Fertility in Beef Cows
Tom Geary
Reproductive Physiologist

Fertility of a Single Service: Beef Cattle
Early Embryonic Loss
Maternal Recognition of Pregnancy Failure
- most loss occurs before d 20

Annual Cost of Early Embryonic Loss to U.S. Beef Industry

<table>
<thead>
<tr>
<th>Beef cows &amp; heifers</th>
<th>40,000,000</th>
<th>x 25%</th>
<th>x 3 lb/d</th>
<th>600,000,000</th>
<th>6.25%</th>
<th>150,000,000</th>
<th>1.5%</th>
</tr>
</thead>
<tbody>
<tr>
<td>20 d later pregnancy</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>40 d later pregnancy</td>
<td></td>
<td>60 d later pregnancy</td>
</tr>
<tr>
<td>calf gain lost</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>lbs lost gain</td>
<td></td>
<td>lbs lost gain</td>
</tr>
<tr>
<td>Pregnancy d 27</td>
<td>70%</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Pregnancy d 42</td>
<td>62%</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Calving</td>
<td>60%</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Dairy Industry Loss:
$ 600,000,000

787,500,000 lbs x $1.10/lb = $ 866,250,000

Loss is to Cow/Calf Producer!

Why / Where is pregnancy failing?
Progress difficult without earlier pregnancy diagnosis

Nutrient Partitioning
1. Maintenance
2. Activity
3. Growth
4. Milk production
5. Body reserves (FAT)
6. Reproduction

Proper nutrition is obligatory to reproductive success, but not focus of presentation.

Reproductive Measures that Result in Pregnancy
Single most important measure of fertility for cow/calf producer?

- Annual BSE

Each bull is expected to contribute to the production of 20 to 30 calves, so the fertility of each bull is at least 20 times more important than a cow.

Limitations:
- Fertility measure on that given day
- Only about 1/3 of the “Fertility Picture”

Importance of Bull Fertility on Breeding Season Pregnancy Rate

<table>
<thead>
<tr>
<th>Year 1</th>
<th>Year 2</th>
</tr>
</thead>
<tbody>
<tr>
<td>BSE</td>
<td>Control</td>
</tr>
<tr>
<td>%</td>
<td>%</td>
</tr>
<tr>
<td>No. Cows</td>
<td>572</td>
</tr>
<tr>
<td>Pregnancy Rate</td>
<td>86%</td>
</tr>
</tbody>
</table>

Wiltbank et al., 1983

Effect of Social Dominance on Percentage of Calves Sired

<table>
<thead>
<tr>
<th>Bull</th>
<th>Year 1</th>
<th>Year 2</th>
<th>Year 3</th>
<th>Year 4</th>
<th>Year 5</th>
</tr>
</thead>
<tbody>
<tr>
<td>A Age</td>
<td>10</td>
<td>11</td>
<td>12</td>
<td>13</td>
<td>14</td>
</tr>
<tr>
<td>B Age</td>
<td>4</td>
<td>5</td>
<td>6</td>
<td>7</td>
<td>8</td>
</tr>
<tr>
<td>C Age</td>
<td>3</td>
<td>4</td>
<td>5</td>
<td>6</td>
<td>7</td>
</tr>
<tr>
<td>D Age</td>
<td>2</td>
<td>3</td>
<td>4</td>
<td>5</td>
<td>6</td>
</tr>
</tbody>
</table>

- What if the dominant bull is sterile?

Effect of Social Dominance on Percentage of Calves Sired

<table>
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<tr>
<th>Bull</th>
<th>Year 1</th>
<th>Year 2</th>
<th>Year 3</th>
<th>Year 4</th>
<th>Year 5</th>
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<td>6</td>
<td>7</td>
<td>8</td>
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<tr>
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<td>4</td>
<td>5</td>
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<td>7</td>
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<tr>
<td>D Age</td>
<td>2</td>
<td>3</td>
<td>4</td>
<td>5</td>
<td>6</td>
</tr>
</tbody>
</table>

Bull Fertility Measures – Fort Keogh

- Annual breeding soundness exams (BSE) on all bulls.
- Puberty exams at 10.5 mo of age (n = 1,100).
  - Phenotypes for genomics
  - Sperm morphology traits
    - Heritability
    - Puberty
Heritability of Bull Fertility Traits

<table>
<thead>
<tr>
<th>Trait</th>
<th>( h^2 )</th>
</tr>
</thead>
<tbody>
<tr>
<td>Scrotal circumference, cm</td>
<td>0.67 ± 0.09</td>
</tr>
<tr>
<td>% Normal</td>
<td>0.16 ± 0.07</td>
</tr>
<tr>
<td>% Knobbed acrosome</td>
<td>0.02 ± 0.07</td>
</tr>
<tr>
<td>% Head defects</td>
<td>0.00 ± 0.05</td>
</tr>
<tr>
<td>% Distal midpiece reflex</td>
<td>0.01 ± 0.04</td>
</tr>
<tr>
<td>% Dag defect</td>
<td>0.50 ± 0.10</td>
</tr>
<tr>
<td>% Bowed midpiece</td>
<td>0.18 ± 0.07</td>
</tr>
<tr>
<td>% Proximal droplets</td>
<td>0.37 ± 0.06</td>
</tr>
<tr>
<td>% Distal droplets</td>
<td>0.09 ± 0.06</td>
</tr>
<tr>
<td>% Coiled principle piece</td>
<td>0.07 ± 0.05</td>
</tr>
<tr>
<td>% Bent principle piece</td>
<td>0.18 ± 0.08</td>
</tr>
<tr>
<td>Gross motility score</td>
<td>0.20 ± 0.07</td>
</tr>
<tr>
<td>% Progressive motility</td>
<td>0.20 ± 0.08</td>
</tr>
</tbody>
</table>

Selection for improved sperm morphology should increase fertility!

Key Parameters of Sperm Fertility

MEMBRANE INTEGRITY (1)
- Broken, opens the door to DNA content

DNA CONTENT (2)
- Controls DNA fragmentation

ACROSOME (3)
- Key role in fertilization

CAPACITATION (4)
- Prepares sperm to fertilize

MITOCHONDRIAL STATUS (5)
- Influences motility

Measuring these key physiological functions provides insight into the fertilization potential of sperm.

Currently: 2 studies on bull fertility with Guava EasyCyte.

Biological Markers Associated with Fertility

Ubiquitin  | PNA  | PAWP

Identify bulls of greater/lesser fertility???
Selection for improved fertility in bulls???

IVF Using Magnetic Sperm Depletion

Sperm sorted with Ubiquitin antibody resulted in 2 to 4x higher fertilization rate.

Bull Fertility Biomarker Trial - UBI

Normal Histogram  | Abnormal Histogram

Future Research Bull Fertility Analysis

<table>
<thead>
<tr>
<th>ASSAY</th>
<th>Microscope</th>
<th>CASA</th>
<th>EasyCt</th>
</tr>
</thead>
<tbody>
<tr>
<td>Motility</td>
<td>++</td>
<td>+++/</td>
<td>-</td>
</tr>
<tr>
<td>Concentration</td>
<td>-</td>
<td>+++/</td>
<td>+++</td>
</tr>
<tr>
<td>Viability</td>
<td>+</td>
<td>+</td>
<td>+++</td>
</tr>
<tr>
<td>Acrosome</td>
<td>+</td>
<td>+</td>
<td>+++</td>
</tr>
<tr>
<td>Mitochondria</td>
<td>-</td>
<td>-</td>
<td>+++</td>
</tr>
<tr>
<td>Capacitation (Ca)</td>
<td>-</td>
<td>-</td>
<td>+++</td>
</tr>
<tr>
<td>DNA Fragment</td>
<td>-</td>
<td>-</td>
<td>+++</td>
</tr>
<tr>
<td>Bacterial count</td>
<td>-</td>
<td>-</td>
<td>+++</td>
</tr>
<tr>
<td>Morphology/physiology</td>
<td>+/-</td>
<td>++/-</td>
<td>++/+++</td>
</tr>
<tr>
<td>Objectivity</td>
<td>+</td>
<td>++</td>
<td>+++</td>
</tr>
</tbody>
</table>

CASA = Computer Assisted Semen Analysis
EasyCt = Flow cytometer with various assays built-in

Additional “Chute Side” measures within 5 years
“Grass always greener on the other side of the fence!” Cow Fertility?

Fertile Mating?

Heritability Rates
- Birth Weight: 40%
- Weaning Weight: 30%
- Yearling Weight: 30%
- Fleshing Ability (BCS): 40%
- Frame Size: 55%
- Pelvic Area: 40%
- Fertility: 10%

Source: “Scientific Farm Animal Production”

Requirements for Reproductive Success
1. Puberty / Resume cycling ✓ / ✓
2. Fertile ovulation ✓
3. Conception (Ovum and Sperm) ✓
4. Pregnancy establishment ✓ d. 27
5. Pregnancy maintenance ✓

Successfully Reproduce

Fort Keogh Research:
- Identify genes/DNA markers involved with fertility in beef cattle.

Identification of New Phenotypic Traits involved in Fertility
- Beef Cow / Heifer
- Used AI to obtain numerous measures & keep bull fertility constant
- Ovulatory follicle size

Distribution of Ovulatory Follicle Size

Effect of Follicle Size on Pregnancy Rate (Timed-AI)

Perry et al., 2005
Fort Keogh Data

Embryonic Loss
Fort Keogh Reciprocal Embryo Transfer Study (2007-09)

**Goal:** Determine how size of the ovulatory follicle affects pregnancy establishment and maintenance. Does it affect oocyte maturation and/or viability? Does it affect the maternal reproductive tract in assisting pregnancy maintenance/establishment?

<table>
<thead>
<tr>
<th>No. of transfers</th>
<th>Donor</th>
<th>Recipient</th>
<th>Purpose</th>
</tr>
</thead>
<tbody>
<tr>
<td>71</td>
<td>Small</td>
<td>Small</td>
<td>Negative control</td>
</tr>
<tr>
<td>111</td>
<td>Small</td>
<td>Large</td>
<td>Effects 1° of oocyte origin*</td>
</tr>
<tr>
<td>122</td>
<td>Large</td>
<td>Small</td>
<td>Effects 1° of maternal origin*</td>
</tr>
<tr>
<td>50</td>
<td>Large</td>
<td>Large</td>
<td>Positive control</td>
</tr>
</tbody>
</table>

n = 1,164 suckled beef cows; 810 = donor and 354 = recipient

Suggests that ovulatory follicle size of the recipient is the only one that matters so effects of follicle size acting through maternal environment prepared for embryo.

Path Diagram: Embryo Donor Cow

**Path Diagram: Additional Fertility Measures**

1. Estradiol at time of breeding 18%
2. Progesterone on d 7 after breeding 15%
3. Progesterone before breeding 8%
4. Ovulatory follicle size 7%
5. Embryo quality 3%
6. Days postpartum at breeding 2%

**NOTE:** Cows were not used if they exhibited estrus.
Take Home Message – RET Study

- Fertility is much more complicated than we hope.
- The most important variable related to pregnancy success was estradiol concentration at the time of breeding.

If using synchronization and timed AI, insemination must coincide with expression of estrus.

Early pregnancy diagnosis to identify causes of pregnancy failure at Fort Keogh.
- d 19 with very good accuracy
  - Endometrial echotexture
  - CL blood flow

Current Technologies of IVF
- Ultrasound Blood – Pregnancy specific proteins

Serum Progesterone vs CL Blood Flow

Doppler Ultrasound Imaging of CL Vascularity

Sequential CL Vascularity for Pregnancy Diagnosis

Sequential CL Vascularity for Pregnancy Diagnosis
Thank You! Questions?

ROY WALLACE, SELECT SIRES

**Ubiquitin (UBI)**
- Proteolytic marker peptide that binds to the surface of defective sperm.
- Ubiquitination causes immobilization and/or resorption of these defective sperm during epididymal passage.
- Increased binding of anti-ubiquitin antibodies to the sperm surface reflect the occurrence of abnormalities.

**Peanut Agglutinin (PNA)**
- High affinity/strong specificity for disaccharides with terminal galactose, especially the D Gal α (1,3) D GalNAc disaccharide.
- Binds to the outer acrosomal membrane (OAM), exposed during the acrosome reaction.

**Lentil Lectin (LCA)**
- Shows a strong specificity to d-glucosyl and d-manosyl residues.
- Binds to the acrosome in normal sperm, whole surface in defective sperm.

**Postacrosomal Sheath WW-Domain-Binding Protein (PAWP)**
- Resides exclusively in the post-acrosomal sheath (PAS) region of the sperm head perinuclear theca (PT) and is expressed and assembled in elongating spermatids.
- Promotes meiotic resumption and pronuclear development during fertilization.
- Abnormal sperm may have unusually high levels of this protein.

**Bull Fertility Biomarker Trial**
- n = 162 samples.
- Samples analyzed via UBI, PNA, LCA, PAWP.
- Flow cytometry via GUAVA.
- Analysis performed via Excel and SAS.