Pooling DNA to Investigate Cattle Infertility

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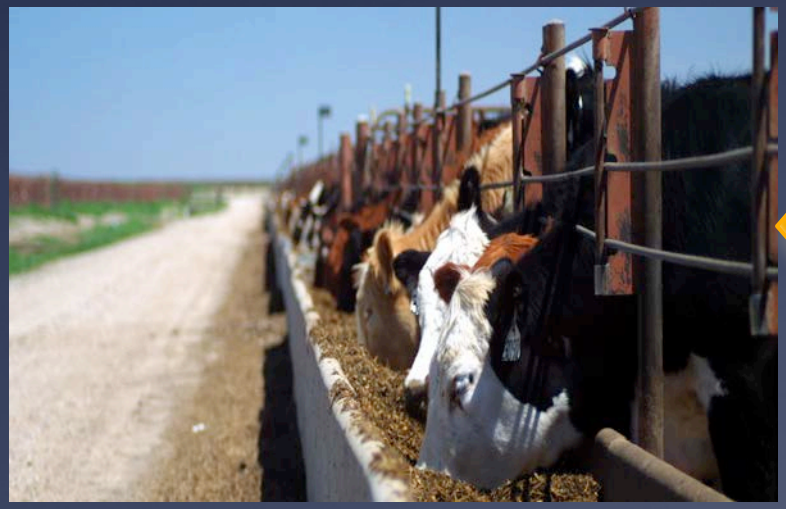
Background

- Reproductive efficiency
  - Lifetime production of the cow
  - Important element of cow-calf component of cattle industry
  - Female becomes a liability in the herd with no calf for producer to market



OR

Objective

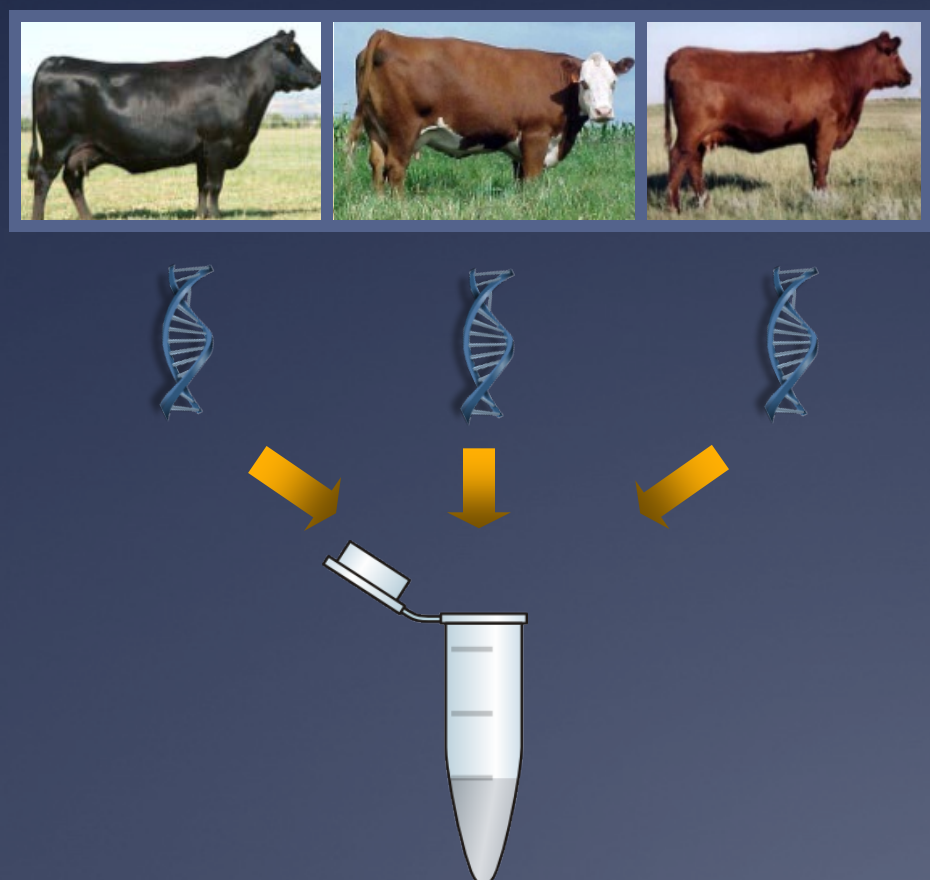
* Identify regions of the genome associated with reproductive efficiency in beef cattle

Challenges when evaluating reproductive traits

- Low heritability (0.10-0.40)
- Complex trait
  - Influenced by multiple genes
  - Creates difficulty when identifying genomic regions

Tools for overcoming challenges

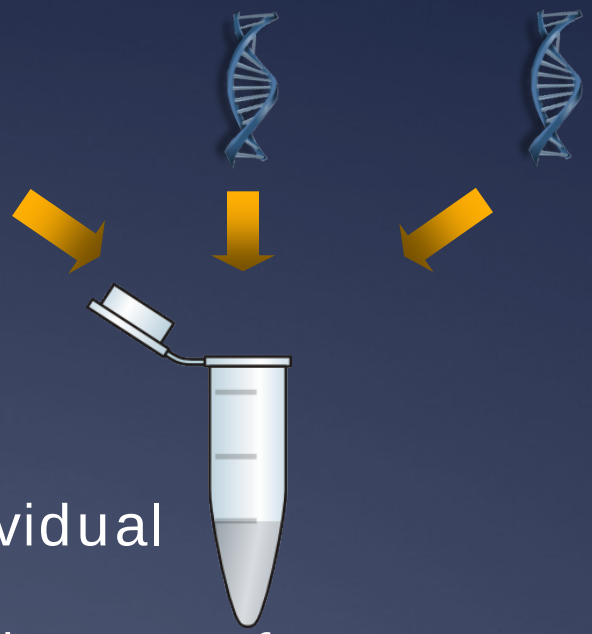
Pooling

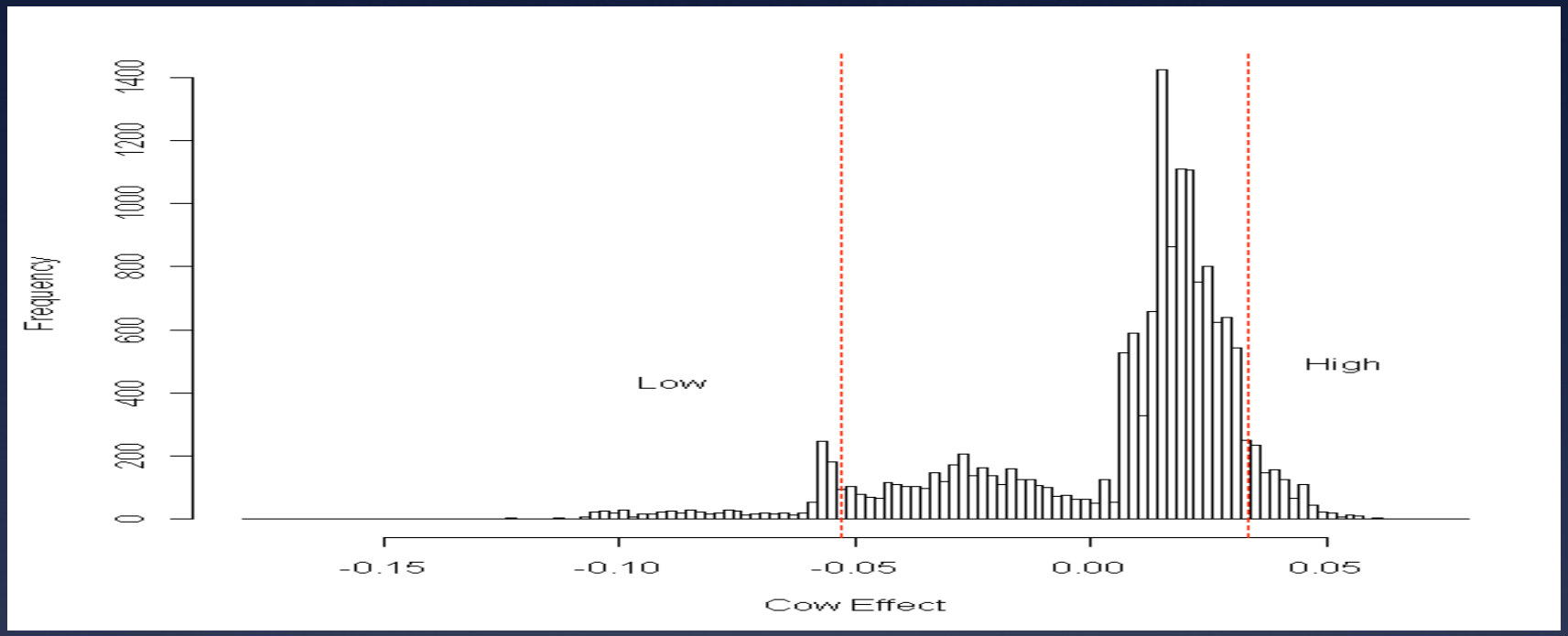


770 K SNP chip

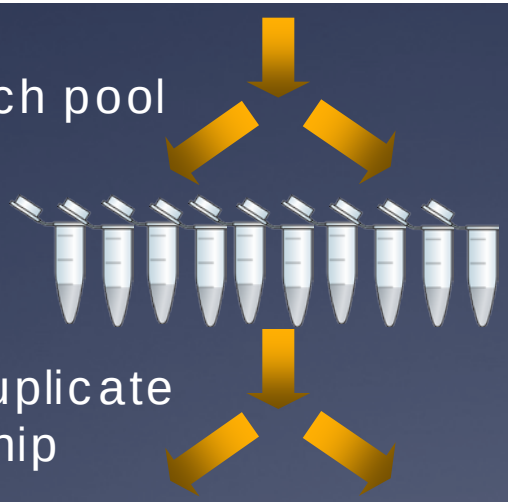
Steps in creating DNA pools

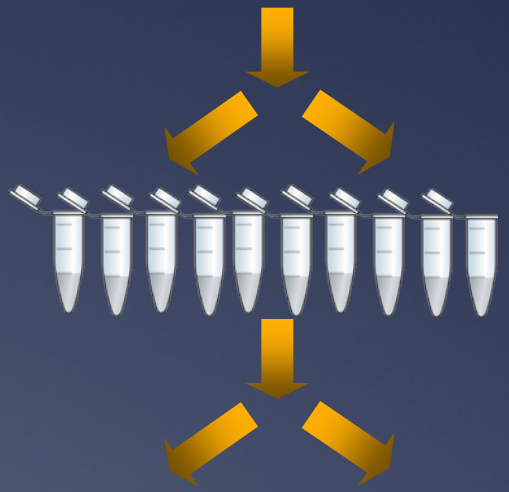
- Extract DNA
  - Tissue
  - Blood
  - FTA cards
- Determine quality and quantity
- Pool equal amount of each individual
- Properly mix pool to ensure consistency of individuals in pool



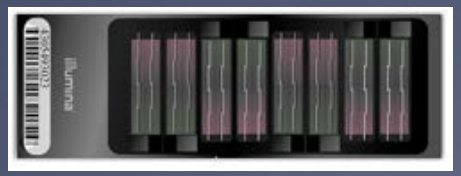


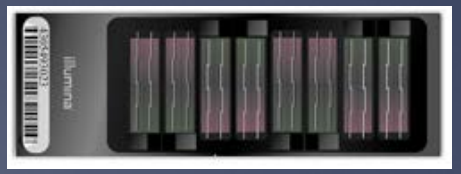
100 animals for each pool





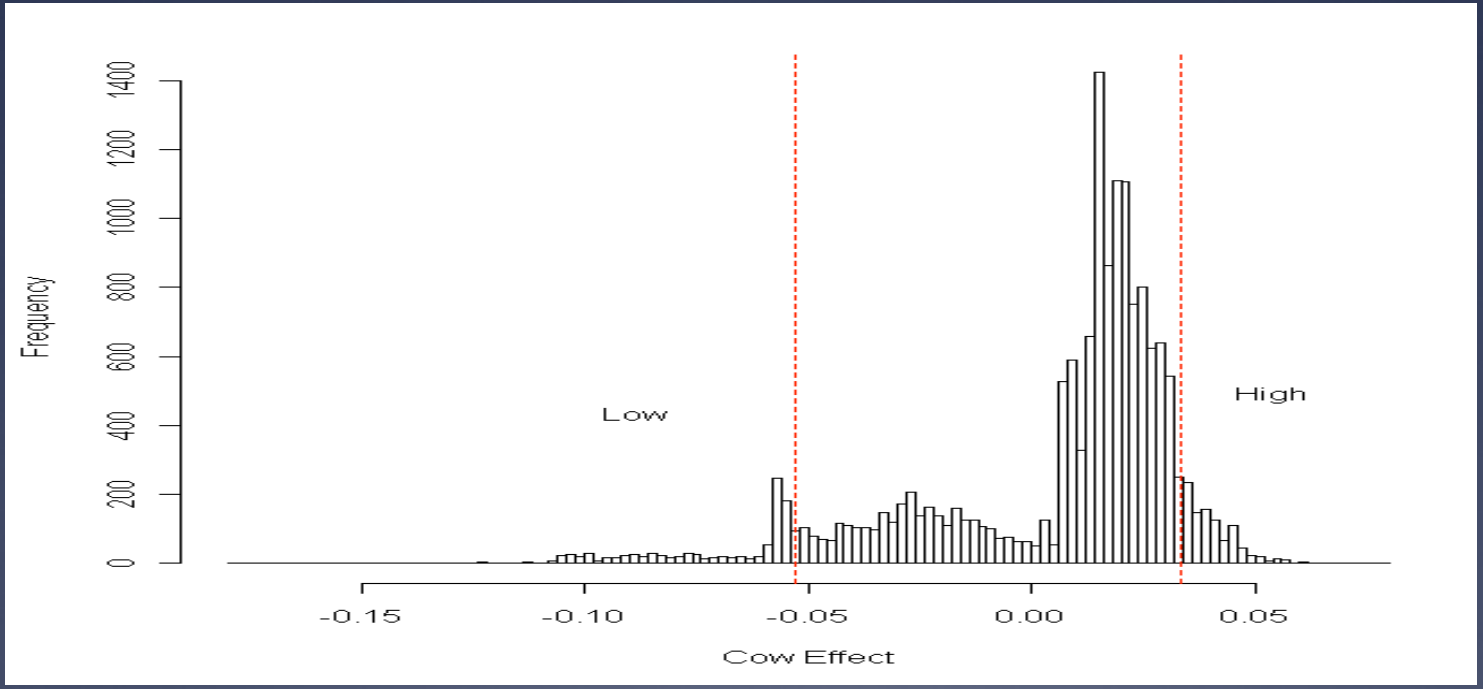
Each pool run in duplicate on 770 SNP chip





Cattle populations

- USMARC population
  - Genetically diverse populations
    - British and Continental breeds
  - 1000 females of low reproductive efficiency
  - 1000 females of high reproductive efficiency
  - 10 pools of 100 individuals for each phenotype



Cattle populations

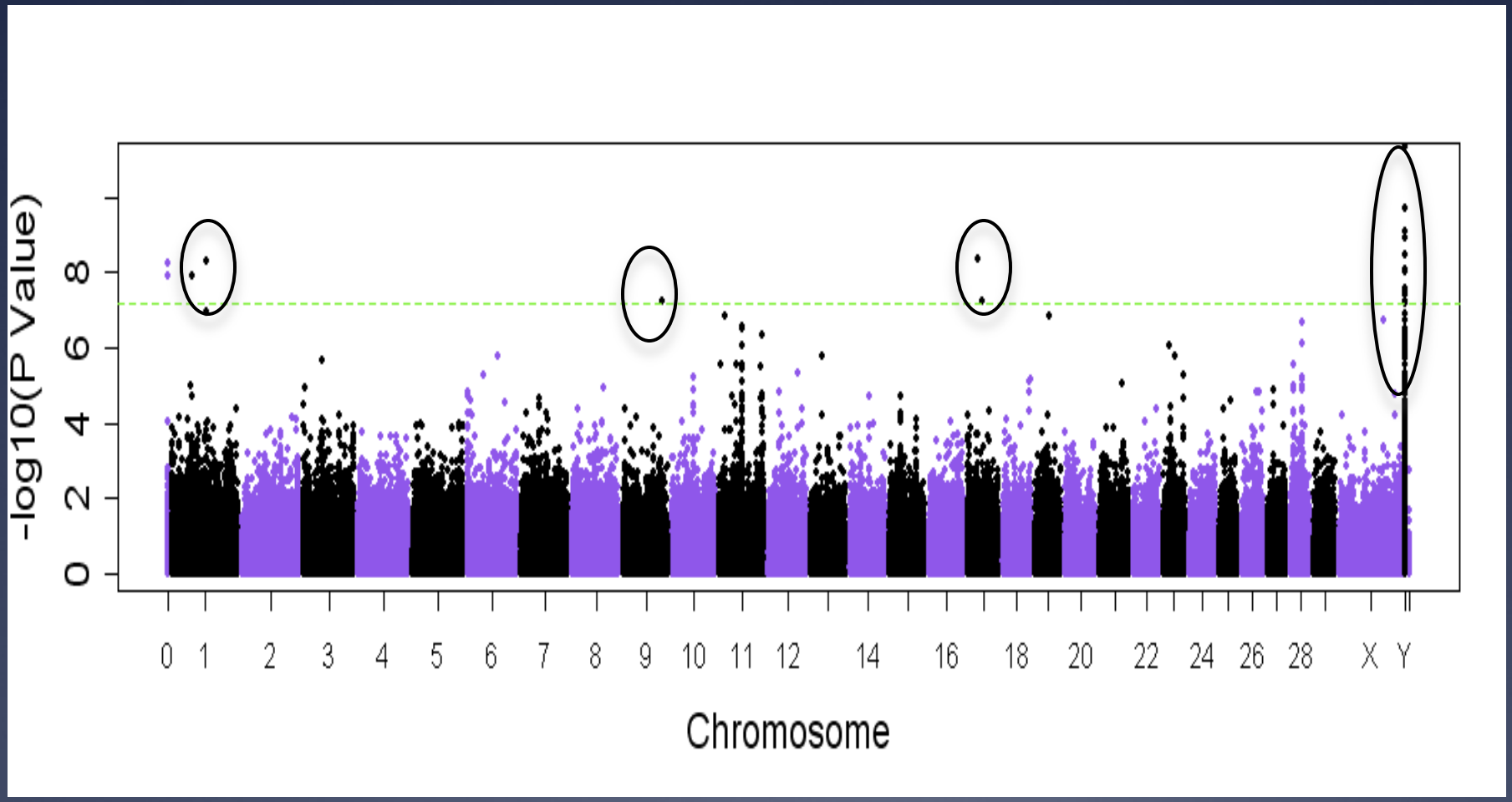
- Central Florida population
  - *Bos indicus* influence
    - Brangus
    - Simbrah
    - Braford
  - Reproductive records from 2 breeding seasons
    - Open/open
    - Open/pregnant
    - Pregnant/open
    - Pregnant/pregnant

Additional populations

- Western Nebraska population
  - Records from 2007 born heifers
    - Records for three consecutive breeding seasons
    - Open females are culled after first failure
- Milt Thomas at NMSU
  - Records from 7 small populations

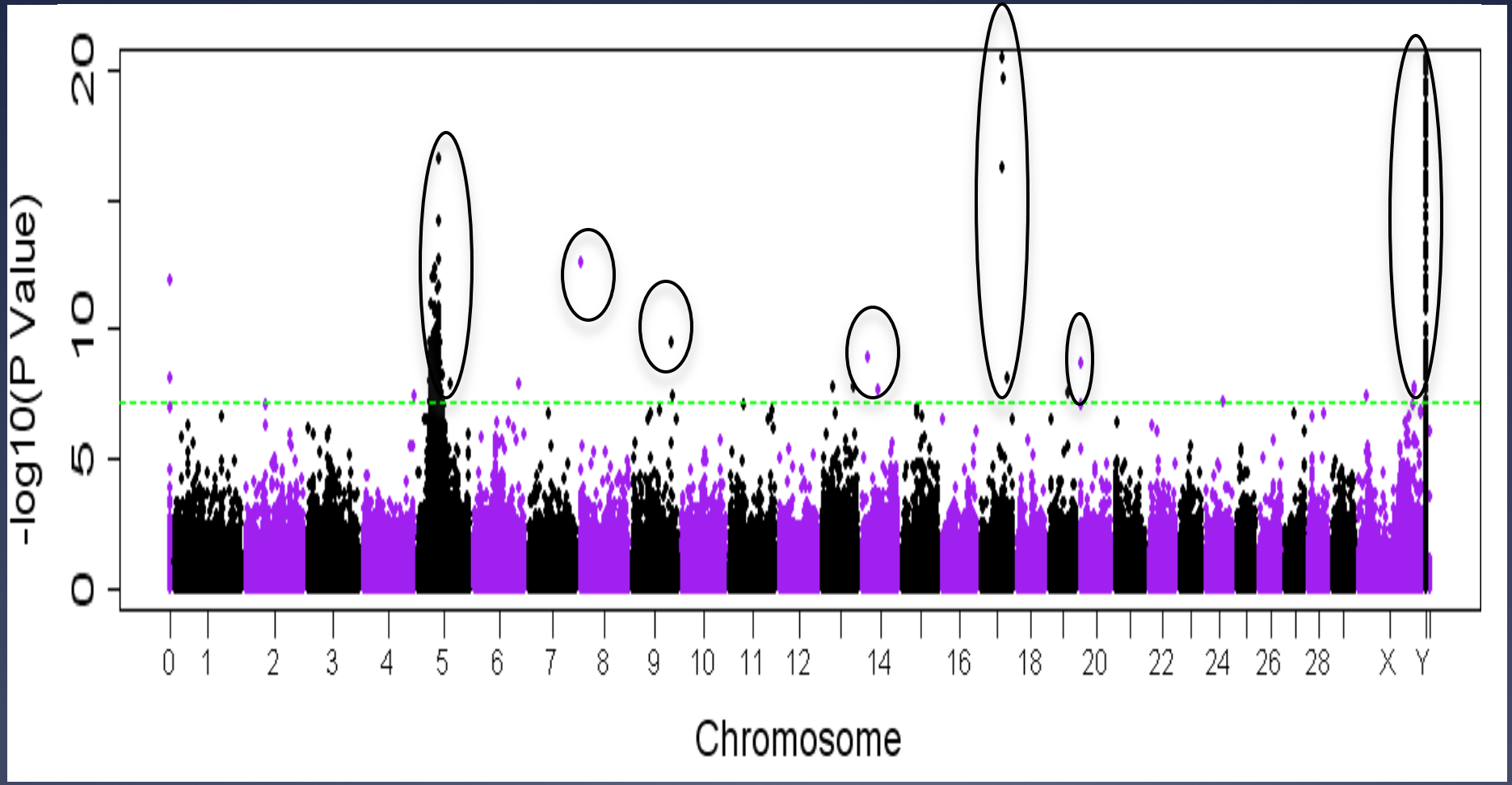
MARC population

- Identified multiple regions across genome associated with reproductive efficiency
  - BTA 1, 9, 17
  - Chromosome Y?



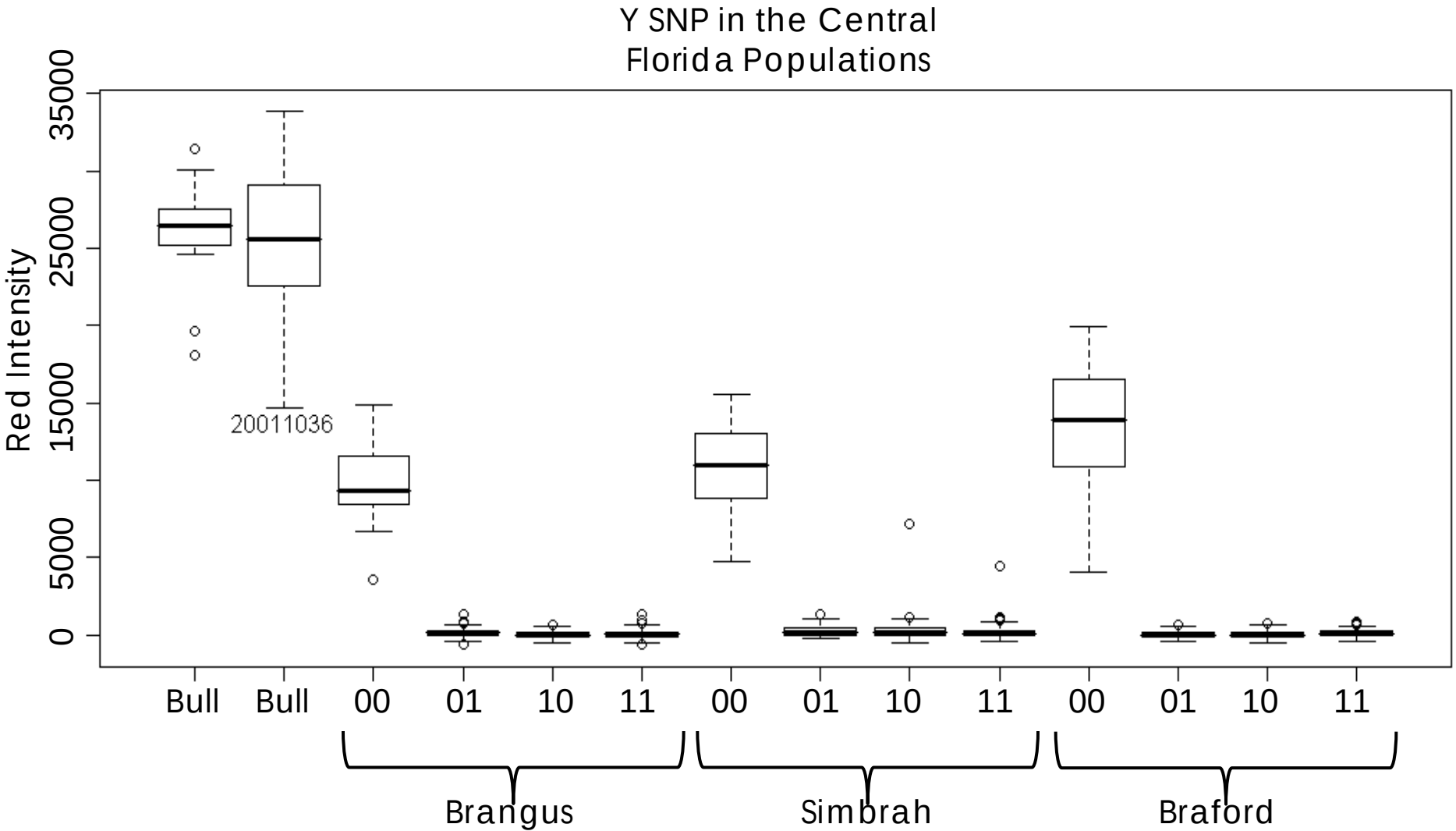
Central Florida population

- Identified multiple regions across genome associated with reproductive efficiency
  - BTA 5, 8, **9**, 14, **17**, and 20
  - Chromosome Y



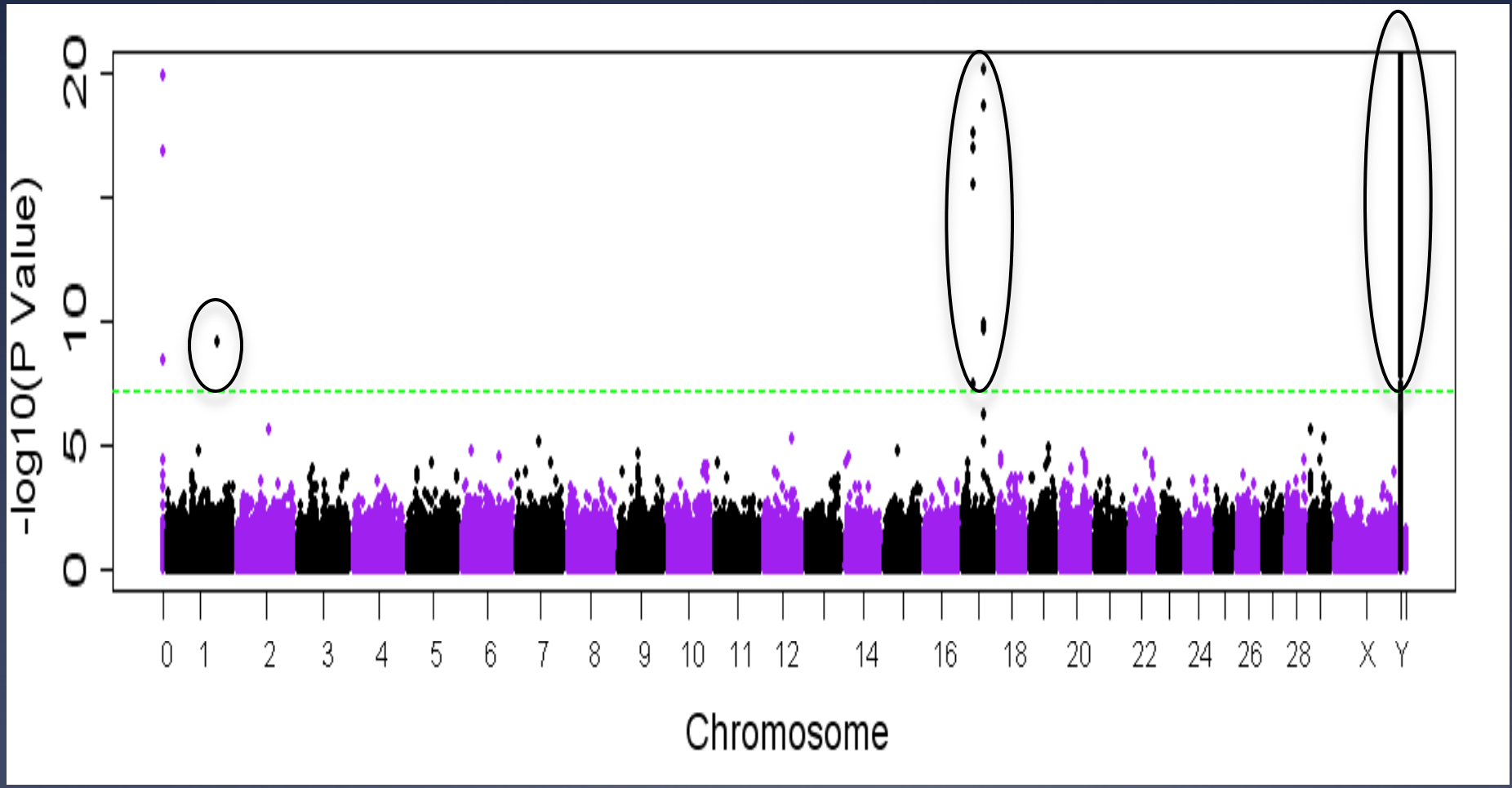
Y SNP in males and females?

- Only see Y SNP in open and low reproductive pools



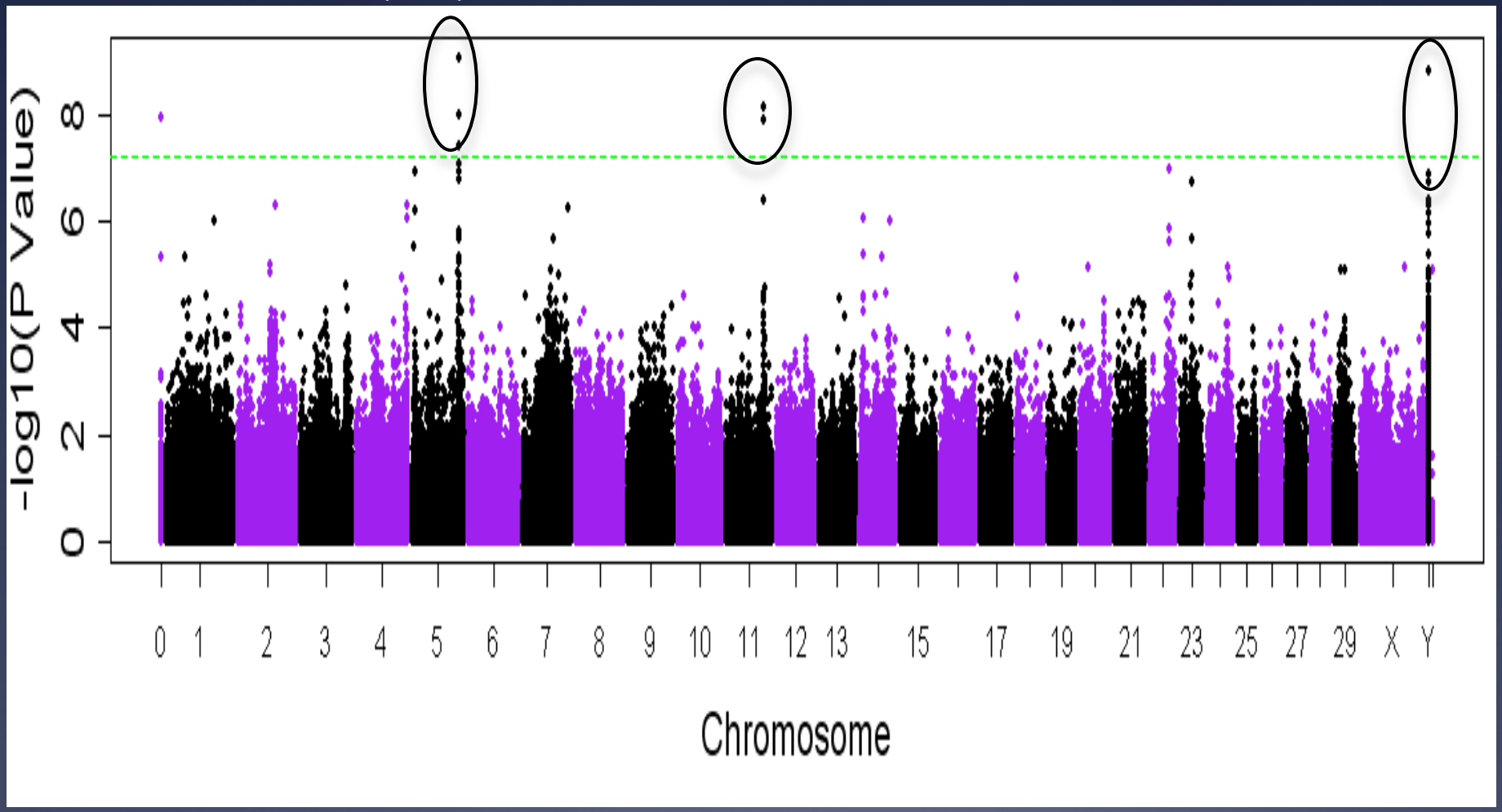
Western Nebraska population

- Identified multiple regions across genome associated with reproductive efficiency
  - BTA **1, 17, Y**

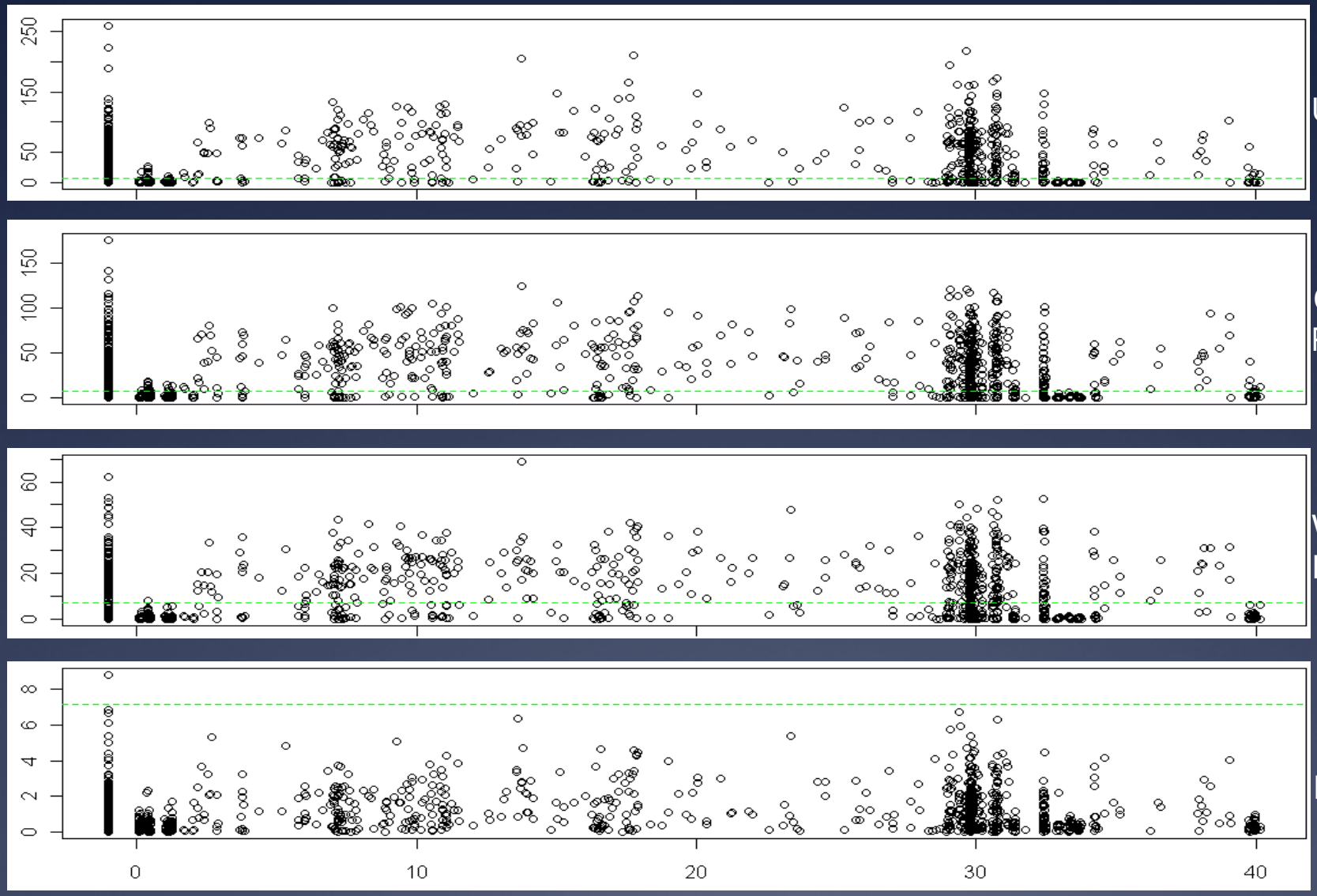


Additional populations

- Identified multiple regions across genome associated with reproductive efficiency
  - BTA **5**, 11, and **Y**



SNP across the Y chromosome

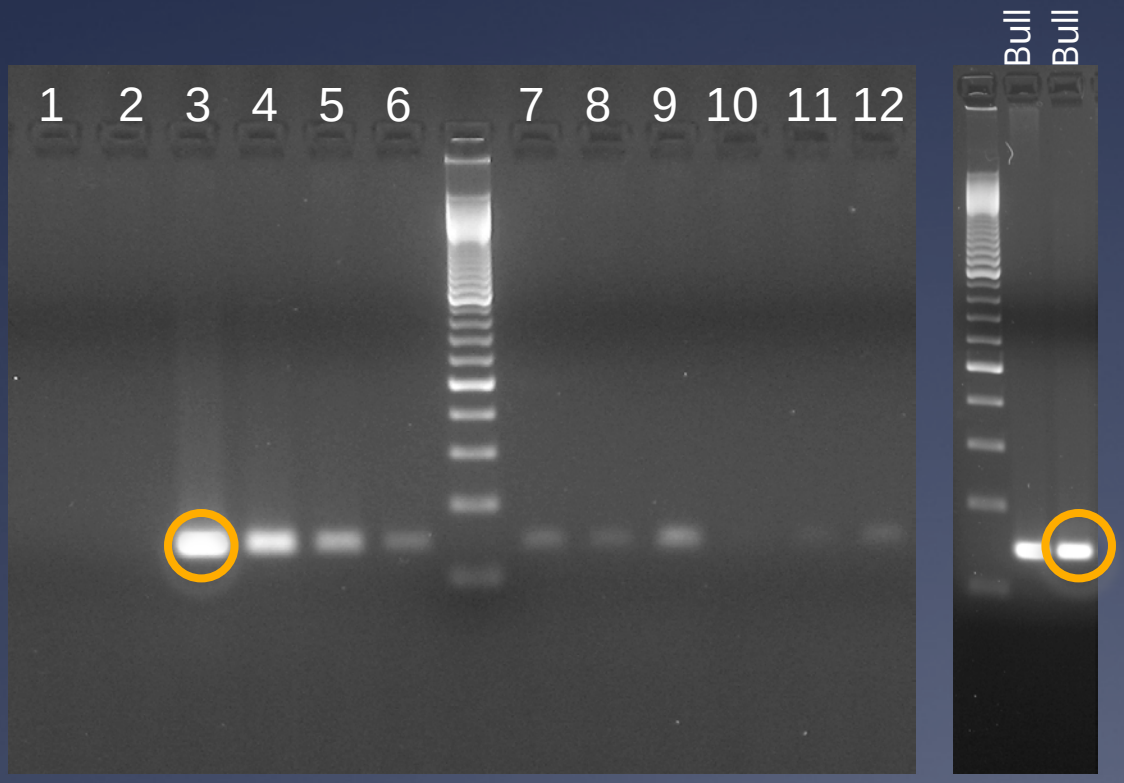


Why do we see Y SNP in the open females?

- Can we determine which females that possess the Y SNP?
- Evaluate individual females that make up open pool
  - Are all the open females contributing to the Y SNP?

Y chromosome in the pools

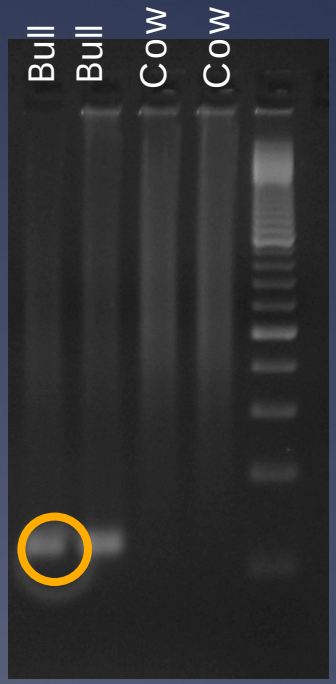
- Evaluate individuals of pools
  - PCR test specific to Y chromosome
    - PCR test is used to sex embryos
    - Only positive for males



% pool positive for sexing test	
Brangus	21%
Simbrah	27%
Braford	29%
USMARC	21%

Y SNP in pools

- Evaluate specific Y SNP on 770 K SNP chip
  - Developed PCR tests for Y SNP that are significant
  - We see different patterns in females that are positive for sexing PCR test (3-10% open females positive)
  - Different fragments of Y chromosome?

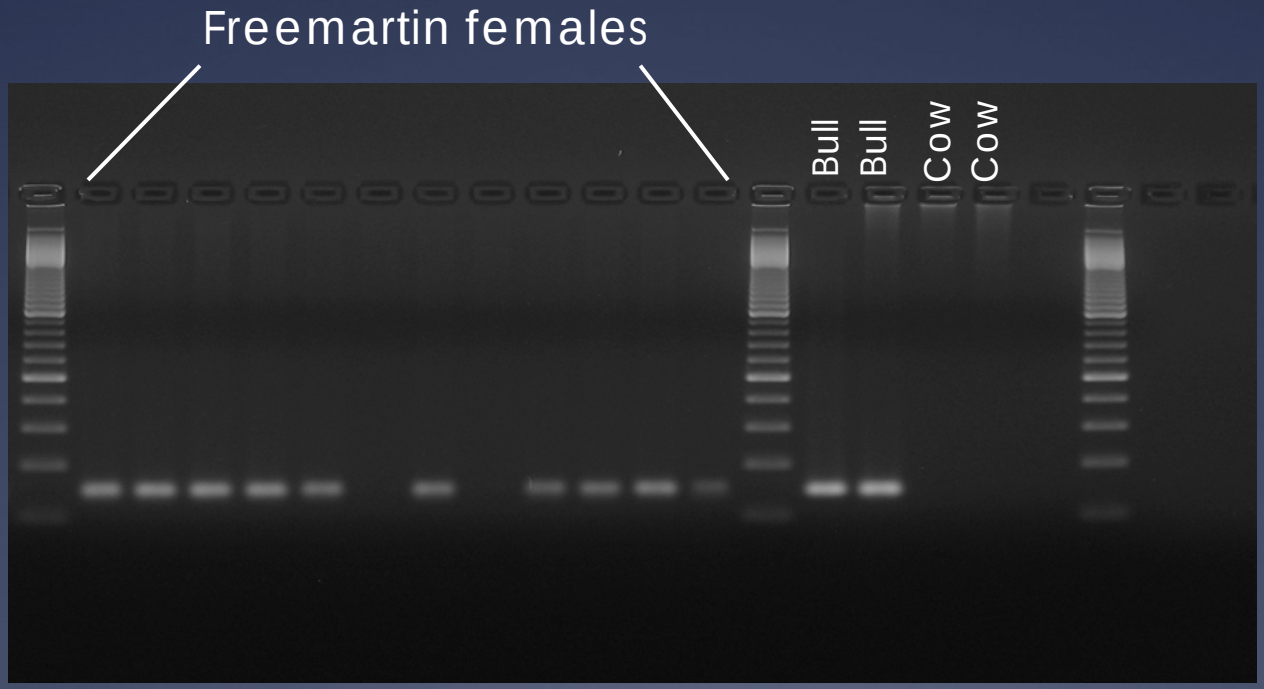


What is causing the Y SNP in females?

- Undetected freemartins?
  - Females commonly calve on pasture
    - May miss calves that are born twins
  - Reproductive tracts are checked in Central Florida population at prebreeding check
  - Twins are recorded at USMARC

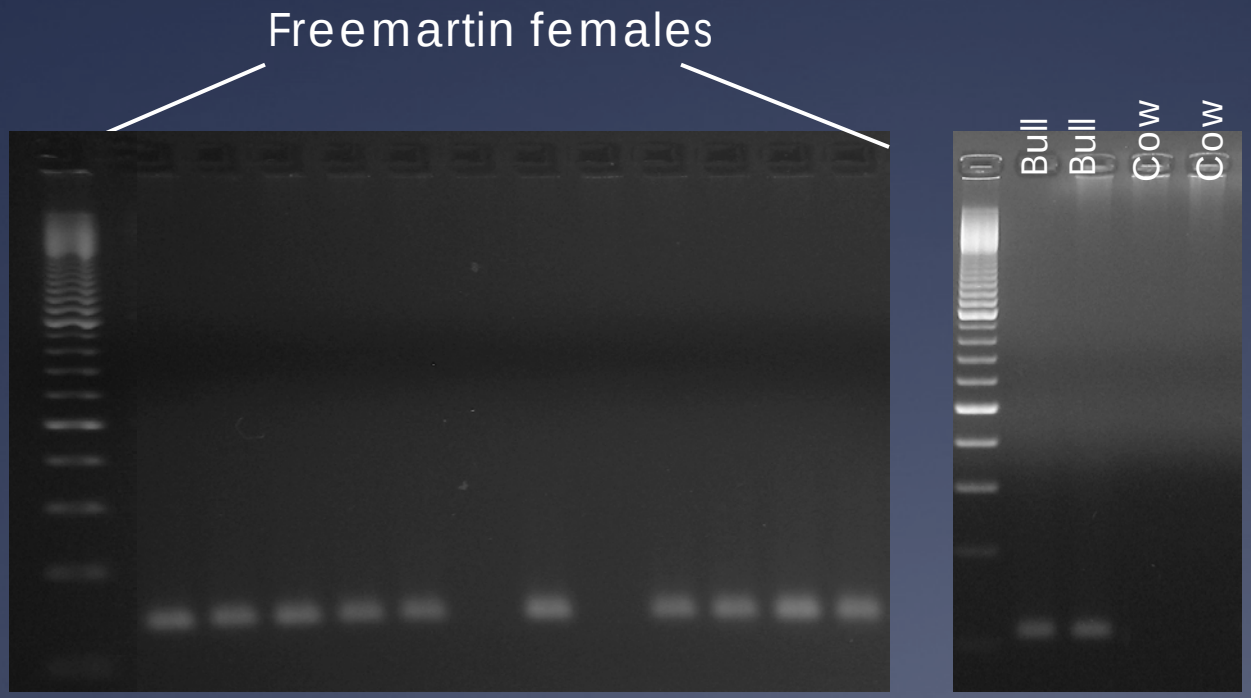
Freemartins in pools?

- Use PCR to test for genomic regions of Y in individuals that made up pool
  - Sexing PCR test



Freemartins in pools?

- Use PCR to test for Y SNP that are significant
  - All females that are positive for sexing primers are also positive for Y-SNP tests
  - Are freemartins the only cause of the Y?



% + Sexing primer test	% + Y-SNP primer test
96%	96%

Conclusions

- Identified regions of the genome associated with reproductive efficiency
- Able to replicate these results in additional populations
- Identified Y SNP in individuals of open pools that may contribute to low reproductive efficiency (approx. 3-29% of the open population)

Current status

- Evaluate Y-chromosome anomaly further
- Fine map regions on autosomes identified in current project
- Evaluate other chromosomal abnormalities

Acknowledgements

- MARC cattle operations
- Central Florida Ranch
- Western Nebraska Ranch
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Thank You!