Sexed Semen – The newest reproductive technology for the beef industry
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Although available commercially in the dairy industry for almost a decade, gender selected semen or sexed semen is one of the newest reproductive technologies available to the beef producer. The availability of sexed semen from beef bulls along with concerns about success of the technology at the ranch level has limited the use of sexed semen in purebred and commercial beef operation. Recent changes in semen availability combined with continuing research into improving success in beef operations make consideration of sexed semen a reality for some beef producers.

As sorting capacity increased, the number of beef bulls with gender selected semen available increase exponentially over the last four years. For the major US AI studs, the number of beef bulls with gender sorted semen available has increased from 0 to 70 from 2008 to 2011. Sexing Technologies, the major semen sexing company, lists over 45 sires with sexed semen in their catalog. In addition, it appears that custom collection of bulls with subsequent semen sexing will be available in the near future. While not an overwhelming selection of bull and genetics, there are now sufficient numbers of beef bulls with sexed semen to begin to meet the needs of the seedstock sector, and address the wanted traits for the commercial producer.

Sorting process increases cost and limits availability
Many different techniques for sorting Y and X bearing sperm were tried over 20 to 30 years. Only one, flow cytometery, is effective. This process consistently results in semen sorted with 90% of the desired sex. While the accuracy is great, the speed and yield of the process is slow and low. In addition, the equipment is extremely expensive and specially trained technicians are needed to assure sorting accuracy. Sexing of semen is a service provided by independent companies such as Sexing Technologies to bull studs (Select Sires, Genex, Accelerated Genetics, ABS Global, etc). Therefore, there is a fixed cost associated with sorting above the normal semen price.

To reduce costs and maximize availability of sexed semen. Gender selected semen is packaged in ¼ cc straws with 2.1 million cells per straw. In contrast, conventional semen is distributed in ½ cc straws containing 10 to 20 million cells per straw. Currently, sexed semen is available for $25 to $75 per straw.

Semen is actually sorted one sperm at a time and yield is low. Sperm with an X chromosome (which results in females) has slightly more DNA than Y-bearing sperm (which result in males). The sperm are treated with a fluorescent die that allows differentiation of the amount of DNA in the sperm. Sperm are then diluted and placed in droplets so each individual sperm is in a droplet. The droplets enter the detector where a laser is used to energized the dye

During the sorting process the machine identifies the sperm as X or Y, and it puts a charge on the drop that the sperm is placed in for sorting. An electrical field deflects the sperm towards the collection vessel. The sperm hit the fluid in the collection vessel at about 50 kph (30 mph). The
sorted sperm are the centrifuged and re-suspended. Although the sorting process is 90% accurate, approximately half the sperm cannot be sorted because they are damaged or the machine could not determine X or Y. Therefore, 50% of the ejaculate is discarded.

**Ranch level results with sexed semen**

*Artificial insemination.* Several large scale studies with use of sexed semen in dairy heifers indicate that pregnancy rates are 10% to 20% lower with sexed semen compared to conventional semen (Seidel et al., 1999; Seidel et al., 2000; DeJarnette et al., 2009). Using information from 39,763 inseminations with sexed semen and 53,718 inseminations with conventional semen, DeJarnette and coworkers (2009) reported heifer pregnancy rates of 45% and 56% for sexed and conventional semen, respectively. As typical with lactating dairy cows, pregnancy rates are considerably less in dairy cows than in dairy heifers. This led to the general recommendation that sexed semen should be use preferentially in heifers.

Data on pregnancy rates to sexed semen in beef cows and heifers are more limited. In general, technical services personnel from the major AI studs report at 10% to 15% reduction in pregnancy rates to sexed semen compared to conventional semen (Simmons, personal communication). The idea that using sexed semen heifers would be more successful than cows at the University of Idaho Nancy M. Cummings Center, we bred postpartum lactating beef cows with either sexed (n = 235) or conventional (n = 507) semen over the last three breeding seasons (Hall et al., 2010). Our pregnancy rates to sexed semen averaged 52% (range 48% to 58%) while pregnancy rates to conventional semen averaged 58% percent (range 52% to 69%). Most of the 235 cows we bred with sexed semen were bred using the CO-Synch + 5d CIDR fixed-time AI protocol (see figure). Other researchers had pregnancy rates ranging from 30% to 55%. Rhinehart and coworkers (2010) reported a 4% to 38% reduction in pregnancy rates when using sexed semen in heifers, and a 33% reduction in postpartum cows. In general, beef producers can expect a reduction in AI pregnancy rates of 10% to 20% with sexed semen compared to conventional semen.

The idea that using sexed semen in heifers would be more successful than in cows may not be correct in beef cattle. Over a large number of study members of the Beef Reproduction Task Force reported pregnancy rates of 65% using fixed-time AI systems with conventional semen (Lamb, 2010). In contrast, same group (and the industry in general) appears to show lower pregnancy rates and greater variability in pregnancy rates to fixed-time AI systems in heifers (Patterson, 2010). The one exception is the 14d CIDR-PG system which resulted in 65% AI pregnancy rates with conventional semen in heifers. One theory is that mature postpartum beef cows in good body condition and at least 50 days postpartum may be as fertile a female as we have on the ranch. One study tested the hypothesis that the fertility of sexed semen was not different between heifers and postpartum cows (Rhinehart et al., 2010). These researchers saw no difference in the performance of sexed semen in heifers vs. cows. However, the AI pregnancy rates to sexed semen were only in the 30 to 35% range. Another
theory is that just as in estrus synchronization and AI with conventional semen, we need to find the proper estrus synchronization system and timing of AI for sexed semen.

Calves produced from gender selected semen are normal with growth rates comparable to their herdmates that are products of conventional semen. Across two calving seasons at the University of Idaho, there was no difference in weaning weights of AI calves from sexed or conventional semen. Similarly, Tubman and coworkers (2004) found no difference in abortion rates, birth weight, calving ease, calf vigor, calf health, weaning weights, or mortality before weaning in over 1100 calves from sexed semen compared to 793 calves from conventional semen.

*Multiple Ovulation Embryo Transfer – MOET applications.* Using sexed semen in superovulated cows to produce embryos also results in decreases in reproductive efficiency. Researchers noted a 20% to 35% reduction in the number of transferable embryos when using sexed semen (Table 1). Most of this reduction is due to an increased number of unfertilized ova. The decrease in transferable embryos may be due in part to sperm number as a dose of 20 million sexed sperm resulted in similar numbers of transferable embryos to 40 million unsorted sperm. A few studies reported delay in development of embryos.

Table 1. Percentage of transferable embryos as affected by sorting and sperm dosage

<table>
<thead>
<tr>
<th>Experiment</th>
<th>% Transferable embryos</th>
<th>Semen dosage (million)</th>
<th>Heifers or cows</th>
</tr>
</thead>
<tbody>
<tr>
<td>Schenk et al., 2006*</td>
<td>18.6/16.5</td>
<td>43.5</td>
<td>10.0/2.0</td>
</tr>
<tr>
<td>Hayakawa et al., 2009</td>
<td>53.4</td>
<td>68.1</td>
<td>5.0</td>
</tr>
<tr>
<td>Peippo et al, 2009 (Expt. 1)</td>
<td>70.3</td>
<td>75.0</td>
<td>6.0 to 8.0</td>
</tr>
<tr>
<td>Peippo et al, 2009 (Expt. 2)*</td>
<td>53.9</td>
<td>65.5</td>
<td>6.0 to 8.0</td>
</tr>
<tr>
<td>Larson et al., 2010*</td>
<td>39.5</td>
<td>60.5</td>
<td>8.4</td>
</tr>
</tbody>
</table>

*Effect of semen type on % transferable embryos (P <0.05)

Pregnancy rates after transfer are similar among embryos produced with sexed or unsorted semen (Schenk et al., 2006; Hayakawa et al., 2009).

*In Vitro Fertilization – IVF.* In vitro fertilization drastically reduces the number of sorted sperm needed to fertilize an oocyte. As opposed to millions of sperm for AI or MOET procedures, IVF requires only 600-1500 sorted sperm to fertilize an oocyte (Xu et al., 2009). This greatly increases the potential sexed offspring from a sire.

Pregnancy rates from IVF cultured embryo fertilized with sexed semen range from 30% to 50%. While these pregnancy rates may seem low, they are offset by the sheer number of embryos that can be produced. For example, in a large commercial IVF embryo production system using Bos taurus, Bos indicus, and indicus-taurus cross cows, 5,407 embryo pick up procedure resulted in 16,924 transferable embryos (Pontes et al., 2010). Pregnancy rates were 36%-40% even after some of the embryos had been shipped over 1500 miles during culture. Embryos produced from sexed semen and IVF may have reduced cleavage or blastocyst rates (Zang et al., 2003; Blondin et al., 2009). However, improvements in IVF specifically for sexed semen fertilized embryos are rapidly bringing pregnancy rates of these embryos closer to pregnancy rates of embryos fertilized...
with conventional semen (Xu et al., 2009). In addition, these studies provide insight into potential solutions for decrease fertility of sexed semen in AI or MOET procedures.

**Why fertility is reduced, and what are the solutions.**
The sorting process, while effective, damages the sperm resulting in decreased motility and damage to the cell and acrosomal membranes (Carvalho et al., 2010). Gene expression and organelle development in the embryo may be impaired (Rath et al., 2009). While some damage occurs as a result for dye and exposure to laser light, most of the insult is a result of physical trauma during the sorting process (Garner, 2006). In contrast, sorting reduces the percentage of sperm with damaged DNA as sperm with damaged DNA are discarded (Gosalvez et al., 2011). Recent changes to the sorting process such as reduced sorting pressure and use of pulse lasers reduces damage to sperm and increases fertility (Schenk et al., 2009; Sharpe and Evans, 2009). Implementation of new semen preservation protocols (Sexcess®) during and after sorting may result in pregnancy rates near those of unsorted semen (Rath et al., 2009). However, further studies are needed to confirm the benefits of Sexcess® or similar procedures.

Increasing sperm number in the straw seems like a plausible solution to increase fertility. However, several studies clearly demonstrated only modest gains (5% to 7%) in pregnancy rates by doubling or tripling sperm numbers (DeJarnette et al., 2007; Schenk et al., 2009). Increasing sperm numbers from some bulls enhances pregnancy rates with sexed semen, but not with other bulls. Simply stated the damage done to sperm during sorting cannot be compensated for merely increase sperm number. Practically, reducing the number of available straws by 50% to gain 5% increase in pregnancy rate is a poor use of genetic potential.

There is considerable need for a greater understanding of the role of timing of AI relative to the onset of estrus when using gender sorted semen. This may be related to the initiation of the acrosome reaction which shortens the lifespan of the sperm. There is some indication that breeding later after estrus onset in dairy heifers improves pregnancy rate (Sa Filho et al., 2010). However, more studies are needed in beef cattle. Several universities are engaged in research on this topic. Perhaps alterations to timing of procedures in fixed-time AI programs would increase pregnancy rates to sexed semen. However, studies indicate that alterations in membranes and the acrosome may be bull specific. This would mean that timing of AI for semen from one bull may be different from several others.

Due to the fragile nature of sexed semen and the use of ¼ cc straws, semen handling when thawing, loading, and inseminating is critical. Errors in any part of the process are magnified with sexed semen. Beef producers need to work with their AI stud technical staff to learn the proper techniques for handling that companies sexed semen product. There are differences in packaging and handling requirements from company to company, so follow the recommendations specific to each product.

**Other considerations**

**Genetic diversity.** One of the biggest limitations for seedstock breeders is that a relatively small percentage of AI bulls are available as sexed semen. This severely limits the breeder’s options for genetic selection. If sexed semen is used heavily then the amount of genetic diversity may decrease due to the small number of sires available.
Another drawback may be the risk of overproduction of offspring from a particular bull. For example, if sons of Angus bull Joe Vandal are highly sought by commercial producers, and a great number of seedstock breeders use Y-sorted semen to make Joe Vandal sons then there could be an oversupply. In practice, there is no more danger of this type of over production due to the use of sexed semen than the current possibility of over production using conventional semen. The overproduction of milk by the dairy industry is often erroneously attributed to the use of sexed semen. Hutchison and Norman (2009) reviewed the use of sexed semen in the dairy industry. From 2006 to 2008, only 6.8% of all heifer inseminations and 0.9% of all cow inseminations were with sexed semen. In 2008, 14.2% of all heifer breedings and 2.1% were with sexed semen. Due to the higher number of inseminations per cow and greater number of lactating cows than heifers total unit of semen were almost equally split between cows and heifers. A little cowboy math (below) shows that due to sexed semen the dairy industry cranked out 6.9% more heifers than they would have if they used conventional semen. That is not exactly flooding the market with heifers.

### Impact of sexed semen on heifer production in the dairy industry in 2008-2009

\[
14.2\% \text{ of all heifer inseminations } \times 50\% \text{ pregnancy rate } + 2.1\% \text{ of all dairy cow inseminations } \times 25\% \text{ pregnancy rate } = 7.6\% \text{ sexed calves} \\
7.6\% \text{ sexed calves } \times 90\% \text{ desired sex} = 6.9\% \text{ more heifers}
\]

As each ranch situation is different, one of the best calculators for the cost and returns to using sexed semen can be found on the Genex website at: [http://genex.crinet.com/page2008/GenChoiceSexedSemen](http://genex.crinet.com/page2008/GenChoiceSexedSemen)

In my opinion, this calculator is rather conservative so it gives a realistic analysis if inputs are listed honestly.

**Management strategies when using sexed semen**

- Consider sexed semen in your herd only if AI pregnancy rates with conventional semen are consistently 60% or better.
- Use only in healthy cycling females in good body condition. AI companies suggest using sexed semen only in heifers; however, our research would indicate that cycling mature beef cows are also good candidates.
- Inseminate only animals observed in heat.
- If using fixed-time AI, make sure a high percentage of the animal were in heat before fixed-time AI. We believe this is the advantage to the CO-Synch + 5 day CIDR protocol.
- Use only experienced and proven AI technicians to inseminate cows or heifers.
- Be extremely careful with semen thawing and handling. Follow all of the AI company’s recommendations on semen handling to the letter.
• Keep up on current research on sexed semen as well as the latest recommendations from AI companies.

Conclusions
Rapid increase in the availability of sexed beef semen, and continuing improvements in the technology of producing and using sexed semen is creating an opportunity for leaders in beef reproduction and genetics. Breeders can expect acceptable results with sexed semen, but they need to be aware of the risks. Those using sexed semen should expect AI pregnancy rates that are 10% to 20% lower than the ranch’s normal AI pregnancy rates. In addition, the variability of success is high. Similar reductions in fertility are seen with MOET and IVF technologies.

At UI and other universities, we are still investigating the reasons for decreased pregnancy rates with sexed semen. Possibilities include greater sensitivity to handling during thawing and AI, early capacitation of sperm, and low sperm numbers per straw. Researchers and commercial suppliers of sexed semen are working together to decrease the impacts of sorting on sperm integrity and improve semen quality in sex-sorted semen.

References


