



Updates on USDA Bovine Respiratory Disease, Feed Efficiency and Heifer Fertility Projects



United States Department of Agriculture
National Institute of Food and Agriculture


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University of Missouri
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The Rumors Are True...




Got Neanderthal DNA?

An estimated **2.7%** of your DNA is from Neanderthals.

Jeremy Taylor (You)  **2.7%** 72nd percentile

Average 23andMe user  **2.5%**

Respiratory Disease



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Bovine Respiratory Disease

| Clinical signs | Cell Health Scoring Criteria | | |
|--|------------------------------|---|--|
| | 0 | 1 | 2 |
| Respiratory rate 100-120 bpm | 120-140 bpm | 140-160 bpm | >160 bpm |
| Cough | None | Intermittent single cough | Intermittent repeated coughs or occasional spontaneous coughs |
| Nasal discharge | None | Small amount of unilateral cloudy discharge | Excessive quantity of excessive mucous, mucopurulent discharge |
| Eye scores | Normal | Small amount of ocular discharge | Excessive amount of bilateral discharge |
| Ear scores | Normal | ESP tick or heat visible | Slight unilateral discoloration |

S. McGuirk's Diagnostic Criteria


Identify genomic regions associated with BRD resistance/susceptibility in beef and dairy cattle

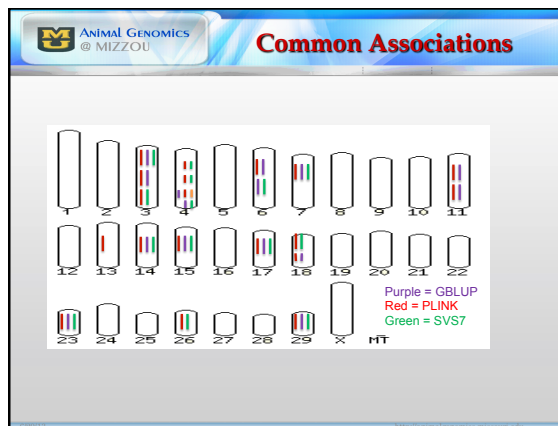
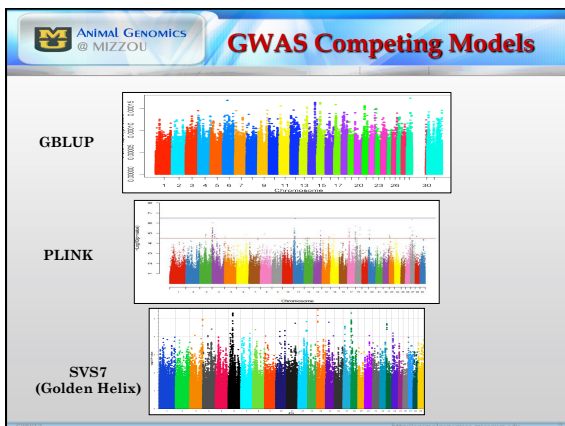
- Population A - 2000 Holstein calves
- Population B - 800 Holstein replacement heifers
- Population C - 2000 *Bos taurus* feedlot cattle
- Population D - 1000 *Bos taurus* purebred bulls



Population A Dairy Calves

- Samples on 2013 calves collected and clinical scores obtained
- Diagnostics for *Mycoplasma*, *P. Multocida*, *M. Haemolytica*, *H. Somni*, Bovine respiratory syncytial virus, Bovine viral diarrhea virus, IBR completed
- DNA extracted, and genotyped for 778,000 SNPs





- ### ANIMAL GENOMICS @ MIZZOU GBLUP Heritabilities
- BRD (21%)
 - Nasal discharge (9%)
 - Cough (19%)
 - Total clinical score (18%)
 - *Pasturella multocida* (2%)
 - *Mycoplasma* (2%)
 - BRSV (6%)
 - *Mannheimia haemolytica* (8%)
 - Temperature (10%)
 - Ocular discharge/ear tilt (8%)

- ### ANIMAL GENOMICS @ MIZZOU NM Replacement Holstein Heifers
- Samples on 800 heifers collected and clinical scores obtained
 - Diagnostics for *Mycoplasma*, *P. Multocida*, *M. Haemolytica*, *H. Somni*, bovine respiratory syncytial virus, bovine viral diarrhea virus, IBR completed
 - DNA extracted, and 778,000 SNPs genotyped
 - Heritabilities similar for BRD Case-Control
 - BUT... p_G between SNP effects in CA and NM = 0!
 - BRD is a mixture of diseases and pathogens differ between NM and CA
-

ANIMAL GENOMICS @ MIZZOU Identify Sires of these Calves

~70% of Holstein calves sire identified
All AI Holstein bulls are genotyped with the SNP50 or HD
We can use these genotypes to:

- Match bulls to calves
- Predict the MBVs of sires of these calves to BRD susceptibility

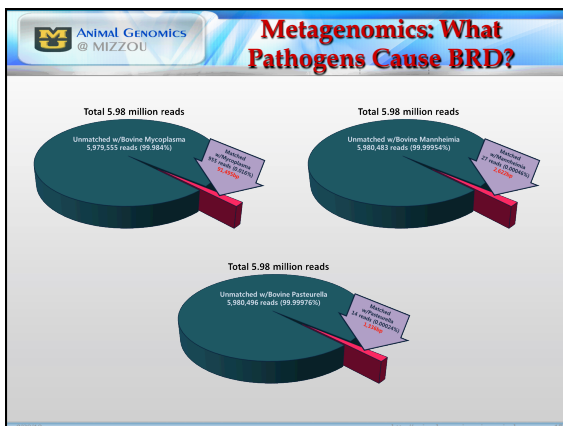
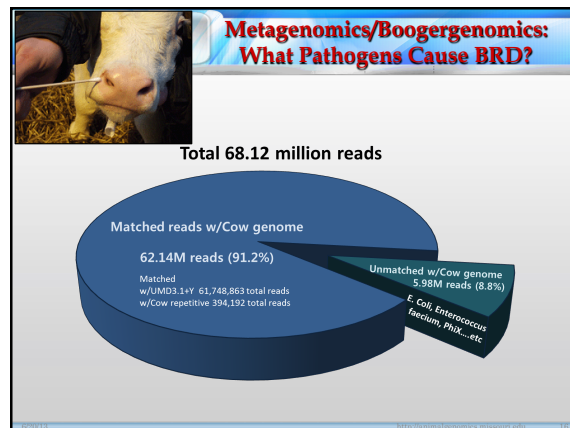
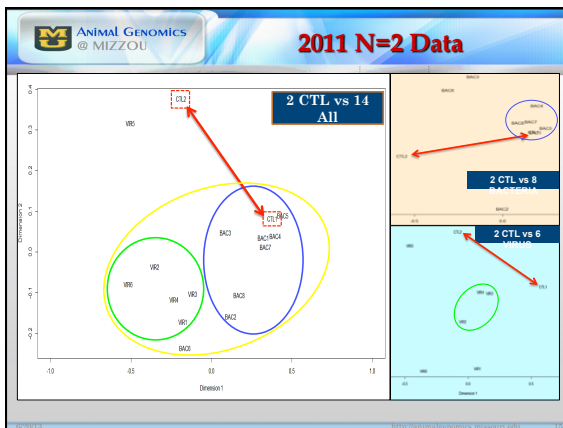
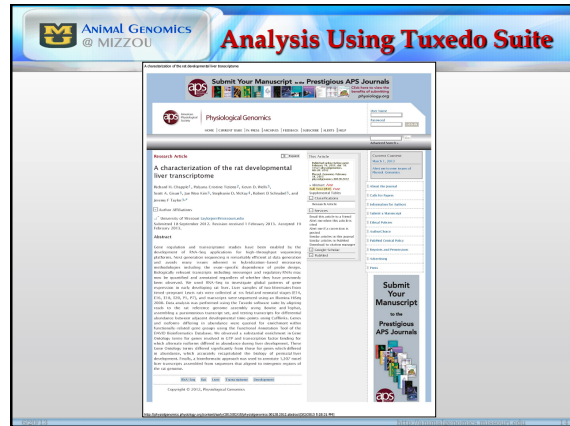
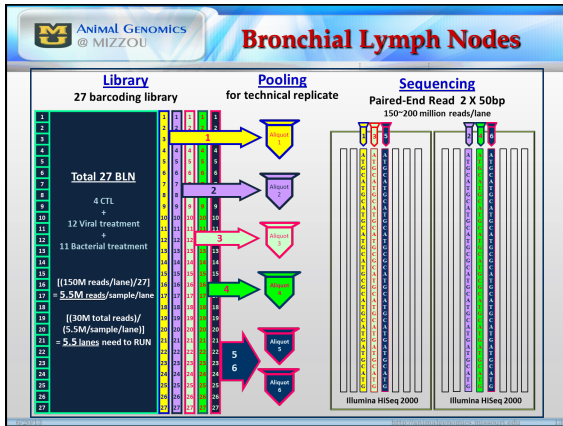
Mean MBV for BRD Risk = -2.2

BRD Unaffected Calves

Mean MBV for BRD Risk = +12.8

BRD Affected Calves

- ### ANIMAL GENOMICS @ MIZZOU Challenge Study
- 300 kg Angus calves challenged with *Mycoplasma bovis*, *P. Multocida*, *M. Haemolytica*, Bovine viral diarrhea virus, and Bovine respiratory syncytial virus (N=2 in 2011 and N=4 in 2012)
 - Identify genes expressed with given pathogens via RNA-seq of bronchia-alveolar lymph nodes
 - Characterize immunological responses
-
-
-



Genomic Selection

| Breed | SNP50 | HD | #Animals | #SNPs ¹ | 2013 Animals ² | Total Animals ² |
|------------------|--------------|--------------|--------------|--------------------|---------------------------|----------------------------|
| Angus | 1,093 | 510 | 1,603 | 747,473 | 435 | 2,038 |
| Charolais | 24 | 24 | | N/A | | 24 |
| Charolais×Ang | | | | | 450 | 450 |
| Commercial Xbred | | | | | 220 | 220 |
| Getzweh | | 369 | 369 | N/A | | 369 |
| Hereford | 361 | 491 | 852 | 684,458 | 300 | 1,152 |
| Limousin | | 37 | 37 | 568,501 | 45 | 82 |
| Normande | | | | | 3 | 3 |
| Pied×Ang×Simm | 236 | | 236 | N/A | | 236 |
| Red Angus | | 155 | 155 | 694,847 | 3 | 158 |
| Simm×Ang | 2,251 | 589 | 2,840 | 690,184 | 909 | 3,749 |
| Wagyu | | 35 | 35 | N/A | 150 | 185 |
| Total | 3,941 | 2,210 | 6,151 | | 2,515 | 8,666 |

¹Imputed using Beagle
²Does not include the possibility of a data swap with TEASUG, the Canadian Feed Efficiency Consortium or USMARC animals

- ### Four Data Sets (N=5,021)
- ◆ **Hereford Cattle fed at Olsens (HD)**
 - 847 animals in 10 contemporary groups
 - ◆ **F1² composites fed at USMARC (50K)**
 - 1,160 animals in 15 contemporary groups
 - ◆ **Legacy Simmental cattle fed at Illinois (HD)**
 - 1,444 animals in 202 contemporary groups
 - ◆ **Legacy Angus fed at Circle A (HD) and Angus fed at MU**
 - 1,580 animals in 102 contemporary groups

Dry Matter Intake

| Study | Genetic Var | Residual Var | Heritability |
|-----------|-------------|--------------|--------------|
| Hereford | 3.2 | 4.6 | 0.41 |
| USMARC | 1.9 | 3.4 | 0.35 |
| Simmental | 1.35 | 3.65 | 0.27 |
| Angus | 4.1 | 7.5 | 0.35 |

Mid-Test Metabolic Weight

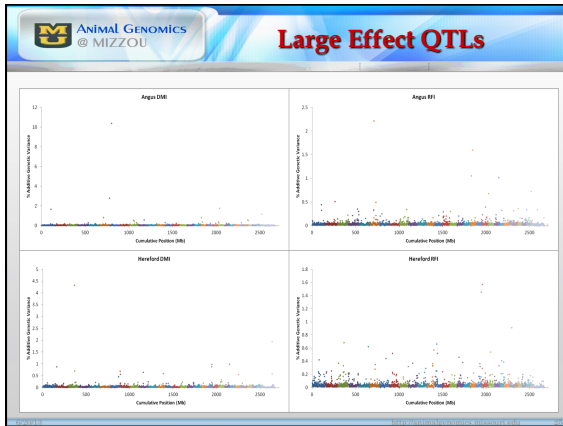
| Study | Genetic Var | Residual Var | Heritability |
|-----------|-------------|--------------|--------------|
| Hereford | 79 | 78 | 0.50 |
| USMARC | 84 | 97 | 0.47 |
| Simmental | 28 | 36 | 0.48 |
| Angus | 125 | 130 | 0.49 |

Average Daily Gain

| Study | Genetic Var | Residual Var | Heritability |
|-----------|-------------|--------------|--------------|
| Hereford | .09 | .23 | .27 |
| USMARC | .07 | .16 | .30 |
| Simmental | .04 | .13 | .23 |
| Angus | .06 | .24 | .19 |

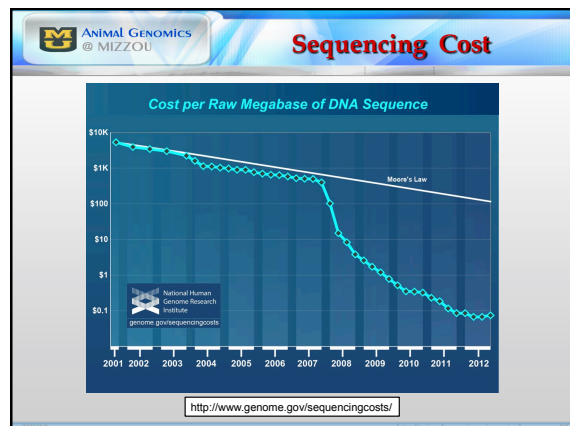
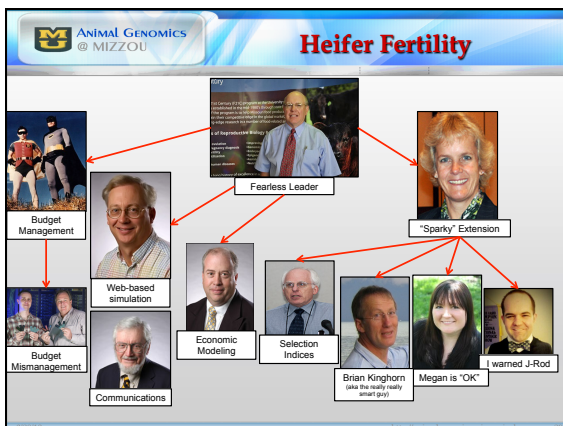
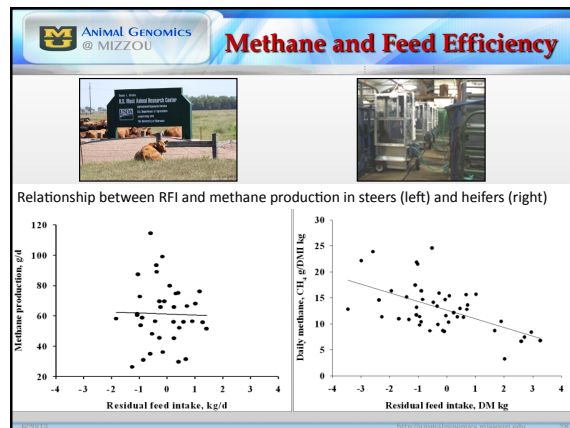
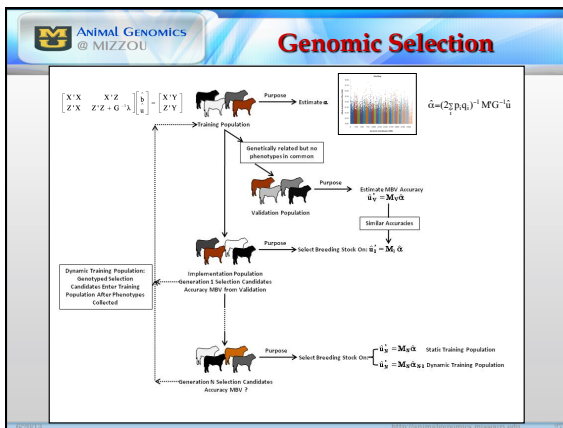
RFI

| Study | Genetic Var | Residual Var | Heritability |
|-----------|-------------|--------------|--------------|
| Hereford | 1.6 | 1.9 | 0.45 |
| USMARC | 0.91 | 0.94 | 0.49 |
| Simmental | 0.96 | 2.02 | 0.32 |
| Angus | 1.3 | 4.8 | 0.21 |



Large Effect QTLs

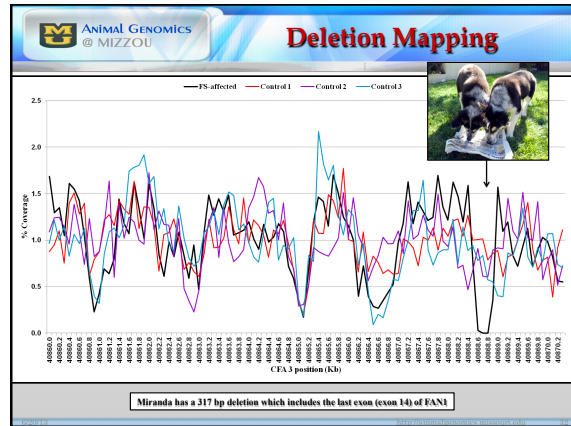
| | Dry Matter Intake | Mid-test Metab Wt | Gain on Test | ResidFeed Intake | | | | | |
|-----|-------------------|-------------------|--------------|------------------|------|-------|-----|------|-------|
| Chr | SNP | War | chr | Mo | SNP | War | chr | Mo | |
| ANN | 261 | 18.29 | 7.23 | 261 | 1.85 | 9.53 | 384 | 2.21 | 6.58 |
| ANN | 219 | 2.78 | 7.0 | 219 | 3.99 | 7.0 | 292 | 0.78 | 7.25 |
| ANN | 277 | 1.75 | 21.13 | 277 | 1.97 | 21.13 | 184 | 0.41 | 3.82 |
| ANN | 210 | 1.64 | 1.107 | 265 | 1.94 | 1.98 | 233 | 0.38 | 4.69 |
| ANN | 638 | 1.16 | 30.145 | 340 | 0.71 | 10.44 | 280 | 0.34 | 15.79 |
| ANN | 384 | 0.81 | 6.58 | 246 | 0.65 | 8.83 | 253 | 0.32 | 4.2 |
| ANN | 453 | 0.80 | 18.6 | 250 | 0.45 | 5.70 | 324 | 0.31 | 14.34 |
| ANN | 301 | 0.58 | 26.32 | 220 | 0.44 | 17.64 | 268 | 0.31 | 1.34 |
| ANN | 195 | 0.13 | 3.78 | 299 | 3.78 | 29.4 | 183 | 0.18 | 7.93 |
| HER | 214 | 1.95 | 31.115 | 183 | 2.55 | 7.93 | 312 | 3.13 | 5.186 |
| HER | 223 | 0.98 | 22.51 | 225 | 1.76 | 18.63 | 287 | 2.68 | 8.0 |
| HER | 316 | 0.97 | 19.57 | 195 | 1.25 | 3.70 | 182 | 1.26 | 11.34 |
| HER | 308 | 0.87 | 19.55 | 336 | 1.89 | 7.111 | 370 | 1.88 | 14.5 |
| HER | 212 | 0.87 | 2.2 | 247 | 0.78 | 17.25 | 213 | 0.98 | 8.2 |
| HER | 283 | 0.70 | 3.73 | 274 | 0.70 | 8.3 | 267 | 0.67 | 8.1 |
| HER | 287 | 0.69 | 8.0 | 211 | 0.66 | 8.2 | 394 | 0.46 | 16.74 |
| SIN | 382 | 0.86 | 15.52 | 192 | 3.14 | 14.24 | 295 | 1.23 | 2.11 |
| SIN | 345 | 0.78 | 18.33 | 288 | 2.58 | 28.6 | 197 | 0.82 | 4.58 |
| SIN | 388 | 0.69 | 28.33 | 321 | 1.72 | 19.77 | 211 | 0.79 | 7.92 |
| SIN | 297 | 0.61 | 15.55 | 189 | 1.71 | 21.71 | 285 | 0.54 | 4.25 |
| SIN | 348 | 0.51 | 17.41 | 218 | 1.48 | 18.58 | 288 | 0.58 | 16.38 |
| SIN | 196 | 0.45 | 2.49 | 288 | 0.67 | 19.59 | 243 | 0.47 | 14.29 |
| SIN | 282 | 0.44 | 18.48 | 193 | 0.66 | 14.25 | 223 | 0.39 | 15.83 |
| SIN | 278 | 0.39 | 14.46 | 344 | 0.64 | 3.115 | 312 | 0.37 | 6.94 |
| CPT | 23 | 0.99 | 8.52 | 21 | 3.49 | 6.38 | 25 | 2.42 | 20.8 |
| CPT | 22 | 0.94 | 13.48 | 25 | 2.88 | 8.29 | 21 | 1.43 | 6.38 |
| CPT | 13 | 0.76 | 2.44 | 16 | 1.41 | 11.24 | 25 | 1.27 | 12.54 |
| CPT | 21 | 0.74 | 10.14 | 13 | 1.23 | 6.35 | 23 | 0.98 | 2.46 |
| CPT | 13 | 0.57 | 6.35 | 23 | 0.84 | 7.27 | 18 | 0.67 | 11.33 |
| CPT | 15 | 0.51 | 27.4 | 26 | 0.82 | 12.45 | 32 | 0.58 | 14.10 |
| CPT | 25 | 0.41 | 24.38 | 25 | 0.81 | 20.4 | 20 | 0.57 | 2.14 |
| CPT | 28 | 0.35 | 13.41 | 8 | 0.72 | 12.71 | 19 | 0.55 | 13.68 |
| CPT | 25 | 0.35 | 3.85 | 24 | 0.70 | 1.42 | 18 | 0.55 | 7.93 |
| CPT | 25 | 0.35 | 3.85 | 24 | 0.70 | 1.42 | 18 | 0.55 | 7.93 |



Whole Genome Sequencing

You are a nobody in genomics until you sequence your own dog...

| Name | Breed | Disease | Coverage | Total Reads | Total Base Pairs | Chromosomal Region |
|------------|-----------------------------|--------------------------------|----------|----------------|-------------------|-----------------------|
| Jasper | Chinese Crested Dog | Multiple system degeneration | 23.4 | 498,404,908 | 56,447,845,446 | CFA1: 54.8-57.5 Mb |
| Oliver | Standard Poodle | Polymicrogyria | 15.2 | 336,244,627 | 36,569,611,910 | CFA4: 72.7-76.4 Mb |
| Satties | Irishdale | Cerebellar ataxia | 17.9 | 368,514,264 | 43,040,534,507 | CFA11: 66.9-70.0 Mb |
| Miranda | Basenji | Fanconi syndrome | 10.2 | 284,619,056 | 24,618,832,286 | CFA3: 44.3-50.1 Mb |
| Maggie | Soft Coated Wheaten Terrier | Paroxysmal dyskinesia | 23.1 | 559,144,094 | 55,619,462,414 | N.D. |
| Allystar | Henry Blue Terrier X Beagle | Cerebellar ataxia | 27.2 | 659,250,940 | 65,586,246,963 | N.D. |
| Clifford | English Cocker Spaniel | Degenerative myopathy | 21.0 | 514,950,236 | 50,735,392,704 | N.D. |
| Katie | Miniature Schnauzer | Neuronal ceroid lipofuscinosis | 8.3 | 207,671,762 | 20,045,383,134 | N.D. |
| Dandy | Labrador Retriever | Laryngeal paralysis | 12.5 | 304,021,990 | 30,104,892,536 | N.D. |
| Tennis | Chowchow | Dyskinetic epilepsy | 20.9 | 512,709,827 | 50,561,256,583 | N.D. |
| Woody | Pembroke Welsh Corgi | Degenerative myopathy | 30.2 | 732,686,470 | 72,788,166,647 | N.D. |
| Tango | Pembroke Welsh Corgi | Degenerative myopathy | 28.9 | 701,707,104 | 69,713,170,780 | N.D. |
| Mandy | Pointer | Cerebellar ataxia | 28.1 | 681,486,600 | 67,717,943,370 | CFA9: near centromere |
| Sabre | Jack Russell Terrier | Spinocerebellar ataxia | 34.3 | 831,348,994 | 82,695,065,197 | N.D. |
| Jack | Italian Greyhound | Heart murmur | 29.1 | 704,439,702 | 70,073,668,907 | N.D. |
| McGrickick | Scottish Bernard | Epilepsy | 28.4 | 693,114,974 | 68,573,872,841 | N.D. |
| River | Kerry Blue Terrier | Multiple System Degeneration | 28.4 | 687,612,564 | 68,398,334,405 | N.D. |
| Peanut | Portuguese Podengo | Leukodystrophy | 28.4 | 684,948,160 | 68,127,936,572 | N.D. |
| Toby | Border Collie | Sensory Neuropathy | 30.3 | 694,948,160 | 68,127,936,572 | N.D. |
| Jewels | Airedale Terrier | Cornell Dystrophy | 36.5 | 884,009,792 | 88,044,667,556 | N.D. |
| Totals | | | 482.3 | 11,547,745,239 | 1,157,494,196,358 | |



This is a Loss of Function Allele!!!

Sequence alignments for various breeds (Cani lupus familiaris, Canis lupus familiaris, etc.) showing deletions in the FANX1 gene region. Gel electrophoresis images show bands for Control 1, Control 2, and Control 3, and the affected allele.

Let's Sequence Some (Angus) Bulls!

| Reg | Name |
|----------|------------------------|
| 12309327 | GDAR SVF Traveler 234D |
| 12798179 | Circle A 200A Plus |
| 11418151 | B/R New Design 036 |
| 12223258 | J LB Exacto 416 |
| 10776479 | N Bar Emulation EXT |
| 11356596 | G A R Traveler 1489 |
| 11994601 | TC Stockman 365 |
| 12240991 | High Valley 4C6 Ambush |
| 12309326 | SVF Gdar 216LTD |
| 13395344 | G A R Predestined |
| 12655222 | Circle A 216LTD 6563 |

We have sequenced each of these bulls to an average depth of 30X and have generated whole genome polymorphism reports...

Data Analysis

- 2 libraries - 1 per HiSeq 2500 lane
 - 2 x 100 bp paired-end reads
 - 32X coverage
 - \$4,650
- Trim/Filter reads:
 - Illumina adapter sequences
 - Base quality scores
 - Remove reads containing repetitive elements
- Error correct using MSR-CA:
 - Removes most sequencing errors
- Align to UMDS1 using NextGene:
 - High stringency/paired-end alignment algorithm
 - Generate mutation report
 - Capture 40 pieces info including gene annotation
 - Upload to PostgreSQL relational database
- Process mutation report in db into 65 categories (increases monthly):
 - 2463 genes with LOF alleles! Some are assembly errors!

Numbers of Broken Genes

| No. LOF Mutations | No. Genes LOF Homozygotes | No. Genes LOF Heterozygotes | No. Candidate LOF Genes |
|--------------------|---------------------------|-----------------------------|-------------------------|
| 1 | 694 | 1722 | 1274 |
| 2 | 181 | 311 | 169 |
| 3 | 64 | 81 | 29 |
| 4 | 26 | 19 | 4 |
| ≥5 | 21 | 8 | |
| Total Genes | 986 | 2141 | 1477 |

Candidates for embryonic or early developmental lethals

ANIMAL GENOMICS @ MIZZOU **Comparison to Human**

| Description | Avg per bull | 1000 Genomes |
|--|--------------|--------------|
| Splice site +/- 2bp | 584.5 | |
| UTR | 25,489.3 | |
| Indels non-genic | 208,239.2 | |
| Indels genic | 91,742.7 | |
| Indels (inframe) that affect 1-2-3 amino acids (AA) | 160.2 | 190-210 |
| Indels that cause frameshifts | 524.5 | 300-350 |
| Stop codon usage | 401.5 | |
| High quality SNP synonymous AA | 11,741.8 | 10-12,000 |
| High quality SNP nonsynonymous AA | 11,729.2 | 10-11,000 |
| High quality SNP genic region | 1,422,460.6 | |
| High quality SNP | 3,524,459.0 | |
| High quality homozygous SNP (differing in Dominette) | 2,406,798.5 | |
| High quality heterozygous SNP | 5,502,478.4 | |

Broken Genes!!!!

Mutant!

ANIMAL GENOMICS @ MIZZOU

They can't all be lethal.... Can they??

- On average a bull would carry **134 lethals!**
 - We suspect that many are not real – assembly errors!
 - Our analysis pipeline is continually evolving
 - Many may not be essential for life!
 - We know from human studies that every human carries about 7 lethals
 - Cows are going to be very similar
 - We just didn't sequence enough bulls to find homozygotes that would eliminate these genes from consideration as being essential for life
 - There still must be a LOT of broken genes floating around in the Angus population and they really must have negative effects on SOME traits!!
 - Of the 1,477 genes with predicted loss of function alleles, 176 (16.7%) are identified in the mouse genome informatics database data base as possessing a lethal phenotype (2,838 mouse genes have mouse phenotype terms which include "lethal") in knockout homozygotes

ANIMAL GENOMICS @ MIZZOU **Objectives & Updates**

- Sequence the genomes of additional 89 registered Angus bulls**
 - To find as many LOF alleles as possible and remove from consideration those that are found as homozygotes in any bull (can't be essential for life)
 - Sequencing costs have decreased from \$5,500 to \$4,650
 - American Angus Association \$50,500
 - American Hereford Association \$25,000
 - Beefmaster Breeders United \$23,250
 - American Gelbvieh \$23,250
 - American International Charolais, American Simmental Association, American Maine-Anjou Association, American Wagyu Association?
 - We will sequence at least 150 bulls from 6-8 breeds
- Exchange sequence data**
 - USDA MARC, Genome Canada and INRA projects

ANIMAL GENOMICS @ MIZZOU **Objectives & Updates**

- Develop a genotyping assay with: 1) All LOF alleles, 2) 10,000 SNPs from BovineSNP50, and 3) As many other functional variants as we can fit**
 - AFFYMETRIX will build custom assay with 53,000 SNPs for ~\$46
 - Need 10,000 from the BovineSNP50 to impute genotypes up to 50K to allow use of developed molecular EPD (MEPD) equations
 - Likely to find 5-6,000 LOF alleles in all breeds of cattle
 - Remainder will be variants that are likely to have functional effect – i.e., change the amino acid composition of proteins
- Genotype 10,000 heifers**
 - Missouri Show-Me-Select™ Replacement Heifer Program
 - Recruited from multiple breeds (primarily Angus)
 - Any LOF allele that never turns up in homozygous form is probably lethal
 - Develop MEPDs for fertility in males and females (via embryonic loss)

ANIMAL GENOMICS @ MIZZOU **Objectives & Updates**

- Develop economic selection indexes that support multi-trait selection, inclusive of fertility**
- Develop decision support software to optimize breeding schemes via the implementation of selection indexes and mate selection based upon sire and dam recessive lethal genotypes**
 - We will have MEPDs for about 18 traits including Feed Intake, Shear Force, Fertility, Carcass, Growth, etc (From previously funded grants)
 - We will also have genotypes on heifers and bulls for lethals
- Develop:**
 - Web-based educational training program for extension livestock specialists, allied industries, producers, veterinarians, and students to enable successful adoption of genomic technologies for beef cattle breeding decision-making
 - Develop simulation exercise that demonstrates effects of MEPDs for heifer and sire fertility on reproductive performance and profitability
 - Develop innovative educational and outreach materials for the enhancement of the Beef Cattle Community of Practice

ADDITIVE GENETICS
This is how it works

16/ MotivatedPhotos.com

