

Dairy Pipeline

School of Animal Sciences

Volume 46, No. 6 ● July/August 2025

Optimizing Double-Ovsynch: Timing Is Everything

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The introduction of the first Ovsynch program in 1995 marked a pivotal shift in reproductive management for dairy cattle. Since then, these protocols have evolved significantly, with the **Double-Ovsynch program** being one of the most popular timed artificial insemination (TAI) programs in modern dairy herds (Figure 1). As its name suggests, the Double-Ovsynch program layers two Ovsynch sequences to optimize fertility outcomes, particularly in high-producing cows preparing for their first postpartum insemination.

This protocol provides multiple reproductive benefits: it induces estrus in anovulatory cows, promotes the turnover of suboptimal follicles, and fosters the development of healthy ovulatory follicles. A less discussed, yet equally critical, advantage is the ability of this program to generate multiple corpora lutea (CLs), the ovarian structures that produce progesterone. Elevated progesterone levels support improved follicular development, enhance oocyte quality, and create a more receptive uterine environment, all of which boost conception success.

Timing Is Critical—But Often Mismanaged

Anyone familiar with TAI protocols understands that strict adherence to the injection schedule is vital to success. However, one part of the Double-Ovsynch program that often sees inconsistent compliance is the timing of AI relative to the final GnRH injection. Producers frequently adjust this TAI event to fit milking schedules, and the extremes of this seem common, where cows may be inseminated at the time of the last GnRH (16 hours too early) or wait until the following morning (8 hours too late).

So, does this really matter? Based on our understanding of bovine reproductive physiology, the timing of AI is crucial. Inseminating too early reduces fertility because sperm have a limited lifespan within the uterus and oviduct. While the oviduct provides a temporary sanctuary for sperm, this reservoir begins to decline 24–30 hours postinsemination.

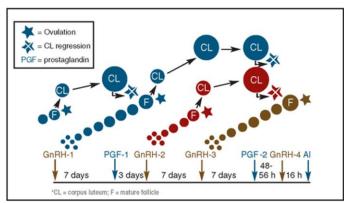


Figure 1. The ovarian reaction to Double Ovsynch

In contrast, inseminating too late jeopardizes oocyte viability. Oocytes can still be fertilized for extended periods after ovulation, but the competency of the embryo produced from these late fertilizations is often compromised. Expect to see a significant drop in embryo competency for the oocyte not fertilized within 6 six hours after ovulation. Ovulation occurs 24–32 hours after the final GnRH injection, and the sperm require 8-12 hours to travel to the oviduct and undergo capacitation, so this 16-hour period is the ideal time to provide capacitated sperm to a recently ovulated oocyte.

What the Research Says

A study published recently in the *Journal of Dairy Science* analyzed over 20,000 inseminations from lactating Holstein cows (Santos et al., 2025). The findings are clear: deviating too far from the recommended 16-hour post-GnRH insemination window significantly reduces fertility. Specifically, conception rates dropped by 22% when AI was performed at the time of the final GnRH injection, rather than 16 hours later.

However, there is some good news. Research indicates a degree of flexibility exists in this timing. Two follow-up studies discussed in this same manuscript found that optimal conception rates could be maintained when inseminating between 13 and 23 hours after the final GnRH injection. So, while the exact 16-hour mark isn't rigid, AI should not be performed at the time of GnRH administration or delayed for 24 hours, or beyond.

Special Considerations when using Sexed Semen

Timing of insemination is even more critical when using sexed semen. The current recommendation is to delay AI to better align with the shorter lifespan and fertilization window of sexed sperm. For estrusbased breeding, inseminate at 18–20 hours postestrus instead of the traditional 12 hours. For TAI protocols without heat detection, inseminate 22 hours post-GnRH instead of 16 hours.

Still, the same flexibility applies when using the Double-Ovsynch program. The 13-23-hour post-

GnRH window remains effective with sexed semen. But again, conception rates decline rapidly if AI is postponed beyond 23 hours.

Takeaways for Manipulating the Timing of TAI in a Double-Ovsynch Program:

- 1. **Avoid inseminating too early or too late.** AI at the time of the final GnRH injection or 24 hours or more after the final GnRH is strongly discouraged.
- 2. Use the 13-23 hour post-GnRH window. While 16 hours is ideal, this range maintains high conception rates.
- 3. **Adjust for sexed semen with care.** Aim for insemination 13-23 hours post-GnRH, with a preference toward the later end of the window.

With four decades of research behind us, Ovsynch strategies continue to evolve, offering both flexibility and precision. Adhering to these refined timing recommendations will allow dairy producers to maintain efficient, high-fertility TAI programs where reproductive success can be achieved while balancing the practical demands of herd management.

In Utero Heat Stress Limits Milk Yield for Generations

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As climate change continues to raise global temperatures, the impact of heat stress on livestock has become a growing concern. The effects of heat stress on lactating dairy cows have been well established. It increases the chance of transition diseases for fresh cows and lowers dry matter intake, milk production, immune function, and fertility in all stages of lactation. However, the impacts of heat stress during late gestation have not received as much attention as the effects of heat stress on lactating cows, despite being a critical issue for dairy farmers. The consequences of heat stress are far-reaching, affecting not only the cows themselves but also their offspring and subsequent generations. This article highlights the impact of

late-gestation heat stress on dairy cows and their progeny.

The last two months of gestation coincide with the dry period, a time when dairy cows are not lactating, ranging from 45-60 days. This period is imperative for mammary involution, as their energy shifts from milk production to fetal development and preparing for the next lactation. This period is also characterized by accelerated fetal growth, as the fetus accumulates approximately 60% of its birth weight during the last two months of gestation (Bauman and Currie, 1980). The foundation for mammary function is laid in utero with early mammary development, so exposure to stressors prenatally could affect immediate and long-term mammary morphology and synthetic capacity, reducing lifetime production and economic profit (Larsen and Laporta, 2024).

The economic implications of not cooling dry dairy cows are substantial. Heat stress during the dry period can lead to decreased milk production in subsequent lactations, increased health issues, and higher culling rates. Studies estimate that the dairy industry loses approximately \$810 million annually to the effects of heat stress on dry cows and its impact on progeny for multiple generations (Ferreira et al., 2016). These losses stem from reduced milk yield for both the dam exposed to heat stress during the dry period and future progeny following exposure to heat stress in-utero (Casarotto, 2021). It also involves increased veterinary costs from reduced immune function and increased transition diseases. By investing in cooling systems for dry cows, such as large shade structures, fans, and soakers, dairy farmers can mitigate these economic losses and improve the overall health and productivity of their herds (Laporta and Skibiel, 2024).

Heat stress affects lifetime production through many facets, beginning with exposure to heat during late gestation, which alters the placental structure and its function in multiparous dairy cows. This changes the placenta's ability to support the developing fetus. The placenta is crucial for fetal development, facilitating nutrient transfer, heat dissipation, and gas exchange. Dry cows that experience heat stress have shorter gestation lengths and reduced calf birth weights (Casarotto et al., 2025). The placenta itself has a reduced total weight and cotyledon number in

heat-stressed cows; however, placenta efficiency is increased. Cooled cows have healthier placentas overall, while cows exposed to heat stress must work harder to adapt and compensate for these changes to guarantee the success of the pregnancy. This leads to impaired body and mammary development and reduced lifetime production for the calf in-utero, setting the stage for long-term health and productivity issues (Casarotto et al., 2025).

Impacts on mammary development are not limited to the daughter exposed to heat stress in utero but continue to the next generation (granddaughter). Carryover effects of maternal late-gestation heat stress have been shown to reduce body growth and mammary gland development of daughters and granddaughters (Larsen and Laporta, 2024). Through ultrasound and mammary biopsy observations, the parenchyma and fat pad area are greatly reduced in heat stress progeny, when compared to progeny that were cooled during the dry period and late gestation. The parenchyma and fat pad give rise to the ductal system, lumen, lobules, and alveoli, and adipose and connective tissue. This reduction in mammary gland development can lead to slower mammary growth rates, smaller milk synthetic capacity, and decreased milk production. By not cooling dry cows, the overall productivity and profitability of dairy operations are greatly affected (Larsen and Laporta, 2024).

As suggested through mammary development observations, there is a negative effect of lategestation heat stress on the lifetime performance of daughters and granddaughters. Data that was analyzed from multiple farms and generations of dairy cows, found that heat-stressed progeny during late gestation significantly impairs survivability and lifetime milk production (Laporta et al., 2020). Daughters of heat-stressed dry cows were more likely to be culled across three lactations when compared to cooled daughters, but this was not as significant in the grand-daughter observations. Milk yield was analyzed for daughters and granddaughters across three lactations, with heatstressed progeny consistently producing less than their cooled counterparts, across both generations (Laporta et al., 2020). It has been known that heat stress in dry cows affects milk production in the subsequent lactation, but now it also shows the same effect on multiple generations following heat stress exposure during late gestation.

There is a clear picture of the detrimental effects of late-gestation heat stress on dairy cows and their offspring. From altering placental structure and function to impairing mammary gland development and reducing lifetime performance, the consequences of heat stress are far-reaching and multi-generational. Cooling dry cows will increase milk yield in their subsequent lactation and improve the lifetime production of daughters and granddaughters. By doing so, the dairy industry can ensure the sustainability and profitability of their operations for generations to come.

Upcoming Events

July 2025

Southeast Dairy Business Innovation Initiative Grants Opening

- Dairy Business Planning Grant
- Specialty Processing Equipment Grant

July 26, 2025

Dairy fitting and showing workshop Rockingham County Fairgrounds

July 31, 2025

Dairy Youth Field Day

August 2, 2025

VA State Dairy Shows

August 7, 2025

Virginia Ag Expo

August 12, 2025

Rockingham County Fair Dairy Show

September 14-17, 2025

All-American Dairy Show

September 26, 2025

VA State Fair Jr. Dairyman's Contest

September 27, 2025

VA State Fair Dairy Show

October 13, 2025 (Monday)

Hokie Cow Classic – Save the Date

If you are a person with a disability and require any auxiliary aids, services, or other accommodations for any Extension event, please discuss your accommodation needs with the Extension staff at your local Extension office at least 1 week prior to the event.

Additional Notes:

- The dairy extension group is working with VDH to assist in distributing PPE to dairy farms. Request a kit online at https://shorturl.at/ethov or contact your local extension agent. Requests will be filled as supplies allow.
- Your input could guide future programming! Please complete the short survey at https://tinyurl.com/dairy-extension.

For more information on Dairy Extension or to learn more about our current programs, visit us at VTDairy—Home of the Dairy Extension Program online at www.sas.vt.edu/extension/vtdairy.html

Dr. Christina Petersson-Wolfe, Dairy Extension Coordinator & Extension Dairy Scientist, Milk Quality & Milking Management

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2025 VCE-179NP