Extension
UNIVERSITY OF WISCONSIN–MADISON

Optimizing use of sexed semen in dairy herds

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Take home messages

- Use of sexed semen to inseminate Holstein and Jersey females in the U.S. has increased during the past 7 years in conjunction with an increase in reproductive performance of lactating dairy cows.
- In randomized, controlled experiments, sexed semen yields pregnancy per AI (P/AI) that are approximately 80% to 90% of that achieved with conventional semen; however, combining sexed semen with reproductive management strategies can optimize use of sexed semen in both heifers and lactating cows.
- Use of sexed semen in conjunction with a 5-d CIDR-Synch protocol for first insemination in nonlactating Holstein heifers tended to increase P/AI and decrease the cost per pregnancy compared with heifers inseminated to a detected estrus.
- Earlier induction of ovulation relative to timed AI so that insemination with sexed semen occurred closer to the time of ovulation decreased P/AI in primiparous Holstein cows. Thus, timing of AI within a Double-Ovsynch protocol should occur 16 hours after the final GnRH treatment when using sexed semen.
- Lactating Jersey cows submitted to a fertility program for first service had greater fertility than cows submitted to AI after a detected estrus when inseminated with sexed semen.

Introduction

During the past two decades, a reproduction revolution has occurred in the dairy industry. Based on data from nearly 20 million inseminations during >8 million lactations of >5 million cows in >23,000 U.S. herds, phenotypic performance for reproductive outcomes in U.S. Holstein and Jersey cows as well as genetic merit for daughter pregnancy rate reversed their historical declines and began to increase in 2002 (Norman et al., 2009; Fricke and Wiltbank, 2022). Although many factors are associated with the dramatic increase in reproductive performance (genetics, nutrition, management, etc.), development of fertility programs (Carvalho et al., 2018) and their adoption by dairy farmers (Caraviello et al., 2006) has driven much of this change (Fricke and Wiltbank, 2022). Fertility programs for first timed artificial insemination (**TAI**) as well as strategies for resynchronization of ovulation after nonpregnancy diagnosis have not only increased the AI service rate, but also have increased pregnancies per artificial insemination (**P/AI**) in high-producing Holstein cows by about 10-percentage points compared with AI to a detected estrus (Santos et al., 2017). Coupling fertility programs with new technologies for detection of estrus based on activity monitoring systems is an effective strategy for achieving high pregnancy rates in dairy herds (Fricke, 2017).

A consequence of increased reproductive performance was a concurrent increase in replacement heifer inventories which in turn drove down replacement heifer values to less than the cost

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required to rear them. Based on the UW-Extension Dairy Replacement ICPA Survey (Akins and Hagedorn, 2015), rearing costs of Holstein heifers from birth to calving averaged $2,100 from 1999 to 2015 and exceeded $2,500 in 2015. Under these economic conditions, the cost of raising heifers is $1,300 more than their market value and raising 100 extra heifers per year on a 1,000-cow dairy results in $130,000 in excess rearing costs. Further, the value of Holstein bull calves has also decreased because of a lack of demand by feedlots and slaughterhouses that desire animals that qualify to be marketed as Certified Angus Beef. Further, the value of dairy bull calves has also decreased because of increased liver abscess condemnations and decreased total carcass muscling compared with conventional beef cattle (Foraker et al., 2022).

To compensate for these changing economic conditions, dairy farmers have rapidly implemented dairy herd inventory management strategies to right-size replacement heifer inventories and maximize profit. One of the key reproductive technologies they have turned to is sexed semen. Use of sexed semen increases genetic progress in dairy herds through increased dam selection intensity (Khalajzadeh et al., 2012). Other strategies include use of genomic testing or pedigrees to identify genetically superior heifers and cows, use of sexed semen to inseminate genetically superior dairy heifers and lactating cows balanced for replacement needs (Weigel et al., 2012), and use of beef semen to inseminate low genetic merit heifers and cows to produce crossbred calves with increased value in the beef market (Ettema et al., 2017). This has led to a rapidly evolving trend to use sexed Holstein semen, conventional Holstein semen, and conventional beef semen to inseminate Holstein females in the U.S. (**Figure 1**). In 2020, 20% of Holstein females were inseminated using sexed semen, whereas 23% of inseminations used beef semen in Holstein females. The trends in Figure 1 are aggregated among many farms; however, some dairy farms no longer use conventional Holstein semen and now exclusively use sexed Holstein semen and conventional beef semen to inseminate Holstein females to maximize efficiency and profit.

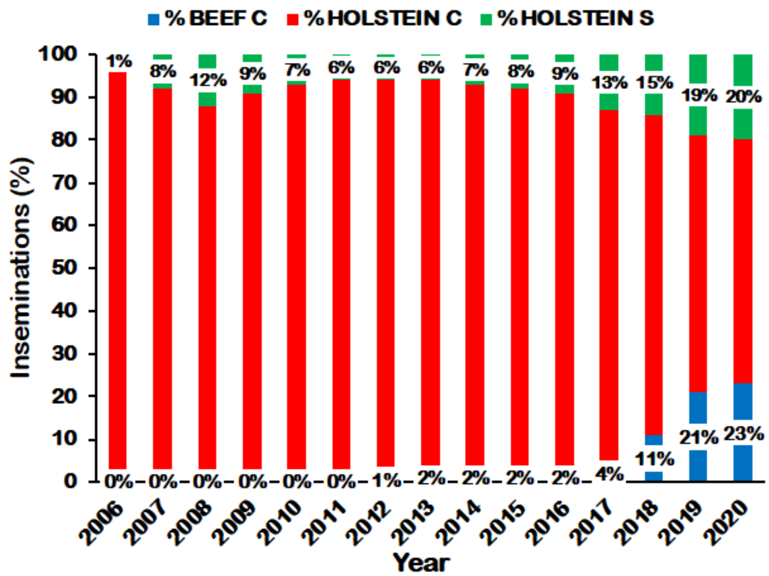

Figure 1. Proportion of inseminations using conventional beef (%BEEF C), conventional Holstein (% HOLSTEIN C), and sexed Holstein (% HOLSTEIN S) semen to inseminate Holstein females (heifers and lactating cows) in the U.S. from 2006 to 2020. Data from AgSource Dairy, Madison, WI.

The purpose of this paper is to overview several recent studies from our laboratory that have investigated management strategies to improve the fertility of sexed semen in both nonlactating heifers and lactating dairy cows.

Use of Sexed Semen to Inseminate Nulliparous Dairy Heifers at First Service

The rate at which heifers become pregnant is determined by an interaction between AI service rate and conception rate (P/AI). Service rate can be increased by using synchronized breeding programs tailored specifically for dairy heifers. In contrast to lactating cows, dairy heifers respond poorly to synchronization protocols based solely on GnRH and $PGF_{2\alpha}$ such as Ovsynch (Pursley et al., 1997). This is due to differences in circulating progesterone concentrations (Sartori et al., 2004) and an increased rate of follicular wave turnover in heifers (Bisinotto and Santos, 2011) which decreases protocol synchrony (Lima et al., 2013). Inclusion of a controlled intravaginal progesterone insert (EAZI-Breed **CIDR**, Zoetis, Madison, NJ) during the protocol prevents heifers from displaying estrus until CIDR insert removal thereby increasing synchrony to the protocol (Rivera et al., 2005). The 5-d CIDR-Synch protocol is recommended for use in dairy heifers by the Dairy Cattle Reproduction Council (Lima et al., 2013).

The mean conception rate for U.S. Holstein heifers was 57% based on 537,938 inseminations of 362,512 heifers in 2,668 herds from 41 states (Kuhn et al., 2006). The 5-d CIDR-Synch protocol for TAI yields conception rates between 50 to 60% when using conventional semen (Rabaglino et al., 2010; Lima et al., 2011) and increases the AI service rate thereby decreasing days to first AI and days to pregnancy compared with heifers receiving AI to a detected estrus (Silva et al., 2015;

Masello et al., 2019). The decreased days to first AI and pregnancy using the 5-d CIDR-Synch protocol in turn decreases days on feed (Silva et al., 2015), which is the greatest cost associated with raising dairy replacements (Akins and Hagedorn 2015; Karszes and Hill, 2020). A caveat with the 5-d CIDR-Synch protocol is that 27% to 33% of heifers display estrus >24 hours before scheduled TAI (Masello et al., 2019; Silva et al., 2015). This makes detection of estrus during the 5-d CIDR-Synch protocol a requirement to achieve acceptable conception rates.

A major limitation of sexed semen is the decrease in fertility compared with conventional semen, and this decrease in fertility may be greater when heifers are inseminated to estrus using sexed semen. In a recent experiment from our laboratory (Niles et al., 2019), the decrease in fertility using sexed vs. conventional semen to inseminate Holstein heifers once-daily based on rubbed tail chalk was 54% of that of conventional semen, and the difference in P/AI between conventional and sexed semen exceeded 30-percentage points. In a post-hoc analysis that included a small number of heifers, the decrease in fertility using sexed semen to inseminate heifers once-daily based on rubbed tail chalk exceeded 30-percentage points, whereas the decrease in fertility using sexed semen to inseminate heifers to TAI after a 5-Day CIDR-Synch protocol was less than 10-percentage points (Silva et al., 2015). Thus, heifers submitted for TAI using sexed semen after a 5-day CIDR-Synch protocol had a 20-percentage point increase in fertility compared with heifers receiving AI using sexed semen once-daily based on rubbed tail chalk. Despite the advantage in fertility of using sexed semen in conjunction with a TAI protocol, 69% of dairy heifers in the U.S. are inseminated at first service based on natural or induced estrus (NAHMS, 2014).

We conducted a field trial in collaboration with three commercial dairy farms in southern Wisconsin to compare reproductive management programs for submission of Holstein heifers for first insemination with sexed semen (Lauber et al., 2021). The experimental design is depicted in **Figure 2**. Briefly, nulliparous Holstein heifers (n = 736) were randomized within each of the three farms to three treatments for first AI with sexed semen (ABS Sexcel Sexed Genetics, ABS Global, DeForest, WI): 1) **CIDR5**; 2) **CIDR6**; and 3) **EDAI** ($PGF_{2\alpha}$ on day 0 was followed by once daily detection of estrus [visual detection of tail-chalk removal and other signs] and AI). Our control treatment using a single $PGF_{2\alpha}$ treatment administered on day 0 is representative of how many farms manage dairy heifers for first insemination. The comparison between the CIDR5 and CIDR6 treatments was based on an experiment using conventional semen in which delaying progesterone insert removal decreased early expression of estrus before scheduled TAI and resulting in no difference in P/AI (Lauber et al., 2021).

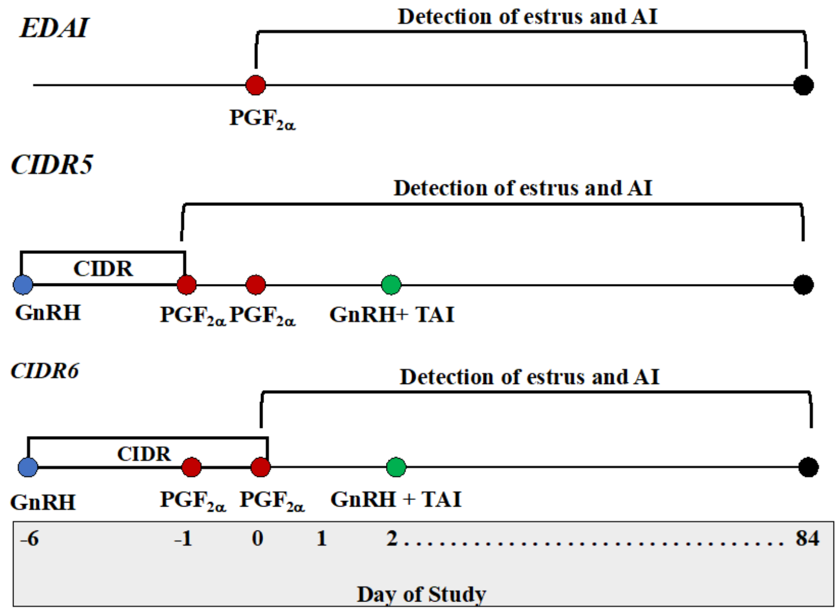

Figure 2. Schematic diagram of reproductive protocols. Holstein heifers were randomized to receive TAI after a 5-d CIDR-Synch protocol (CIDR5), a 6-d CIDR-Synch protocol (CIDR6), or treatment with $PGF_{2\alpha}$ on d 0 and once-daily AI after estrus (EDAI). Adapted from Lauber et al., 2021

Our hypotheses were that 1) delayed CIDR removal would decrease early expression of estrus before scheduled TAI while maintaining P/AI with sexed semen; 2) submission of heifers to TAI protocols for first insemination will increase P/AI and decrease days to AI and pregnancy with sexed semen compared with EDAI; and 3) submission of heifers to TAI protocols for first insemination will decrease the cost per pregnancy compared with EDAI because of decreased days on feed. Delaying CIDR removal decreased early expression of estrus before scheduled TAI (0.004 vs. 27.8%); however, CIDR5 heifers tended to have more P/AI at 35 ± 5 and 64 ± 5 days after AI than CIDR6 and EDAI heifers, respectively (**Table 1**). Overall, CIDR5 and CIDR6 heifers had fewer days to first AI and pregnancy than EDAI heifers resulting in less feed costs than EDAI heifers because of fewer days on feed until pregnancy.

Table 1. Effect of treatment on pregnancies per AI (P/AI), return to estrus, and pregnancy loss in nulliparous Holstein heifers inseminated with sexed semen at first insemination. Adapted from Lauber et al., 2021.

	Treatment[1]			
Item	EDAI	CIDR5	CIDR6	*P*-value
P/AI 35 ± 5 d, % (n)	46^{A} (227)	53^{B} (255)	45^{A} (254)	0.09
EDAI[2]	. .	62^{**} (71)	$0^{‡}$ (1)	
Scheduled TAI[3]	. . .	50^{*} (184)	$46^{†}$ (253)	
P/AI 64 ± 5 d, % (n)	45^{A} (227)	52^{B} (255)	45^{A} (254)	0.10
EDAI[2]	. . .	$62^{‡}$ (71)	$0^{‡}$ (1)	
Scheduled TAI[3]	. . .	$48^{†}$ (184)	$45^{†}$ (253)	
Return to estrus[4] (d)	22.01 ± 0.63	21.19 ± 0.38	21.72 ± 0.39	0.51
Pregnancy loss, % (n)	4 (106)	2 (135)	1 (114)	0.20

$^{A\text{-}B}$ Within a row, percentages with different superscript uppercase letters tend ($P < 0.10$) to differ.

$^{*\text{-}**}$ Within a column, percentages with different superscript tend ($P < 0.10$) to differ.

$^{†\text{-}‡}$ Within a column, percentages with different superscript differ ($P < 0.05$).

[1] Nulliparous Holstein heifers were submitted for first insemination with sexed semen to either $PGF_{2\alpha}$ on day 0 with once-daily detection of estrus and AI (EDAI) or a CIDR-Synch protocol with CIDR removal on either day -1 (CIDR5) or day 0 (CIDR6).

[2] CIDR-Synch heifers detected in estrus ≥ 24 hours and inseminated before scheduled TAI.

[3] Heifers submitted to CIDR-Synch protocols and inseminated at scheduled TAI.

[4] Nonpregnant heifers that returned to estrus and were inseminated before pregnancy diagnosis (Mean ± SEM).

A partial budget analysis based on actual farm costs (US$) was used to determine the cost per pregnancy for heifers in each treatment during the 84-d breeding period. Costs used to calculate the cost per pregnancy for each heifer included: hormonal treatment, detection of estrus, semen and AI, pregnancy diagnosis, and feed costs. Labor associated with treatments and detection of estrus was $13/hour, and hormones for synchronization of ovulation or induction of estrus were $1.60, $2.06, and $12.27 per GnRH treatment, $PGF_{2\alpha}$ treatment, and CIDR insert, respectively. We then conducted a sensitivity analysis to determine the effect of varying feed costs on total costs per pregnancy to reflect differences in costs per pregnancy because of market conditions or geographical regions on these three strategies for submission of heifers to first insemination using sexed semen (**Figure 3**). Despite greater up-front hormonal treatment costs for CIDR5 heifers, costs per pregnancy were less for CIDR5 than for EDAI heifers when the cost of feed per day was greater than $1.50 per heifer per day (**Figure 3**). In conclusion, although delaying CIDR removal by 24 hours in a 5-CIDR-Synch protocol suppressed early expression of estrus before TAI, delaying CIDR removal by 24 hours tended to decrease P/AI for heifers inseminated with sexed semen. Further, submission of heifers to a 5-d CIDR-Synch protocol for first AI tended to increase P/AI and decrease the cost per pregnancy compared with EDAI heifers.

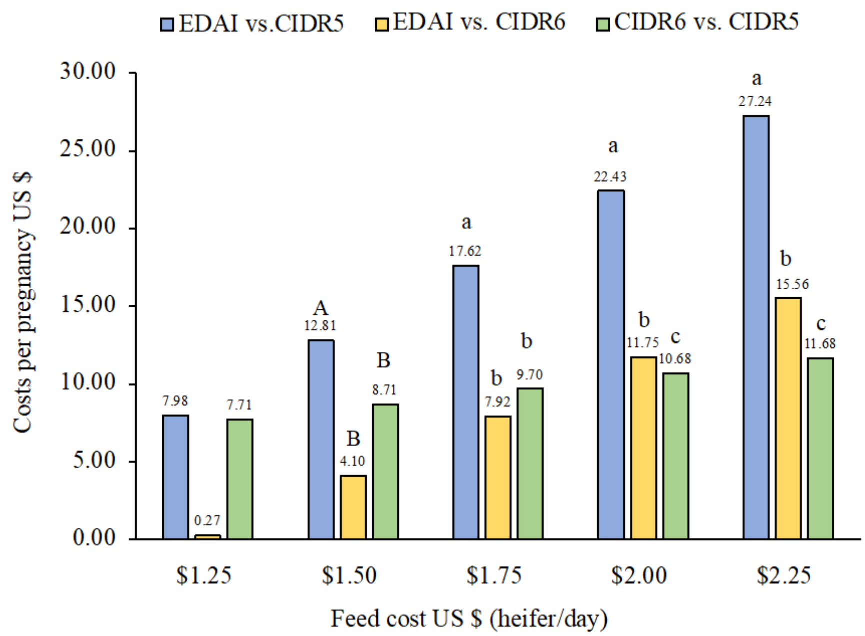

Figure 3. Sensitivity analysis of feed cost (US$ per heifer/day) on cost per pregnancy. All other input costs in the analysis were held constant among treatments. Means with different lowercase letters (a-c) differ ($P < 0.05$), whereas means with different capital letters (A-B) tended to differ ($P < 0.10$). Adapted from Lauber et al., 2021.

Effect of Timing of AI Relative to Ovulation using Sexed Semen in Primiparous Holstein cows

Despite the advances in fertility of lactating dairy cows resulting from fertility programs (Carvalho et al., 2018), fertility of sexed semen remains less than that of conventional semen (Karakaya-Bilen et al., 2019). Pregnancy outcomes of sexed semen are 80 to 90% that of conventional semen resulting in a 5- to 10-percentage point decrease in P/AI (Seidel et al., 1999; DeJarnette et al., 2009; Drake et al., 2020). One reason for the decrease in fertility is damage to sperm during the sexing and cryopreservation processes (Seidel, 2014). It is thought that this damage causes sexed sperm to capacitate at a faster rate than conventional sperm thereby decreasing fertility. A new idea is that timing of AI relative to the onset of estrus may optimally occur closer to the time of ovulation (Bombardelli et al., 2016; Nebel 2018). Capacitation is a biochemical process sperm must undergo to fertilize an oocyte (Austin, 1967). Membrane damage is a compensable trait, however, empirical data that validate the idea that sexed sperm undergo early capacitation are not available. Compensable traits can be overcome by increasing sperm numbers per dose which is critical for aspects of sperm transport and function in the female reproductive tract including progressive motility, acrosome integrity, and cell membrane integrity (Saacke, 2008). Increased fertility resulting from greater sperm numbers reaches a plateau at approximately 4.0 x 10^6 sperm (den Daas et al, 1998; Fearon and Wegener 2000). The decreased fertility associated with sexed semen, however, is not overcome as sperm concentration is increased (DeJarnette et al., 2008; 2010; 2011). Thus, fertility of sexed semen is a uncompensable rather than a compensable trait.

One of the first experiments to evaluate use of sexed semen in lactating cows used an activity monitoring system in Jersey cows to time AI based on increased activity (Bombardelli et al., 2016). Overall, P/AI using sexed semen was greatest for Jersey cows inseminated between 23 and 41 hours after the onset of activity (Bombardelli et al., 2016), which is later than the optimal timing for conventional semen of 4 to 12 hours after the onset of standing activity established using a radiotelemetric system for determining standing activity (Dransfield et al., 1998) or 8 to 16 hours after the onset of activity using an activity monitoring system (Stevenson et al., 2014). The observation that inseminating cows later relative to the onset of estrus when using sexed semen was corroborated in a recent field study (Nebel, 2018). Thus, an idea in the dairy industry today is that timing of AI using sexed semen should occur closer to the time of ovulation than conventional semen. We have reported a positive relationship between the interval from onset of activity associated with behavioral estrus using an activity monitoring system to ovulation and milk production near the time of estrus (Valenza et al., 2012). Thus, inseminating high-producing cows later using sexed semen may be optimal for cows inseminated to estrus because ovulation occurs later relative to the onset of estrus in high-producing cows as milk production near the time of estrus increases. A critical knowledge gap is that the optimal timing of AI using sexed semen has not been established when the interval from timing of AI to ovulation is controlled using a fertility program at first service.

To address this question, we conducted a field trial in collaboration with three dairy herds in three different U.S. states to determine the effect of inseminating primiparous Holstein cows closer to the time of ovulation (Lauber et al., 2020). Our objective was to determine the effect on fertility of increasing the interval from induction of ovulation to TAI thereby decreasing the interval from TAI to ovulation using sexed semen within a synchronized breeding program. Our hypothesis was that induction of ovulation earlier relative to TAI would increase P/AI. Primiparous Holstein cows from three commercial dairy farms in the U.S. were submitted to a Double-Ovsynch protocol for first service that included a second $PGF_{2\alpha}$ treatment 24 hours after the first in the breeding-Ovsynch portion of the protocol as described by Brusveen et al. (2009). The last GnRH treatment (**G2**) was varied between treatments and TAI. To vary the interval between G2 and TAI, cows were randomized to two treatments to receive G2 either 16 (**G2-16**; n = 373) or 24 (**G2-24**; n = 357) hours before TAI, which was fixed at 48 hours after the second $PGF_{2\alpha}$ treatment of the breeding-Ovsynch portion of the Double-Ovsynch protocol. All cows were inseminated with sexed semen (ABS Sexcel Sexed Genetics, ABS Global, DeForest, WI), and each herd used sires of their choosing which were randomly allocated between treatments. Pregnancy diagnosis was conducted by herd veterinarians using transrectal ultrasonography.

Contrary to our hypothesis, G2-24 cows had fewer P/AI than G2-16 cows at 34 ± 3 days (44% vs. 50%) and 80 ± 17 days (41% vs. 48%) after TAI (**Table 2**). Pregnancy loss and fetal sex ratio did not differ between treatments. In conclusion and in disagreement with our hypothesis, induction of ovulation earlier relative to TAI after a Double-Ovsynch protocol decreased P/AI in primiparous Holstein cows, whereas pregnancy loss and proportion of female fetuses did not differ between treatments. Delaying insemination based on detection of estrus may increase P/AI because of the longer interval from the onset of estrus to ovulation and shorter duration of estrus in high-producing lactating dairy cows but not in cows submitted to TAI after synchronization of ovulation in which timing of AI relative to ovulation is precisely controlled. Based on these data, inseminating cows with sexed semen 16 hours after G2 yielded more P/AI than when cows were inseminated 24 hours after G2. The decreased P/AI observed by increasing the interval from induction of ovulation to TAI may be attributable to decreased time for sperm transport and capacitation and/or differences in size of the ovulatory follicle and the hormonal milieu at critical time points near the end of the synchronization protocol.

Table 2. Effect of altering timing of induction of ovulation (G2) relative to timed artificial insemination (TAI) with sexed semen on pregnancies per artificial insemination (P/AI), pregnancy loss, and fetal sex in primiparous Holstein cows. Adapted from Lauber et al., 2020.

	Treatment[1]		
Item	G2-16	G2-24	*P*-value
P/AI 34 ± 3 d, % (n)	50 (373)	44 (357)	0.05
P/AI 80 ± 17 d, % (n)	48 (370)	41 (355)	0.03
Pregnancy loss, % (n)	5 (184)	6 (154)	0.70
Female fetuses,[2] % (n)	92 (126)	90 (100)	0.64

[1]Cows were submitted to a Double-Ovsynch protocol for first TAI with sexed semen and randomized to receive G2 16 hours before TAI (G2-16) or 24 hours before TAI (G2-24).
[2] Fetal sex was diagnosed by herd veterinarians 71 ± 4 days after TAI using transrectal ultrasonography on two of the three farms.

Effect of First Service Insemination Strategy on Fertility of Jersey Cows Inseminated with Sexed Semen

A review of studies that compared P/AI of cows inseminated after a detected estrus to P/AI of cows receiving TAI after submission to Presynch-Ovsynch or Double-Ovsynch protocols supports that fertility programs yield more P/AI than when cows are inseminated after a detected estrus (Fricke and Wiltbank, 2022). We tested the idea that fertility programs and TAI can yield greater fertility than AI to estrus at first insemination using conventional semen in high-producing Holstein cows (Santos et al., 2017). Lactating Holstein cows (n =578) were assigned randomly to receive TAI after a Double-Ovsynch protocol that included a second $PGF_{2\alpha}$ treatment or submission to AI if detected in estrus after receiving a GnRH treatment followed by 2 $PGF_{2\alpha}$ treatments administered 7 and 21 days later. An important aspect of the experimental design was that treatments were administered so that cows were inseminated at a similar days in milk (DIM) across both treatments (77 ± 3 DIM) to control DIM at insemination as a confounding factor. Overall, cows submitted to Double-Ovsynch had more P/AI than cows inseminated after a synchronized estrus at both 33 ± 3 days (49.0% vs. 38.6%) and 63 ± 3 days (44.6% vs. 36.4%) after insemination (Santos et al., 2017). Thus, the relative increase in P/AI was more than 25% (10.4/38.6 = 26.9%). Synchronization rate to the hormonal protocols was 85.3%, which did not differ between treatments; however, cows determined to have synchronized correctly to the Double-Ovsynch protocol maintained the 10-percentage point advantage in P/AI 33 days after insemination compared with cows inseminated at the correct time relative to estrus (54.7% vs. 44.5%). We concluded based on this analysis that, because the proportion of synchronized cows did not differ between treatments, cows submitted to a Double-Ovsynch protocol had more P/AI than cows inseminated after a synchronized estrus because of an intrinsic increase in fertility rather than differences in synchronization rates to the protocols.

To address this question with sexed semen, we conducted a field trial to compare first insemination strategy using TAI vs. AI to estrus in lactating Jersey cows inseminated with sexed semen (Lauber et al., 2022). Our hypothesis was that lactating Jersey cows submitted to a Double-Ovsynch protocol for TAI at 1st service would have more P/AI than cows inseminated after synchronization of estrus. Lactating Jersey cows (n = 742) from a commercial dairy farm were randomized by ear-tag number (odd vs. even) within parity for submission to first service after a Double-Ovsynch (**DO**) protocol or a protocol for synchronization of estrus with twice-daily detection of estrus (**EDAI**) using sexed Jersey semen (**Figure 4**).

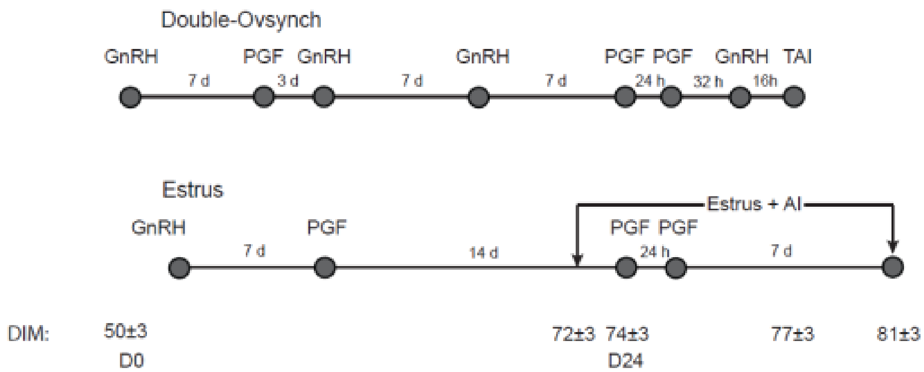

Figure 4. Schematic diagram of treatments and experimental endpoints. Each week, lactating Holstein cows at 50 ± 3 DIM (d 0) were stratified by parity (primiparous vs. multiparous) and randomly assigned to receive their first insemination using sexed Jersey semen as a timed artificial insemination after a Double-Ovsynch (DO) protocol or as an artificial insemination to a detected estrus after a protocol for synchronization of estrus (Estrus). DIM = days in milk; PGF = prostaglandin $F_{2\alpha}$; GnRH = gonadorelin releasing hormone; AI = artificial insemination; TAI = timed artificial insemination. Adapted from Lauber et al., 2022.

Among all cows, mean (± SEM) days from $PGF_{2\alpha}$ (d 24) to AI was greater (P <0.001) for EDAI than for DO cows (4.0 ± 0.11 vs. 3.0 ± 0.01), whereas the proportion of cows inseminated was greater (P < 0.001) for DO than for EDAI cows (100% vs. 75%). Thus, 75% of cows in the EDAI treatment were detected in estrus and inseminated, whereas 25% of cows were not detected in estrus and were submitted to TAI after an Ovsynch protocol for first service. Jersey cows submitted to a Double-Ovsynch protocol and inseminated with sexed semen had more P/AI at 31 ± 2 (52% vs. 43%) or 61 ± 4 (49% vs. 40%) days after AI than EDAI + TAI cows (**Table 3**). These results support our hypothesis that lactating Jersey cows submitted to a Double-Ovsynch protocol for TAI at first service had more P/AI than cows inseminated after synchronization of estrus.

Table 3. Effect of method of submission for first AI on pregnancies per AI (P/AI) of lactating Jersey cows inseminated with sexed Jersey semen. Adapted from Lauber et al., 2022.

	Treatment[1]		
Item	EDAI + TAI	Double-Ovsynch	*P*-value
P/AI 34 ± 3 d, % (n)	43 (344)	52 (398)	<0.01
P/AI 62 ± 4 d, % (n)	40 (341)	49 (398)	<0.01

[1]Cows were submitted to a protocol for synchronization of estrus followed by submission to an Ovsynch protocol for cows not detected in estrus (EDAI + TAI) or to a Double-Ovsynch protocol for first TAI with sexed Jersey semen.

CONCLUSION

A consequence of increased reproductive performance was a concurrent increase in replacement heifer inventories which in turn drove down replacement heifer values to less than the cost required to rear them. In response to these economic conditions, use of sexed semen to insemination Holstein and Jersey females has dramatically increased during the past 7 years and will likely continue to increase in the future. A major limitation of sexed semen is the decrease in fertility compared with conventional semen. Three recent studies from our laboratory have investigated how to increase pregnancy outcomes using sexed semen in conjunction with various management practices. Use of sexed semen in conjunction with a 5-d CIDR-Synch protocol for first insemination in nonlactating Holstein heifers tended to increase P/AI and decrease the cost per pregnancy compared with heifers inseminated to a detected estrus. Induction of ovulation earlier relative to timed AI so that insemination with sexed semen occurred closer to the time of ovulation decreased P/AI in primiparous Holstein cows. Thus, timing of AI within a Double-Ovsynch protocol should occur 16 hours after the final GnRH treatment when using sexed semen. Finally, lactating Jersey cows submitted to a fertility program for first service had greater fertility than cows submitted to AI after a detected estrus when inseminated with sexed semen. Future studies should be aimed at a deeper understanding of the limitations of sexed semen regarding fertility and how to overcome them.

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