# *Genetics of Heifer Fertility*

## Collaborator*S* & Dr. Milt Thomas

### Gerald Thomas Chair Lecture

## Genetics of Heifer Fertility: Molecular Advancements in Reproduction: From hair to calves.

**Outline:**

I. History/Background
II. **Discovery:** spots on chromosomes + genes
III. Building research populations
IV. International-sharing (AU-Beef CRC)
V. Conclusions



## Molecular Advancements in Reproduction: From hair to calves

### Participants: Past to Present

Konni Shirley, Pablo Luna, Sunday Peters, Kasey DeAtley, Gail Silver, Amelia Clayshulte, Tanner Schaub

Juan Medrano, Gonzalo Rincon, David Riley, Chad Chase

Jim Reecy, Rohan Fernando, Kadir Kazilkaya, Dorian Garrick, Zhiliang Hu, John Pollak, Bob Weaber, Warren Snelling

2006-35205-16651, 2008-35205-18751, 2009-35205-05100



National Beef Cattle Evaluation Consortium

Large Herd Managers Symposia (2005)

↓

USDA National Institute of Food and Agriculture

**Identification of Molecular Markers to Improve Fertility of Beef Cattle; 200835205-18751.**

↓

NBCEC Workshop Genetics of Reproduction ➡ **BIF**

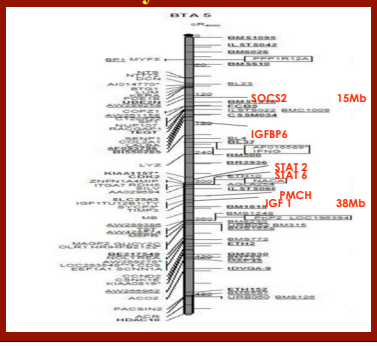


***Long-term goal:*** understand in molecular detail the genetic pathways regulating reproductive performance in beef cattle, with the intent of using this information to develop genetic improvement programs for fertility.

QTL Detection
↓
Fine Mapping
↓
Validation of MAS tools in commercial herds across environments and production systems.
↓
Technology Transfer



## II. Discovery: SNP from the GH-IGF axis




+

**Pregnancy Associated Plasma Protein (PAPPA) or Pappalysin**

***Chromosome 16***

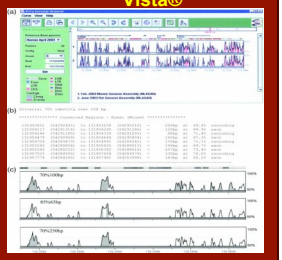
## Bovine Resequencing Population

a. No common ancestors in 3 generation pedigree.
b. Resequenced conserved regions of candidate genes
c. 75 SNP discovered.

Vista®

- Dairy breeds (n = 8/breed):

- Beef breeds (n = 6/breed):





## Assessment Population: Angus – Brahman – Romosinuano (n = 650)

STARS – Brooksville, FL

**Diallele** = Mating system involving all breed-parent combinations to create hybrids.

**Admixed Population** = Group of animals with mixed ancestry because have inherited some fraction of their genome from different ancestry populations (e.g., populations stratification).

**Ancestry-Informative Markers (AIM)** = Set of polymorphisms with significant differences in allele frequencies between breed groups.



## Genotyping Results

| Gene Name | SNP_assayed | SNP with MAF >.10 |
|---|---|---|
| Growth Hormone Receptor (GHR) | 1 | -- |
| Glycosylation-Dependent Cell Adhesion Molecule 1 (GLCM1) | 9 | -- |
| Insulin-like Growth Factor 1 (IGF1) | 6 | -- |
| Insulin-like Growth Factor Binding Protein 6 (IGFBP6) | 5 | -- |
| **Pregnancy-Associated Plasma Protein (PAPPA)** | **9** | **2** |
| Pro-Melanin Concentrating Hormone (PMCH) | 5 | -- |
| **Signal Transducer and Activator of Transcription 2 (STAT2)** | **6** | **1** |
| Signal Transducer and Activator of Transcription 6(STAT6) | 6 | -- |
| Suppressor of Cytokine Signaling 2 (SOCS2) | 12 | -- |
| Total | 59 | 3 |

**3 Polymorphic Markers**
**56 Ancestry Informative Markers**



## Mixed Model

$y_{ijklmn} = \mu + A_i + B_j + C_k + D_l + E_m + F_n + e_{ijklmn}$; **Proc Mixed, Proc Glimmix**

- $y_{ijkl}$= phenotypic value of traits,
- $\mu$ = population mean or frequency,
- $A_i$ = fixed effect of SNP genotype or haplotype,
- $B_j$ = fixed effect of year of birth,
- $C_k$ = fixed effect of age of dam (BIF, 2006),
- $D_l$ = covariate of coefficient of ancestry,
- $E_m$ = covariate of Julian birth date,
- $F_n$ = random effect of sire using the *Z* statistic to test if Ho:$\sigma_w^2 = 0$ (Littell et al., 1996),
- $e_{ijklmn}$ = random residual error.



## Least squares means ± SE and levels of significance for fertility traits among SNP genotypes of PAPPA

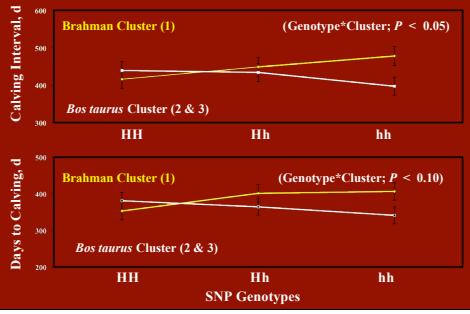
| Trait | N | SNP 1 PAPPA 11 | 12 | 22 | *P*-value |
|---|---|---|---|---|---|
| Age at First Calving, d | 551 | 768.6 ± 36.8 | 787.9 ± 9.6 | 778.8 ± 11.2 | 0.9999 |
| Perfect, % | 551 | 53.8 | 56.3 | 53.7 | 0.8402 |
| Calving Interval, d | 441 | 500.1 ± 37.4$^a$ | 450.4 ± 9.6$^b$ | 421.2 ± 8.0$^b$ | 0.0595 |
| Days to Calving, d | 441 | 448.8 ± 38.9$^a$ | 387.6 ± 10.3$^b$ | 360.9 ± 8.6$^b$ | 0.0289 |
| Pregnancy Rate, % | 640 | 77.8$^a$ | 85.8$^b$ | 89.6$^b$ | 0.0136 |

| Trait | N | SNP 2 PAPPA 11 | 12 | 22 | *P*-value |
|---|---|---|---|---|---|
| Age at First Calving, d | 548 | 773.2 ± 13.7 | 785.7 ± 9.7 | 781.9 ± 13.3 | 0.9999 |
| Perfect, % | 548 | 52.7 | 59.2 | 52.2 | 0.2823 |
| Calving Interval, d | 439 | 403.4 ± 13.2$^a$ | 439.9 ± 8.6$^b$ | 445.1 ± 12.1$^b$ | 0.0484 |
| Days to Calving, d | 439 | 344.9 ± 13.8$^a$ | 379.4 ± 9.1$^b$ | 384.4 ± 12.7$^b$ | 0.0288 |
| Pregnancy Rate, % | 640 | 90.6$^a$ | 87.9$^b$ | 86.3$^b$ | 0.0401 |

$^{ab}$Within a row, means without a common superscript differ ($P < 0.05$)



## Interaction among STAT2 SNP genotypes and ancestral clusters (ALL BREED GROUPS)



## Discovery: Spots on Chromosomes + Genes

***Objective 1:*** **Conduct a SNP-based whole-genome scan to identify QTL associated with heifer pregnancy rate and first service conception rate.**

Relationship fostered from efforts of IBBA –BIC.

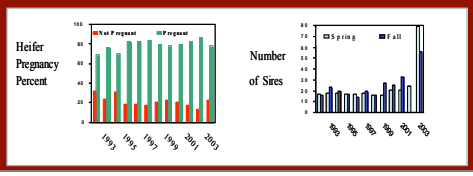


## Population Description

1. Brangus heifer pregnancy records collected since 1993.
2. DNA and phenotypes on >800 heifers from > sires born 2005, 2006, 2007.
3. Trait, pregnant as a yearling, yes (80.6%) or no (19.4%).
4. Simple statistics for growth and carcass traits:

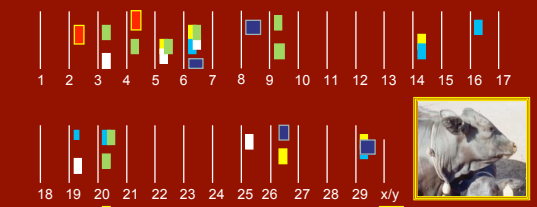
| | |
|---|---|
| Birth weight | 77.2 ± 0.4 lbs |
| 205-day weight | 532.2 ± 2.2 lbs |
| 365-day weight | 799.0 ± 3.3 lbs |
| Ribeye area | 9.8 ± 0.05 in² |
| Fat thickness | 0.24 ± 0.002 in |
| intramuscular fat % | 4.24 ± 0.032 % |

Historic Data provided by Matt Jones






## RESULTS of Objective I: QTL detection



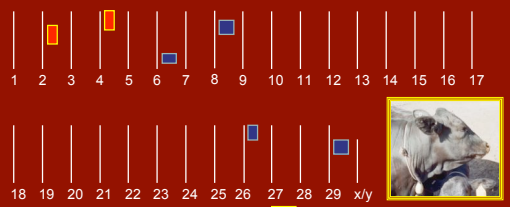

Birth Weight ,
365-day Weight
Intramuscular Fat %
Ribeye Area
Rib Fat

***Pregnant as yearling:*** **(12 to 15 mo) of age to calve by 24 mo.**

***First service conception:***



## RESULTS of Objective I: QTL detection




***Pregnant as yearling:*** **(12 to 15 mo) of age to calve by 24 mo.**

***First service conception:***



## Objectives

***More of Objective 1:*** **identify candidate genes using bioinformatics and a hypothalamic transcriptome reference resource (example from chromosome 8).**

QTL detection
↓
SNPlotZ
↓
GBrowse
↓
Candidate gene
↓
Transcriptome

NRSP-8 Bioinformatics

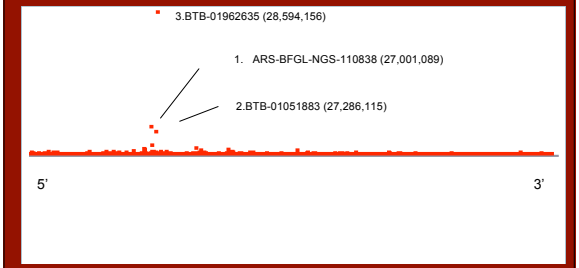
**http://www.animalgenome.org/**

NCGR

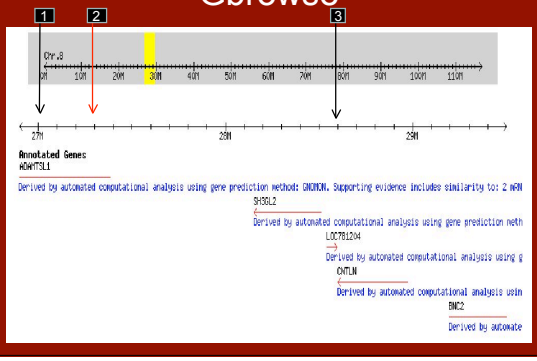
illumina



## SNPlotz Visualization of SNP effects for FSC ON BTA 8

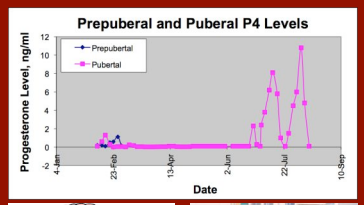


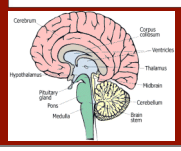
# In-silico gene identification in Gbrowse

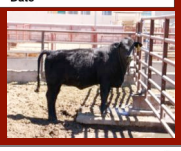



# Pipeline for Marker Discovery: Example from Chromosome 8

QTL detection
↓
SNPlotZ
↓
GBrowse
↓
***Candidates***
↓
***Transcriptome***





# Hypothalamic-expression of candidate genes for First Service Conception

| Gene Name | Transcriptomic expression difference from pre-pubertal to pubertal heifer (pre ---> pubertal) | Proposed function or tissue |
|---|---|---|
| ADAMTS-like 1 | down 4.89 | Extracellular matrix<br>Growth of female reproduction organs |
| SH3-domain GRB2-like2 | down 23.97 | Endocytosis pathway<br>Embryonic stem cells |
| LOC781204 | up 0.94 | Unknown |
| Centlein | up 4.6 | Centrosomal protein |
| Basonuclin 2 | up 8.27 | Stratified squamous epithelia<br>Reproductive germ cells |

QTL detection
↓
SNPlotZ
↓
GBrowse
↓
Transcriptome
↓
Proteome

Functional SNP ID
↓
***Assessment and Validation***

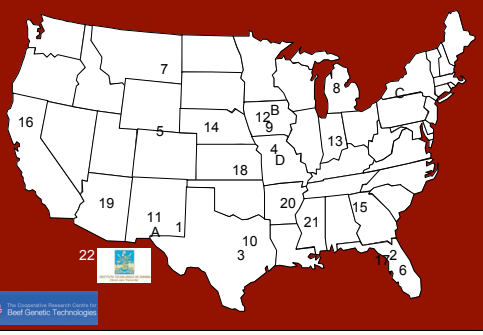
## *Objective 2:* Develop data and DNA resources.

a. Different environments and production systems.
AES = 60%; seedstock = 25%; commercial = 15%
Influence of Angus = 95%; Continental = 25%; Brahman = 40%

b. Opportunity for multiple traits.



**Locations of Cooperators (numbers 1-23) and Research Scientists (letters A-D)**





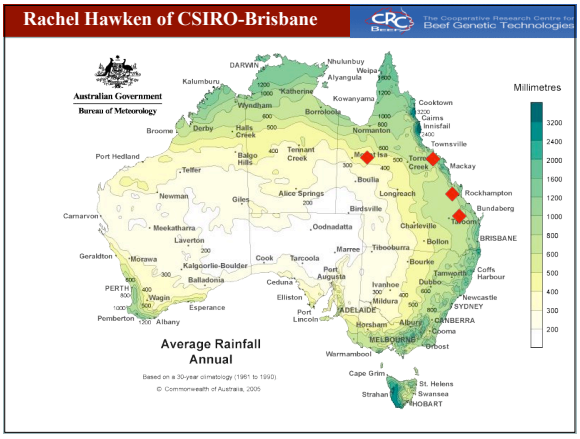

# Verify Results in Other Populations

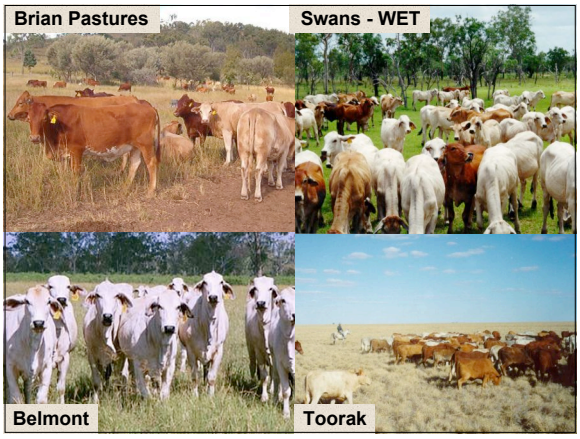
**International Consortium:**

The Cooperative Research Centre for Beef Genetic Technologies

**Rachel Hawken of CSIRO-Brisbane**

Rachel Hawken of CSIRO-Brisbane

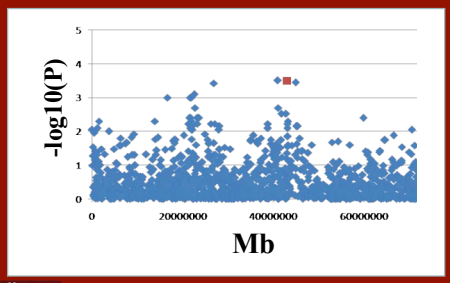



## Rachel's Current Conclusions

- A panel of markers will be chosen for commercialisation and integration into industry
- Little in common between breeds for reproduction traits
  - Factor of distance from causative mutation and linkage disequilibrium
  - Different breeds behave differently
- Require new unrelated populations for validation efforts

## Evaluation of Spots across Research Populations

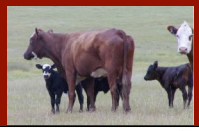



# Conclusions

***Molecular Advancements in Reproduction: from Hair to Calves***

**1.Collaborations needed to move research forward.**
**2.Resequencing candidate genes can yield effective SNP for genotype to phenotype associations.**
**3.Genomic approaches reveal important chromosome regions.**
**4.Multi-breed???**
**5.Gap in research between whole genome approach (BV or Spot on Chromosome) and variation caused by alleles of a gene.**

***Much work to do!***





# Discussion