	
Tenderness		X		X	26 ²
Calving Ease (Direct)	0.20	X	22 ¹	X	11^{1}
Birth weight	0.42	X	32 ¹	X	26 ¹
Weaning Weight	0.20	X	20 ¹	X	27 ¹
Yearling Weight	0.20	X	12 ¹	X	41 ¹
Yearling Height		X			
Calving ease (maternal)		X		X	40 ²
Milking Ability	0.14	X	6 ¹	X	10^{1}
Heifer Pregnancy		X			
Docility	0.37	X	22 ¹		
Mature Height		X			
Mature Weight		X			
Scrotal Circumference		X			
Carcass weight	0.31	X	29 ¹	X	23 ¹
Backfat thickness	0.26	X	25 ¹	X	31 ¹
Ribeye area	0.32	X	34 ¹	X	36 ¹
Marbling score	0.26	X	42 ¹	X	321
Percent choice		X			

^[1] Northcutt, S.L. (2011) Genomic Choices. American Angus Association®/Angus Angus Genetics Inc. release. http://www.angus.org/AGI/GenomicChoice070811.pdf (updated July 7, 2011)

[2] Pfizer Animal Genetics. 2010. Technical Summary. (posted April, 2010)

https://animalhealth.pfizer.com/sites/pahweb/US/EN/PublishingImages/Genetics%20Assets/HD50K/50K%20Te ch%20Summary%204-13-10.pdf.







Summary



- Selection requires accurate identification of genetically superior animals to become parents of next generation
- Accuracy of EBVs/EPDs can be improved by
 - Having performance records on individual and its relatives
 - Maintaining contemporary groups especially in feedlot
 - DNA tests that have been shown to work in Wagyu breed
- Need training populations of Wagyu cattle with records and genotypes to develop accurate DNA tests for Wagyu cattle
- If DNA tests could be developed that explain genetic variation in marbling in Wagyu cattle, there would be great potential to increase the accuracy of marbling score breeding values a trait of great financial importance to Wagyu breeders and considerable value would be generated by this improvement





There are a large number of genetic abnormalities in cattle occurring in a variety of breeds.

Images from an article by David S. Buchanan, NDSU http://www.ag.ndsu.edu/williamscountyextension/livestock/genetic-defects-in-cattle





Myostatin: mutations in cattle, mice and humans



Phenotypic comparisons of: "double muscled" bull homozygous for loss-of-function allele at the myostatin (MSTN) locus, mouse, and human.

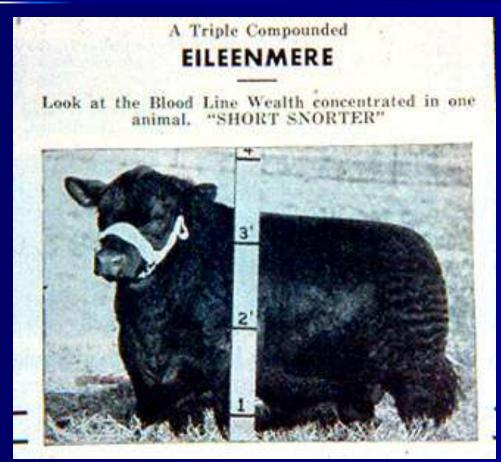


Womack J E Genome Res. 2005;15:1699-1705



Economic implications of recessive genetic factors





An early '50's advertisement that superimposed a measuring stick in the picture of this bull who was nick-named "Short Snorter."

Based upon his height and age, he was less than a frame score 1.

Image from https://www.msu.edu/~ritchieh/historical/shortsnorter.jpg

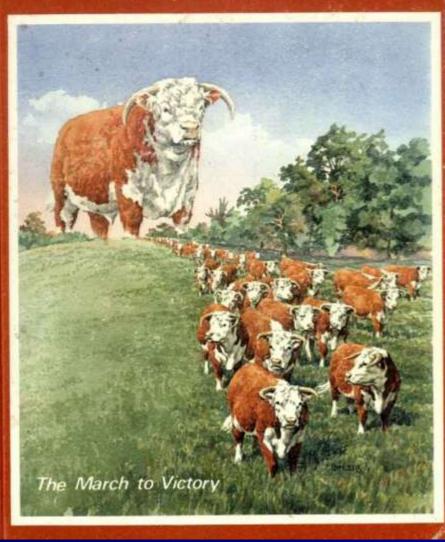






THE BATTLE OF BULL RUNTS

By L. P. McCANN



A 1956 survey of Hereford breeders in the USA identified 50,000 dwarf-producing animals in 47 states.

Through detailed pedigree analysis and test crosses, the American Hereford Association, in concert with breeders and scientists, virtually eliminated the problem from the breed. Because carrier status was difficult to prove and required expensive progeny testing, some entire breeding lines were eliminated.



Arthrogryposis multiplex; aka Curly Calf



- From a scientific standpoint, AM is the complete deletion of a segment of DNA that encompasses two different genes
- One of these genes is expressed at a crucial time in the development of nerve and muscle tissue. The mutation results in no protein being produced from this gene and therefore it is unable to carry out its normal function so homozygotes show phenotype



From September 8 – November 3, 2008 identified genetic problem, developed test, and released carrier status of 736 bulls!



- In the 11 months following the release of the test, the AAA posted the results of tests for AM on about 96,247 cattle.
- This amounts to \$2.4 million in testing costs
- Of these, 20% (19,529) were carriers of AM. That leaves 23,638 bulls and more than 53,000 heifers which tested as free of AM.
- At \$4K/bull and \$2K/heifer ~ \$200 million of suspect animals shown to be free of RGF allele

Based on calculations in Buchanan, D.S. 2009. Genetic Defects in Cattle. http://www.ag.ndsu.edu/williamscountyextension/livestock/genetic-defects-in-cattle



Estimates of US and Australia genetic testing costs (Angus)



	US	AUSTRALIA
AMF	113,526	12,021
NHF	77,067	9,936
CAF	28,837	2,532
TOTAL NUMBER	294,054	34,991
COST (@ \$25/test)	7,351,350	874,775

Numbers kindly shared by Bryce Schumann, American Angus Association; and Carel Teseling, Angus Australia





Angus Australia provides probability data on animal in database





GeneProb used to estimate the probability of every animal in the database being a carrier based on all ancestor and descendant DNA test results



Factor XI deficiency (F11) status (usually non-lethal)



Factor XI (F11) is a plasma protein that participates in the formation of blood clots. Factor XI deficiency is an autosomal disorder that is associated with mild bleeding in Wagyu. Affected animals show prolonged bleeding time and abnormal plasma coagulation after trauma or surgical procedures such as castration or dehorning.

PHOTOGRAPH – Affected F11 Female exhibiting no harmful symptoms





Erythrocyte Membrane Protein Band Deficiency (Spherocystosis) (Band3)



Cattle that are homozygous (have two copies of the recessive allele) have morbidly anemic (bleeding caused by the abnormal red blood cells). This is a disorder of the surface membrane of the erythrocyte (red blood cells). The alves are typically born weak and small (40-55 lbs birth weight) with labored breathing and are not able to stand or suckle at birth. Death normally occurs within the first 7 days after birth. Some cases live to adulthood but there is a severe retardation in growth.



PHOTOGRAPHS - Icterus in the eyes &, anaemic calf @ 5m (90 kg lwt)





Claudin 16 deficiency (CL16) (Type 1)



Claudin 16 or RTD(Renal tubular dysplasia) is a gene disorder on chromosome 1 and causes kidney failure (chronic interstitial nephritis (CIN), often with a build up of excess fibrous connective tissue in kidneys as well as other tissues). This disorder results in terminal kidney failure and the onset can occur any time from late adolescence. Affected cattle typically have short lives (unlikely to live more than 6 years).



PHOTOGRAPH - Elongated Hooves on an affected CL16





Chediak Higashi Syndrome (CHS)



CHS is a macrophage disorder (a white blood cell that has an important role in the immune response to disease). If cattle have a malfunctioning immune system, this makes them unable to resist bacterial challenge. Blood is slow to coagulate so often the first indicator is unusual umbilical cord hemorrhage at parturition (calving). Cattle with this syndrome often have an unusually pale coat color.

PHOTOGRAPH – Affected newborn calf with a pale coat







Factor 13 deficiency (F13) status



This gene contributes to fibrin stability which is an integral part of the blood coagulation pathway (blood clotting ability). Disorders in this gene cause severe hemorrhage (bleeding). In calves hemorrhage is particularly likely to occur in the hindquarters causing blood to pool and stagnate under the hide. In adult cattle any minor trauma (such as hitting the animal) can cause major hemorrhage at the trauma site.



PHOTOGRAPH Navel of an affected
F13 newborn

NOTE, this test is still in R&D as a positive control animal has not been identified in the US or Australia. We will continue to run this test and report the results to the American Wagyu Association (the "Association"), and if and when a Positive animal is identified by the test, staff from iGenix will work with the breeder and the Association to validate the results. When the validation is complete iGenix will begin reporting on this disorder to the member. If you have an animal that has symptoms as described above, please collect a blood sample (purple top blood tube) or nasal swab and contact the iGenix lab.









AgProDX Genetic Testing Services -**Current Offerings:**

Wagyu-breed inherited disorder testing

This test allows you to evaluate your animals for the following recessive genetic mutations that cause:



- 1. Band3
- 2. Chediak-Higashi Syndrom (CHS)
- 3, CL-16
- 4. F11-deficiency (mild bleeding disorder).
- 5. A test for F13-deficiency (severe bleeding disorder) is still in R&D, and results will not be reported until full validation is completed)

2011 Test prices:

Individual Tests	Price
Band3	\$75.00
CHS	\$75.00
CL16	\$85.00
F11	\$100.00

Bundled Tests	Price
Any 2 tests	\$120.00
Any 3 tests	\$135.00
All 4 tests	\$145.00

Please contact us at any time with your questions and comments.







Genetic Testing Status and Offspring Distribution Predictions

The genetic status of each tested animal will be reported as one of the four following results:

Free (F)

- Means the animal has <u>two</u> copies of the normal gene
- Also referred to as Normal and Unaffected

Carrier (C)

- Means the animal has <u>one</u> copy of the normal gene and <u>one</u> copy of the mutated gene
- · Also referred to as Positive

Affected (A)

 Means the animal has <u>two</u> copies of the mutated gene

No Result (NR)

 Means the DNA sample was good but did not yield a result under the applicable test protocol

The Science of Genetics Predicts the Following Results from Each Type of Mating:

	Offspring Distribution							
Mating	Free	Carrier	Affected					
Free x Free	100%							
Free x Carrier	50%	50%						
Carrier x Carrier	25%	50%	25%					
Free x Affected		100%						
Carrier x Affected		50%	50%					
Affected x Affected			100%					

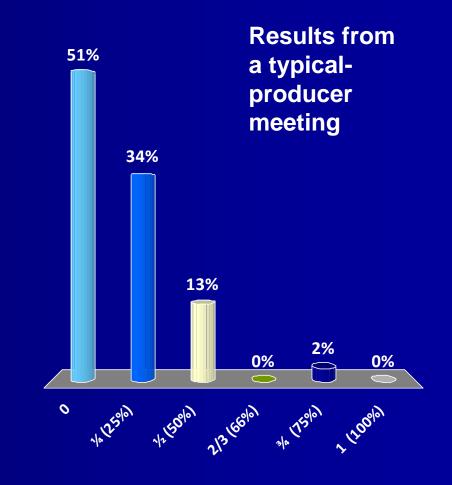




If you breed a curly calf carrier cow (AMC) to an curly calf free bull (AMF), what is the chance that the offspring will be stillborn as a result of being curly calf?



- 1. 0
- 2. 1/4 (25%)
- 3. 1/2 (50%)
- 4. 2/3 (66%)
- 5. 3/4 (75%)
- 6. 1 (100%)





Genetic implications of recessive genetic factors



"Carrier animals....their overall breeding value worth may outweigh the economic value of carrier status"

Chalier C. et al. (2008) Highly effective SNP-based association mapping and management of recessive defects in livestock. Nature Genetics 40:449-454

Should the market decide or should this choice be directed by industry (e.g. disallowing registration of known carriers)?







Important Registration Information and Testing Policy

Registration Information

Effective October 1, 2010, offspring from animals required to be tested in the section below, will not be eligible for registration until such time that the applicable testing has been completed and the results are received by the Association.

Required Testing Effective October 1, 2010

All fullblood, purebred, and percentage Al sires and embryo donor dams are required to be tested for all five genetic disorders. 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 five 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 five genetic disorders.

Annual Association Sales

Only cattle and genetics from cattle testing Free for all five genetic disorders are allowed to be sold through these sponsored events.

Publication of Test Results

All test results will be added to registration certificates and will be posted on the Association website. Once the Breedplan implementation is complete the information will be tracked in the database which is available to the public. Test results will be published in all public sale catalogs.

Fullblood, Purebred, and Percentage Cattle Not Tested

Effective January 1, 2011, any animal that traces to a known Carrier or Affected tested animal will be classified in Association records as a "Potential Carrier" until such time that the animal is tested and the status will be updated as applicable.



Some questions that remain about recessive genetic defects



- If all animals carry recessive genetic factors/defects how should "defects" be managed
- Can 50K or 800K data be also used to identify carrier status to spread the costs of DNA extraction. If so does it infringe on diagnostic test patents/IP
- Can we use genomics to better manage recessive genetic factors/defects
- Are appropriate decision support tools available for producers



Ideally cattle would be genotyped once early in life and genotypes shared among production sectors to derive the maximum value from the fixed DNA collection and extraction costs



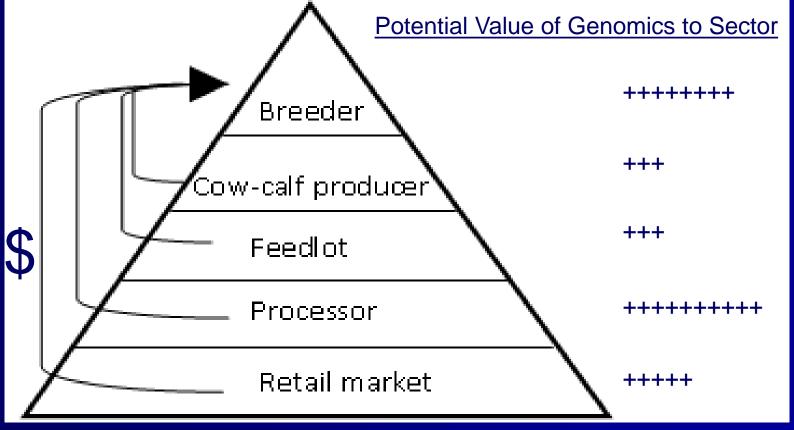


Figure from McEwan, J. C. 2007 Current status and future of genomic selection. Proceedings of the New Zealand Society of Animal Production 67: 147-152.





DNA tests are most valuable for traits that are



- low heritability (influence that genetics rather than environment has on a trait)
- are difficult or expensive to measure (feed efficiency)
- cannot be measured until after selection has occurred (carcass data)
- are currently not selected for due to lack of available phenotypic data (tenderness)