Sex is THE most important genetic trait

10 years of commercially available sexed semen

- ♦ History
- Success Rates
- What has changed
- What to expect in the next 5 years

History

- Early 1980's Lawrence/Livermore Experiments
- 1989 L. Johnson Beltsville experiments with rabbits
- Early 1990's Mastercalf experiments UK and Ireland with IVF
- ◆Late 1990's XY, Inc experments with AI in Colorado
- ◆2002 First Commercial license Cogent
- 2007 XY, Inc. acquired by Sexing Technologies in Texas

Funding for much Ag research has changed from public to private

- Seeds
- Sexed semen

PATENTS ON SEXING SPERM

- ☑ Over 300 patents
- Most patented procedures not
 - efficacious or useful

Private funding made sexed semen practical

- Insufficient public funding
- Patents, licenses, royalties
- Sexing Technologies, Inc owns most relevant intellectual property
- Data information proprietary
- Attorneys are expensive

| US Holsteins Bred with Sexed Sperm | | |
|---------------------------------------|---------|------|
| Year | Heifers | Cows |
| 2006 | 1.5% | 0.1% |
| 2007 | 9.6% | 1.3% |
| 2008 | 14.2% | 2.1% |
| Hutchison and Norman, 2009 | | |

Problems

- Too expensive to use normal 1. numbers of sperm/dose
- 2. Sperm damaged slightly by the sexing process
- 3. Sorting frozen-thawed sperm and refreezing equals poor fertility

Technology Summary

- Speeds of sexing doubled
- Fundamental procedures unchanged
- Sperm still sorted one at a time
- Other methods don't work!

PURITY

- Can exceed 95%
- Industry standard = 90%
- More pure = more expensive
- Similar accuracy X and Y

| Normality of Calves from Sexed Sperm | | | |
|---|-------|---------|--|
| | Sexed | Control | |
| No. ^a | 1158 | 787 | |
| Abortion rate (%) | 45 50 | | |

Increasing numbers of sexed, frozen sperm above 2 million/ inseminate does not increase pregnancy rates greatly

279 279 **Gestation length (d)** Neonatal death (%) 3.5 4.0

1.23

34.1

91.5

241

1.22

33.9

91.7

239

Calving ease score

Live at weaning (%)

Weaning weight (kg)

^a N were lower for some responses.

Birth weight (kg)

Huge Differences

- Heifers
- Lactating dairy cows
- Postpartum beef cows
- Nursing 1st calf beef heifers
- Superovulated females
- Pre-pubertal heifers
- Heat-stressed cattle

Angus Heifers – 2 Bulls and 4 Inseminators – One Herd

| Treatment | No. heifers | % pregnant |
|----------------------------|-------------|-----------------|
| 20x10 ⁶ unsexed | 126 | 67ª |
| 4.5x10 ⁶ sexed | 126 | 51 ^b |
| 1.5x10 ⁶ sexed | 123 | 54 ^b |
| ^{a,b} (P<0.05). | | |
| | | |

Field Trial – Holstein Heifers (3 bulls, 2 inseminators)

| Treatment | No. | % pregnant |
|-------------------------------------|-----|------------|
| 2 x 10 ⁶ sperm sexed | 179 | 56% |
| 10 x 10 ⁶ sperm sexed | 180 | 62% |
| 10 x 10 ⁶ sperm, control | 88 | 61% |
| Schenk et al., 2005 | | |
| | | |

Holstein Heifers

| Treatment | Ν | %Pregnant |
|------------------------------|------|------------------------|
| 2.1x10 ⁶ sexed | 2319 | 38 ª |
| 10.0x10 ⁶ sexed | 2279 | 44 ^b |
| 2.1x10 ⁶ unsexed | 2282 | 55° |
| 10.0x10 ⁶ unsexed | 2292 | 60 ^d |

Estrus Synchronization

- Works well with sexed semen if AI 12-24 h after onset of estrus
- Fixed time Al not recommended

With Sexed Semen

- Best to have egg waiting for sperm
- Sexed sperm degrade while waiting
- Best to inseminate 6-12 h later than with unsexed sperm

| Timing of AI and Pregnancy |
|----------------------------|
| Rates, Sexed Sperm |

| 0.5 day | 266/586, 45.4% |
|---------|----------------|
| 1.0 day | 203/414, 49.0% |

| Estrotect | Semen | Ν | % Pregnant |
|------------|---------|-----|------------|
| Tripped | Control | 105 | 77 |
| Untripped | Control | 113 | 37 |
| Tripped | Sexed | 215 | 47 |
| Untripped | Sexed | 113 | 2 |
| Untripped* | Sexed | 110 | 39 |

Superovulated Dairy Cattle

| Treatment | N | Transferable Embryos | Unfertilized/ Degenerate |
|------------------------|-----|-------------------------|-----------------------------|
| Heifers, sexed semen | 130 | 6.1 | 4.4 |
| Heifers, control semen | 945 | 7.2 | 4.2 |
| Cows, sexed semen | 80 | 4.9 ª | 6.0ª |
| Cows, control semen | 324 | 9.1 ^b | 4.0 ^b |

Kaimio et al, 2013; (a-b, P<0.01)

| Embryos Produced with Sexed Sperm | | | | |
|--------------------------------------|----|-----------|-------------|--|
| | Ν | % Preg | % Female | |
| Sexed sperm | 42 | 47.6 | 90 | |
| Unsexed sperm | 68 | 52.9 | 53 | |
| Acta Sci Vet 2008;36 (Suppl 2):s433 | | | | |

IVF

- 1. Collect oocytes on farm
- 2. Mature oocytes during overnight shipment
- 3. Sort frozen unsexed semen
- 4. IVF and culture
- 5. Ship embryos to recipients or freeze

Sexed Fresh Semen

| Treatment | N | Non-Returnrate | | |
|-------------|--------|----------------|--|--|
| Sexed Semen | 51,712 | 69% | | |
| Control | 57,085 | 73% | | |
| Xu, 2014 | | | | |

| Sexed ULTRA | | | | |
|-------------------|------|--------|--|--|
| Method | Ν | Preg % | | |
| Standard | 3384 | 41.6% | | |
| Sexed ULTRA | 3546 | 46.1% | | |
| Vishiwonath, 2015 | | | | |
| | | | | |

| Timed Sexed Al Protocol | |
|--------------------------------------|--|
| April 8, pm: Insert CIDRs | |
| April 22, pm: Remove CIDRs | |
| May 8, 4 pm: PGF & Estrotect patches | |
| May 11, 9 am: Al if tripped patch | |
| GnRH if untripped patch | |
| May 12, 8 am: Al those given GnRH | |
| | |

Success Rates

- ♦ 46/56 tripped; 35 pregnant
 10/56 not tripped; 3 pregnant
 38/56 = 66%
- 18 not pregnant; 11 synchronized for repeat Al

Protocol for Repeats

| May 23, am: | Insert CIDRs |
|---------------|-------------------------|
| May 31, 9 am: | Remove CIDRs, |
| | + Estrotect patches |
| June 2, 4 pm: | Al those with patches |
| | tripped by 9 am, June 2 |
| June 3, 4 pm: | Al those with newly |
| | tripped patches |
| | |

Current Costs

 \$15/dose extra

 \$25/dose for custom
 sexing

Biggest cost – lower fertility

Extra semen collection

More sperm

- ◆ 2 to 4 million sperm/dose
- ◆ 5 percentage points fertility
- Doubles cost of semen

| | Cost Analysis |
|-------|---|
| | / straw for sexing accurately ╳ |
| | 6 pregnant ∽ |
| | XX 6 female ☆ |
| ♦ 90% | Survived to breeding age S64/ heifer |

Sexed Semen Fertility Higher than Conventional?

- Already discard dead/ dying sperm
- ♦ Already improves IVF
- Already same pregnancy rates sheep
- Already evaluating individual sperm
- Could select for positive sperm traits and against negative ones

Unsexed Sperm Could become obsolete

- One sex or the other always more valuable
- Fertility of sexed semen higher than unsexed?
- Sexing costs would need to be low

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- Farmer and rancher collaborators
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