Identification and Management of Alleles Impairing Heifer Fertility While Optimizing Genetic Gain in Cattle

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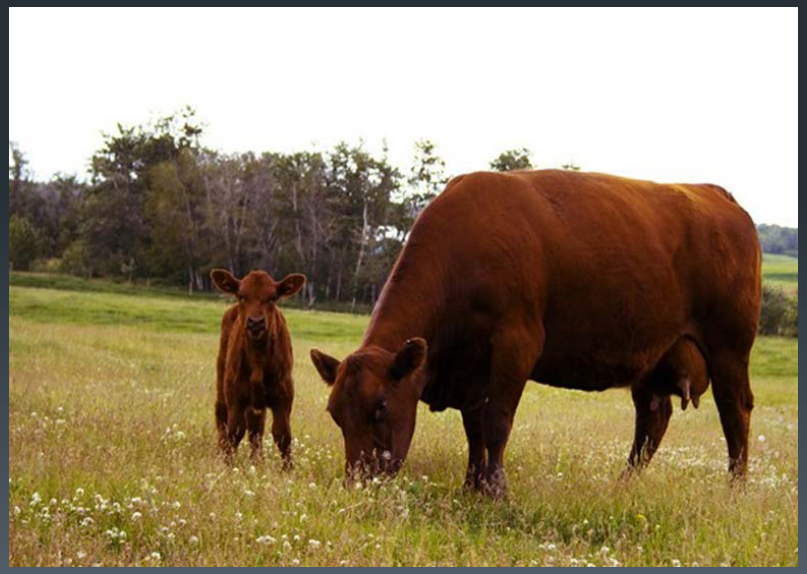
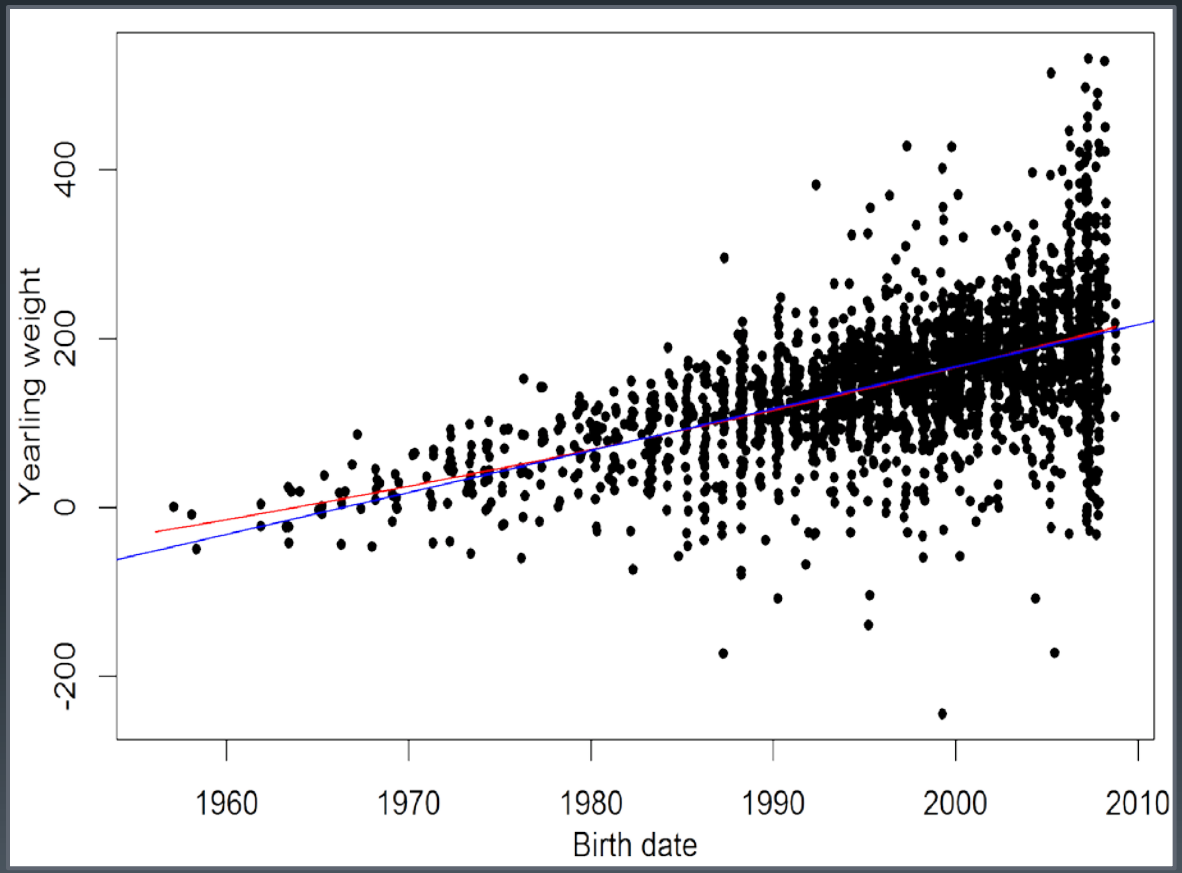


Project Goals and Background

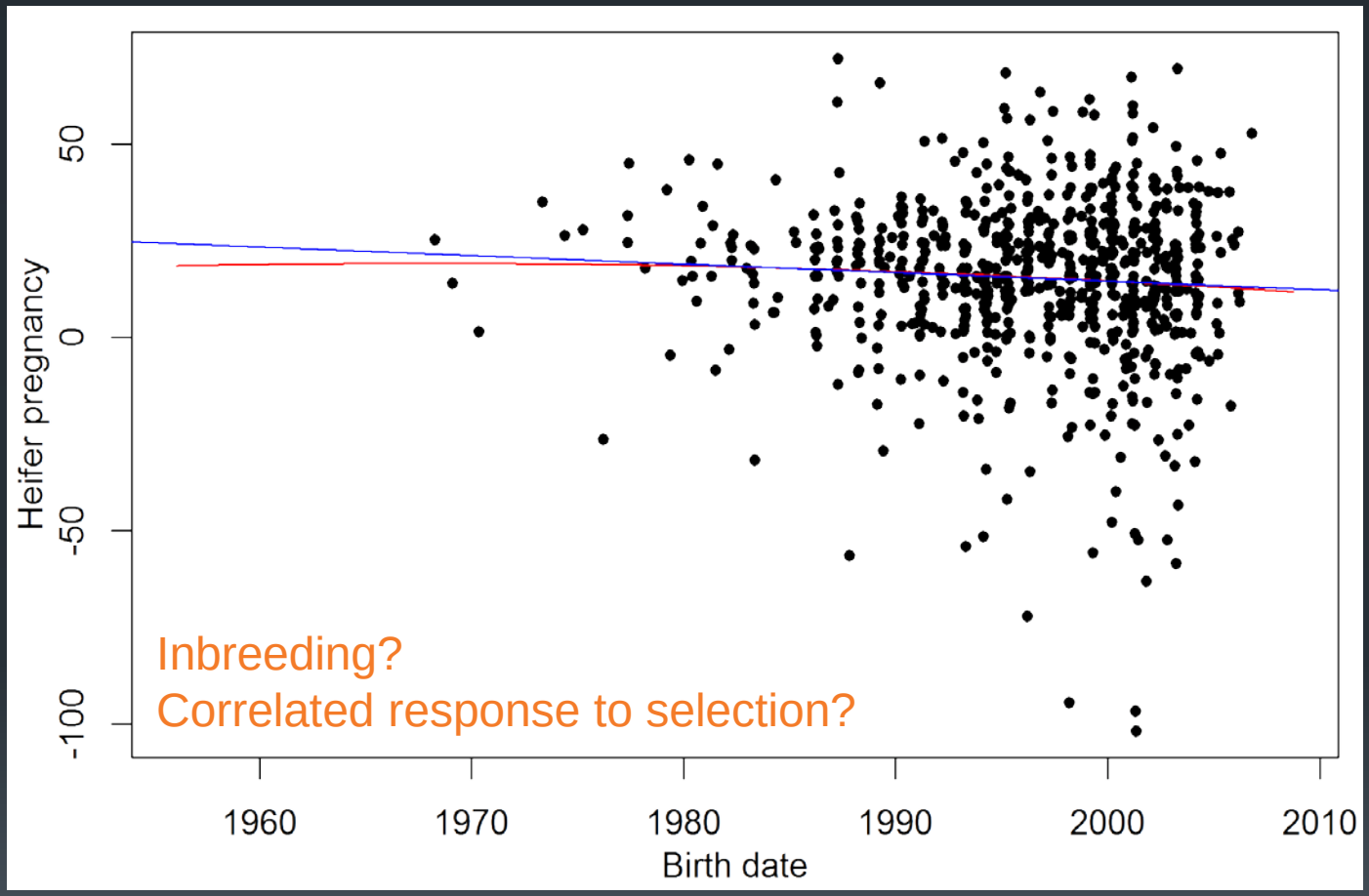
- Improve reproductive rate in US beef cattle
  - Does not sacrifice performance in other ERT
  - Improves overall profitability of the cowherd
- No secret that reproduction is a very important trait in the cowherd
- Maximize number of females that conceive early in the breeding season
- Maintenance of pregnancies that are achieved

Genetic Change



Deregressed EBVs for Yearling Weight of 2,755 registered Angus bulls demonstrates that breeders have achieved an average increase of 4.96 lb per year (blue line) over a 50 year period.

Genetic Change for Heifer Pregnancy



Deregressed EBVs for Heifer Pregnancy Rate for 698 registered Angus bulls indicates that Angus female fertility has decreased by 0.22% per year for about the last 25 years.

Effects of Inbreeding Accumulation

- Increases probability of alleles being homozygous
  - As with all lowly heritable traits (like fertility), reduces fitness

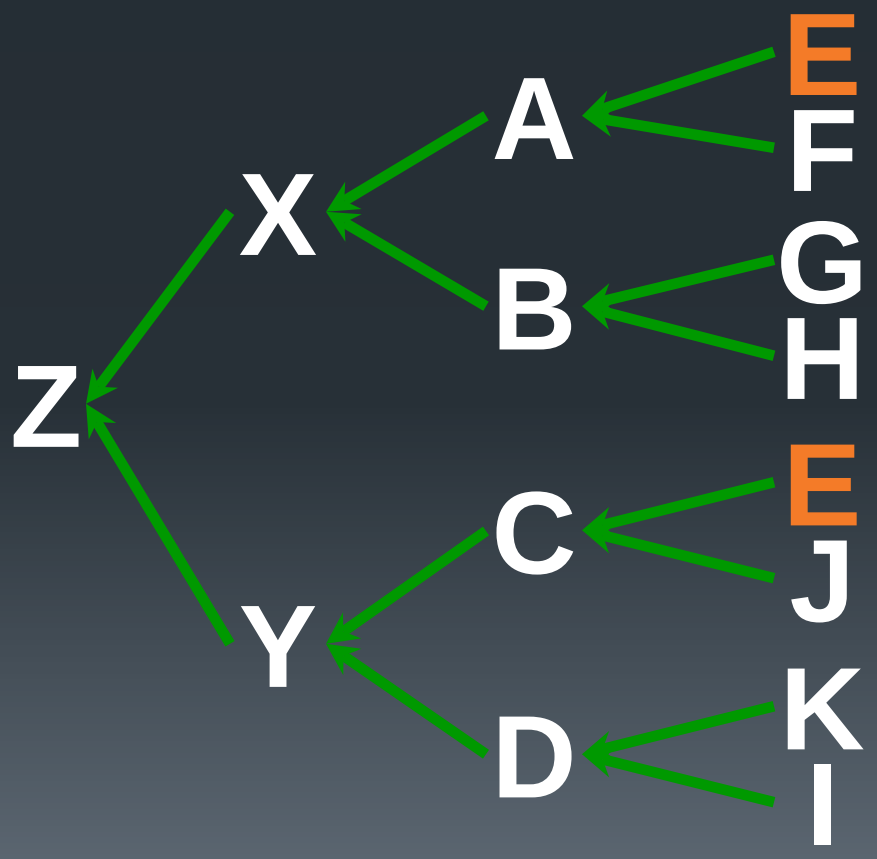
Table 1. Summary of heritability and level of heterosis by trait type.[a]

Trait	Heritability	Level of Heterosis	Inbreeding Depression
Carcass/end product	High	Low (0 to 5%)	Low
Skeletal measurements			
Mature weight			
Growth rate	Medium	Medium (5 to 10%)	Medium
Birth weight			
Weaning weight			
Yearling weight			
Milk production			
Maternal ability	Low	High (10 to 30%)	High
Reproduction			
Health			
Cow longevity			
Overall cow productivity			

[a] Adapted from Kress and MacNeil, 1999.

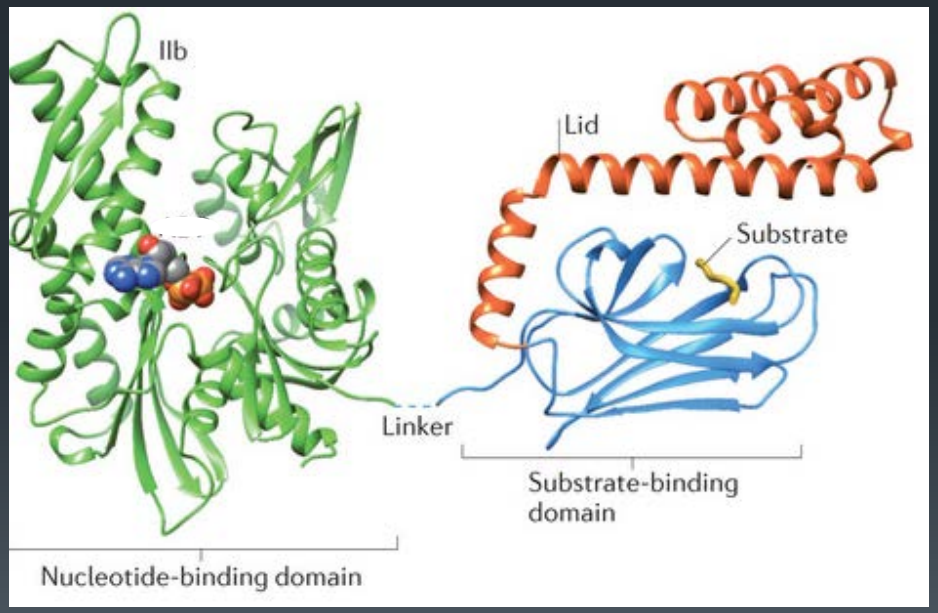
Effects of Inbreeding Accumulation

- Increases probability of alleles being homozygous
  - As with all lowly heritable traits, reduces fitness
  - Increases odds of alleles being identical by descent

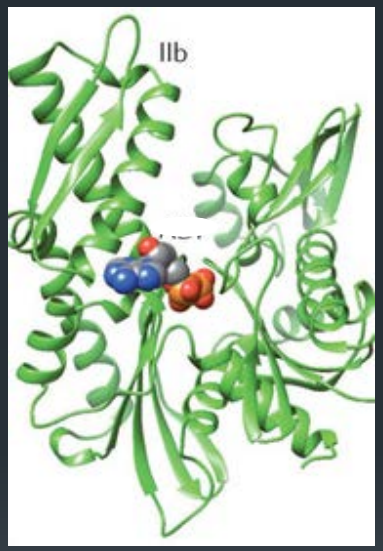


Effects of Inbreeding Accumulation

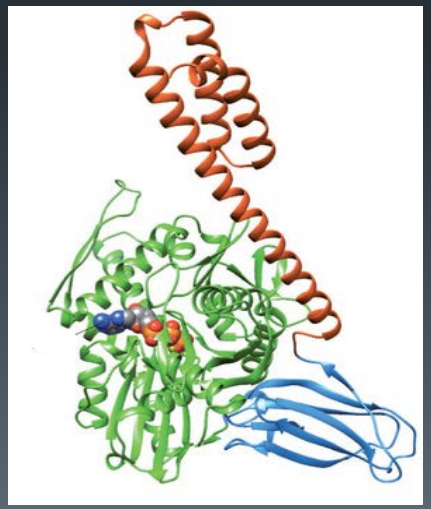
- Increases probability of alleles being homozygous
  - As with all lowly heritable traits, reduces fitness
  - Increases odds of alleles being identical by descent
    - Increases the odds of getting two copies of a broken gene



Truncated



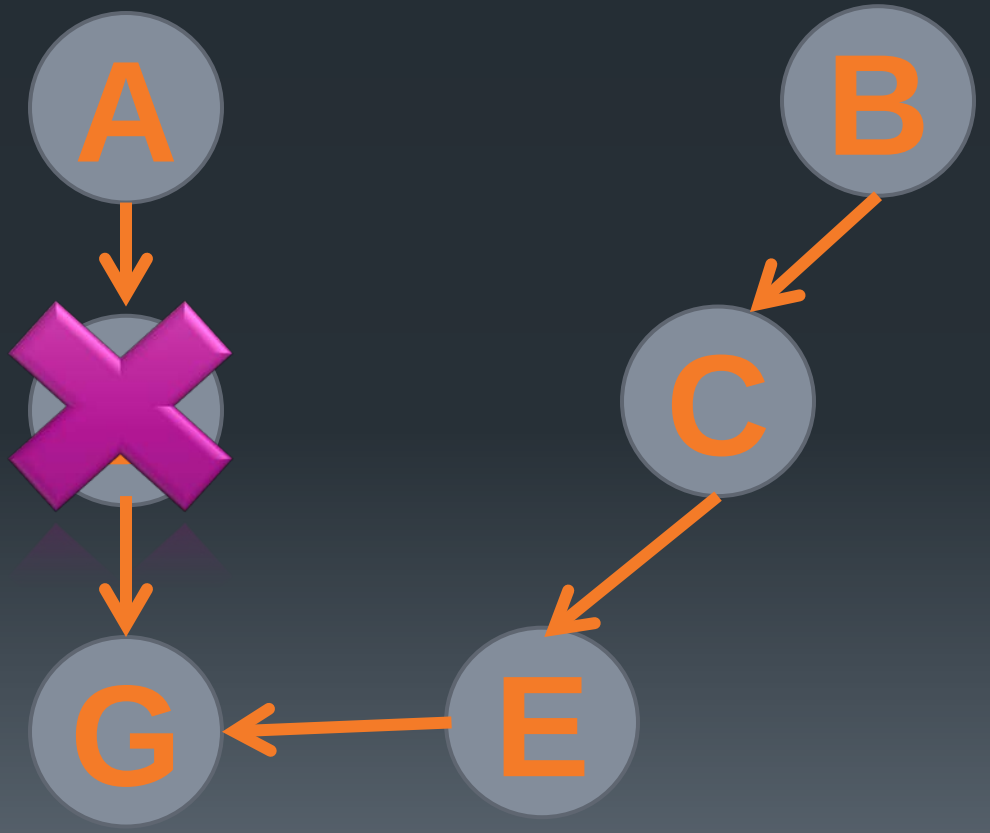
Improper Folding



When a broken gene is encountered…

- An organism has 2 options if that gene is vital to life:
  - Find a way to compensate

Lots of redundancy in biological systems helps to get around this problem



Essential for Life

- Die

Think About Toast as a Gene Product

Assume recessive mutations

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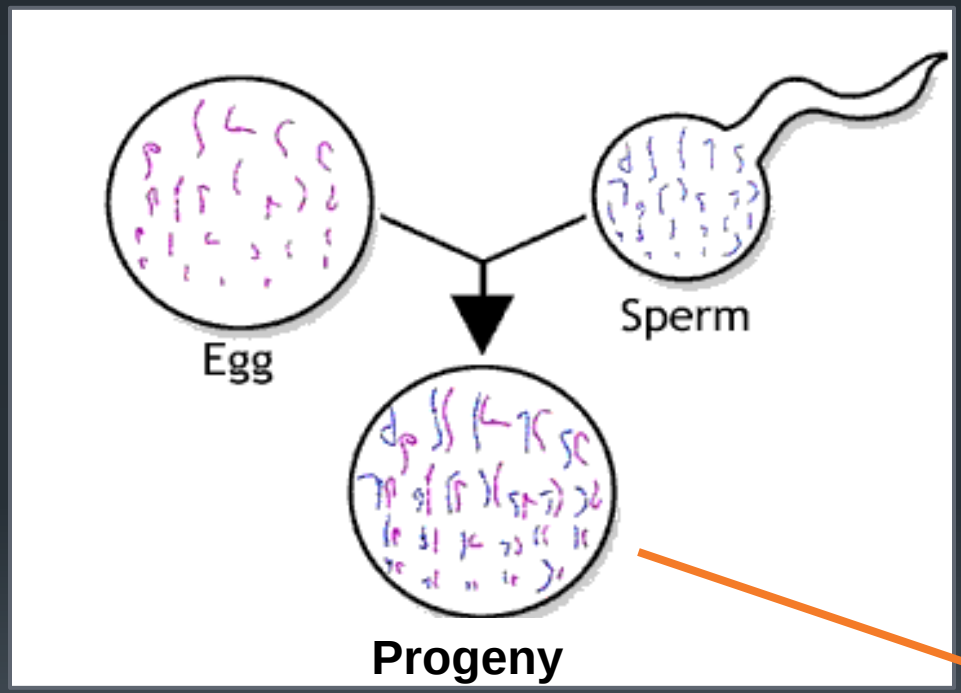
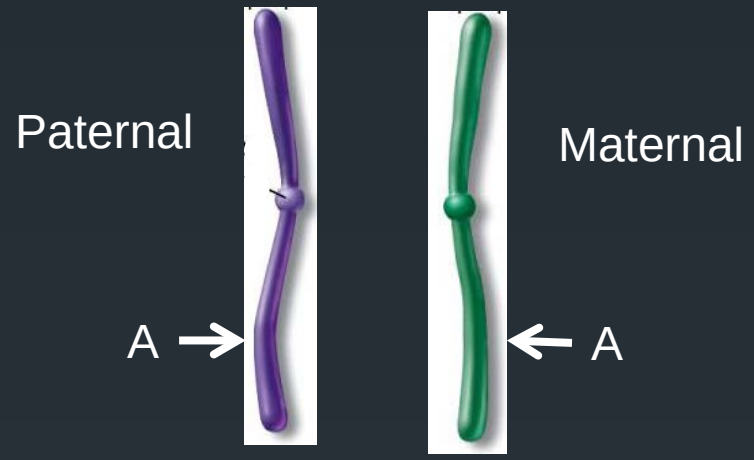
2 Normal Alleles

On the genome level:



On the Allelic level:



Think of the toaster like an animal-it produces proteins that serve a function-one from each of it's chromosomes

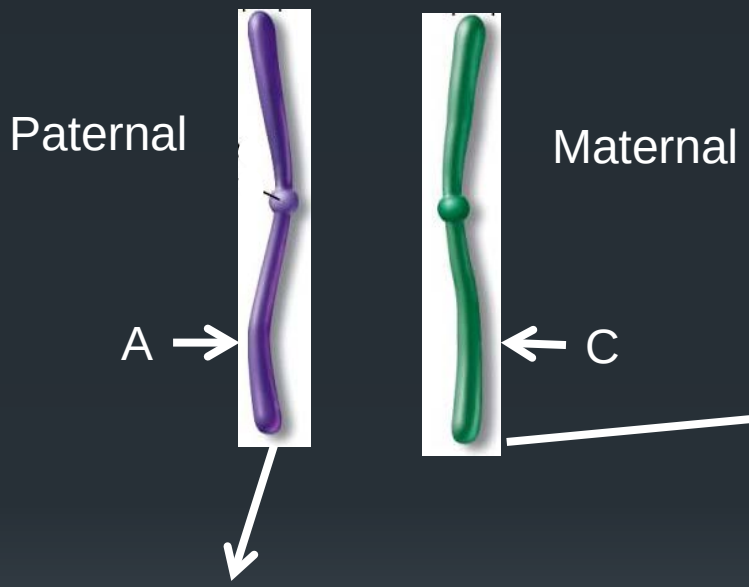
Broken Genes

- Called Loss of Function Mutations (LOF)
- Can be one of two forms

- Not Critical for Life
  - Will see all genotypes in the population (AA, AB, and BB)
  - Animals may have reduced performance or other deleterious effects, but are functional organisms

1 Normal, 1 Nonlethal Mutation

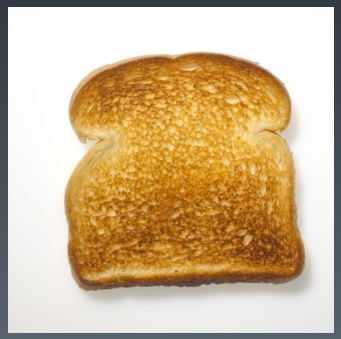
Assume A is the best possible allele at this locus

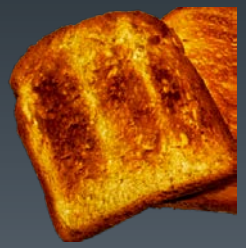


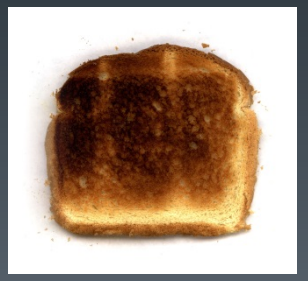
We're still making the gene product!

?

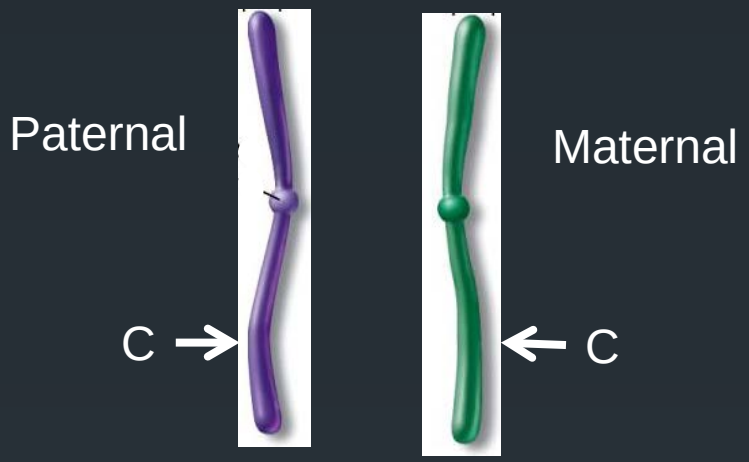
Animal doesn't perform as well, might be perfectly normal







2 Nonlethal Mutations



The animal can still produce the gene product from alt. pathway

?

If it's not necessary for survival, the animal probably doesn't perform as well, but it can live

Assume A is the best possible allele at this locus

Broken Genes

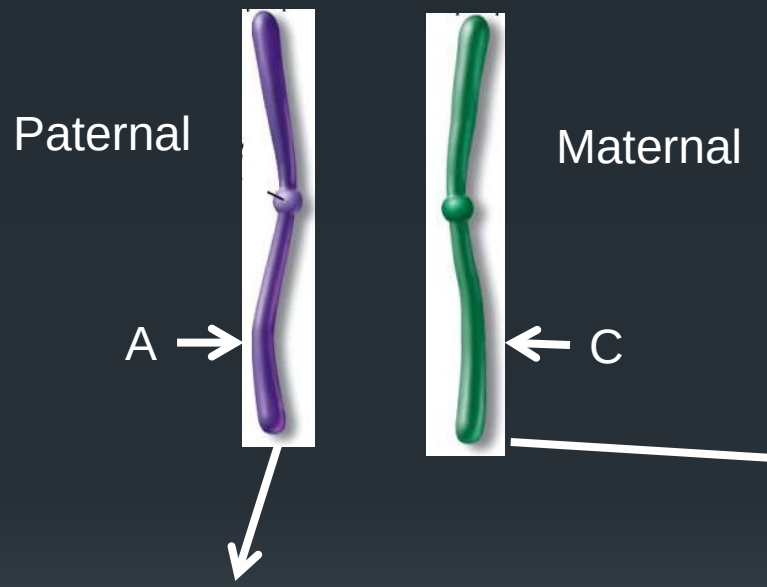
- Called Loss of Function Mutations (LOF)
- Can be one of two forms

- Not Critical for Life
  - Will see all genotypes in the population (AA, AB, and BB)
  - Animals may have reduced performance or other deleterious effects, but are functional organisms

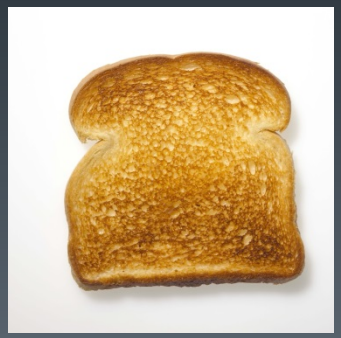
- Critical for Life
  - Animals cannot survive without at least one fully functional version of these genes

1 Normal, 1 Lethal Mutation

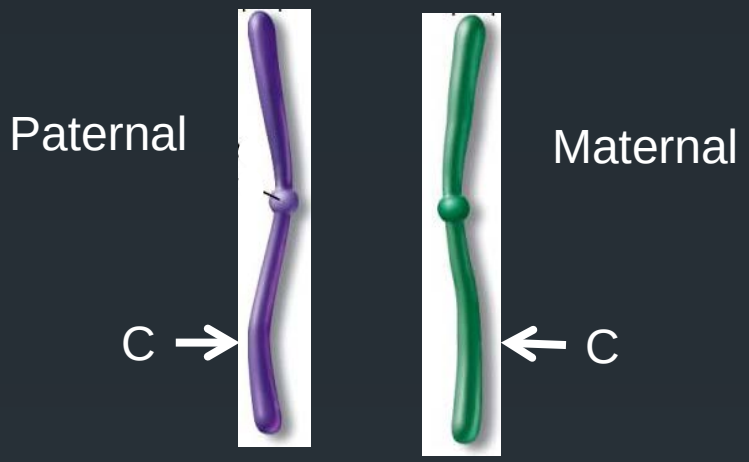
Assume A is the best possible allele at this locus



We're still making the gene product!



2 Lethal Mutations



There is NO functional gene product!

-In practice, these are never observed in the population

?

Animal can not survive

Assume A is the best possible allele at this locus

The Math

$p = frequency\ of\ normal\ allele\ (0.95)$
$q = frequency\ of\ broken\ allele\ (0.05)$

$p^2 = frequency\ of$ homozygous normal
$2pq = frequency\ of\ heterozygotes$
$q^2 = frequency\ of$ homozygous broken

Genotype 10,000 animals-
Expect to see:
9,025 homozygous normal
950 heterozygotes
25 homozygous broken

Genotype 10,000 animals for a lethal-
You would see:
9048 homozygous normal
952 heterozygotes
0 homozygous broken

Need lots of animals to test this!

The Case of the missing homozygotes!

Broken Genes

- Called Loss of Function Mutations (LOF)
- Can be one of two forms

- Not Critical for Life
  - Will see all genotypes in the population (AA, AB, and BB)
  - Animals may have reduced performance or other deleterious effects, but are functional organisms

- Critical for Life
  - Animals cannot survive without at least one fully functional version of these genes (inherits 2 broken genes)
  - When a mutation is lethal
    - Should not see all genotypes in the population (AA, AB)
      - “Missing homozygotes” when sampling within living populations
    - Can observe aborted fetuses, stillborn calves, or animals that die shortly after birth
    - Embryonic and early developmental lethals
      - Observed as poor pregnancy rates from aborted embryos or not observed at all
      - Easy to miss!

Not All Bad News!

- DNA tests can be developed in a couple months rather than several years
  - Can dramatically decrease frequency of these alleles in the population
- We can manage it if we know about it

- There are too many of these broken genes, that will likely be different between individual animals and between breeds, so we need to focus on managing around them rather than getting rid of them entirely
  - New ones will always come along, so it's a continual process
  - No reason to get rid of excellent animals
- Selection and breeding doesn't CAUSE these mutations
  - Just lets us see them by using certain animals widely within a breed

How Do We Find Them?

- A total of ~150 bulls from 9 breeds will be sequenced
  - Angus
  - Beefmaster
  - Charolais
  - Gelbvieh
  - Hereford
  - Limousin
  - Maine Anjou
  - Shorthorn
  - Simmental
- Coverage can vary
  - i.e. 4x vs 30x

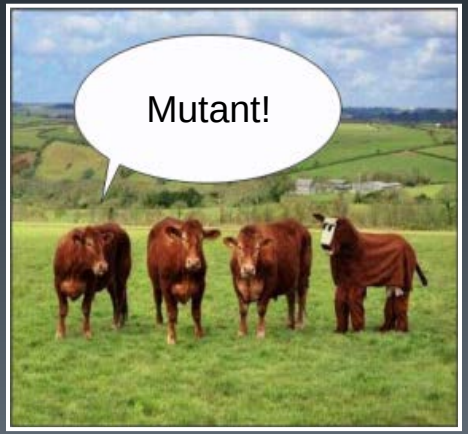
Breed	No. Animals	Av. Raw Coverage
Angus	99	26.86
Hereford	18	28.47
Limousin	12	10.27
Charolais	11	25.15
Simmental	11	27.76
Gelbvieh	8	27.31
Maine Anjou	5	27.85
Romagnola	4	7.73
Shorthorn	2	24.8
Red Angus	14	10.88
Holstein	25	4.42
Jersey	9	5.33
Bison	3	37.07
N'Dama	1	25.34
Brahman	11	5.26
Nelore	8	7.14
Gir	6	9.05
Beefmaster	10	28.65
Dog	120	22.79
Total	377	19.06

How Many Mutations?

Variants	Total	dbSNP 140	≥1 Hom	dbSNP 140	0 Hom	dbSNP 140	Essential	dbSNP 140
Frameshift	3,801	1,603	1,877	1,149	1,924	454	99	24
Premature Stop	1,964	864	1,204	571	760	293	50	24
Stop to Readthrough	419	218	340	187	79	31	4	1
Splice	4,663	2,189	2,605	1,383	2,058	806	194	110
Nonsynonymous AA	126,367	60,114	93,993	47,375	32,374	12,739	2,102	822
Total LOF	10,847	4,874	6,026	3,290	4,821	1,584	347	159
Total LOF + NS	137,214	64,988	100,019	50,665	37,195	14,323	2,449	981

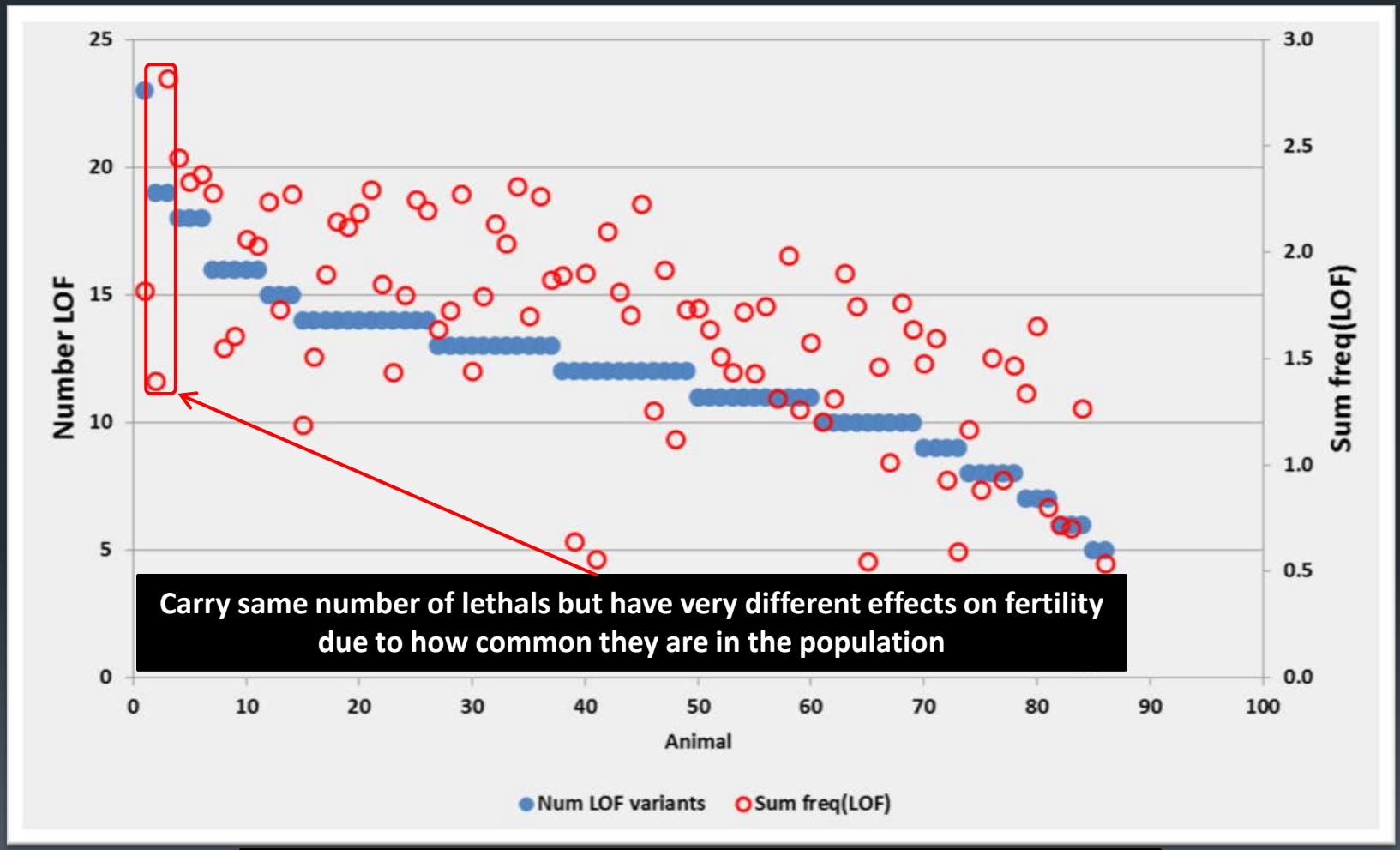
These are probably all lethals

Some of these are going to be lethals



197 sequenced animals
At least two animals with variant (eliminate errors)
Essential = Gene essential for life in mouse

LOF Essential Genes in Angus



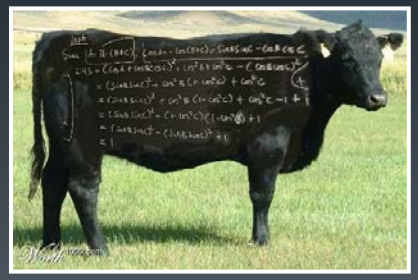
Range 5 to 23
Average = 12.04

Validating LOF Alleles

- Putative LOF candidates need to be tested in a larger population
  - Custom genotyping assay for LOF alleles developed in 2015
  - 10,000 Angus females will be sampled and genotyped on the custom array
    - Lethals can be identified with high confidence
      - No homozygotes in all the samples-implied lethals
    - Allele frequencies can be estimated in a large population
    - 6,050 heifers already sampled from 52 herds

Make a genotyping chip and put all candidates on it
(Practical limit is 200,000 variants)

Genotype 10,000 heifers
THIS IS GOING TO COST ~$400,000



See which ones NEVER turn up as homozygotes
(out of the 200,000 we tested)

More Still to Come!

- Generation of molecular EPDs for fertility based on the genotypes
- Development of selection indexes (Mike MacNeil)
  - Include and appropriately weight fertility in multi-trait selection decisions
- Development of Decision support software to optimize breeding schemes (Brian Kinghorn)
  - MateSel
- Development of web-based educational and training programs (Rolf and VanEenennaam)
- Develop a simulation exercise to demonstrate the effect of DGV for heifer and sire fertility on reproductive performance and profitability (Smith)

United States Department of Agriculture
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