Recent developments in estrus synchronization programs

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Introduction

The economic success of beef cow-calf operations relies on the ability to produce one live healthy and heavy at weaning calf per cow every year. To achieve this goal, beef cow-calf producers need to overcome several obstacles related to the cow, bull and the offspring including, ovulation and fertilization rates and embryonic, fetal and postnatal survivals.[1] Over the last five decades several advances in reproductive biotechnologies such as, artificial insemination (AI), estrus-synchronization, and fixed-time AI (TAI) have helped beef producers improve genetic traits of their cattle, tighten the breeding season and shorten the calving season leading to an increase in overall profitability of cow-calf production systems.[2,3]

Enhanced understanding of the dynamics of the estrous cycle have made possible the development of protocols to manipulate the estrous cycle and control ovulation with great precision and success by utilizing natural and/or artificially synthesized hormones, such as gonadotropin releasing hormone (GnRH), prostaglandin $F_{2\alpha}$ (PGF) and progestins. Use of estrus or ovulation synchronization and TAI has facilitated the widespread utilization of AI and can greatly impact the economic viability of cow-calf systems by increasing total pounds of calf weaned per cow exposed.[3] Implementation of TAI programs by beef producers, however, depends largely on 3 key factors:

1) Limited frequency of handling cattle

2) Elimination of detection of estrus

3) Satisfactory and consistent pregnancy outcomes

Fixed-time AI is possibly one of the most impactful technologies available to beef cow-calf producers with benefits that go beyond the genetic improvement potential of AI, but also has direct impacts on the cow and their offspring, as well as allowing for improved labor, nutritional, health and reproductive management optimization and increased profitability of the operation.

Impacts of Estrous Synchronization on cow-calf productivity and profitability

Estrous synchronization and TAI has facilitated the widespread utilization of AI and can greatly impact the economic viability of cow-calf systems by shortening the breeding season, increasing the proportion of females that are exposed to AI, increasing pregnancy rate earlier in the breeding season and, consequently, increasing calving rate early in the calving season (Figure 1).[3] This increase in number of calves being born earlier in the calving season results in older and heavier calves at weaning, with an increase in weaning weights per cow exposed of 17.5 kg.[3] In that same study, a partial budget analysis revealed an overall increase in net returns of $49.14 per cow exposed for cows exposed to TAI compared to cows exposed to natural breeding only.[3] The increase in net returns is mainly related to greater calving rate early in the calving season and the ability to reduce the number of bulls needed for natural service following TAI. However, this decrease in costs related to the purchase of bulls assumed that if 50% of the cows became pregnant to TAI, then a producer would be able to reduce his number of bulls in half while maintaining a similar bull to cow ratio for the remainder of cows that failed to become pregnant by TAI. Cows that remain non-pregnant will be returning to estrus in a synchronized manner, with the majority returning 20 to 23 d post-TAI[8], and could put added pressure on the bull to breed as many females as possible in a much shorter amount of time.

The current recommended bull:cow ratio is 20 to 30 cows in pasture for every one bull.[9,10] However, the average number of beef cows exposed to yearling bulls is reported at 15.2 and for mature bulls at 22.0, regardless of the use of synchronization and TAI.[4] The recommended 1:25 bull:cow ratio may be too conservative and not reaching the bulls full breeding potential, since no changes in pregnancy rates have been reported when non-synchronized cows were on pasture with bulls in ratios of 1:25, 1:44, or 1:60 bulls per cow.[11] Recently, a retrospective study aimed to determine if the bull:cow ratio affects pregnancy success after estrous synchronization and TAI in beef cattle.[12] Decreasing the bull:cow ratio had a negative correlation with pregnancy rates, but only a small portion of the observed variation (1‑4% for bull to total number of cow ratio, 1‑11% of variation for bull to open cow after TAI ratio) can be attributed to the bull:cow ratio. Overall, bull:cow ratios remained similar to 1:30, yet after TAI, the number of open cows that need serviced was reduced by half. Therefore, a bull:cow ratio of at least 1:50 can be used when implementing estrous synchronization and TAI in combination with natural service using mature bulls that have successfully passed a breeding soundness exam.[12] This decrease in the bull:cow ratio may help alleviate the economic burden of implementing estrous synchronization and TAI, while ensuring an increase in profitability resulting from the greater weaning weights per cow exposed.

Factors affecting pregnancy success of TAI

Success of TAI programs can be influenced by factors related to the cow or heifer, the sire, and the management system imposed to those females. Postpartum anestrous remains as a large obstacle to increase pregnancy rate of beef females early in the breeding season and TAI protocols that use a combination of progestins, GnRH and PGF have the ability to induce cyclicity and increase pregnancy rate of anestrous females.[13] However, fertility of anestrous cows is often less than that of cycling cows enrolled in TAI programs.[13,14] Similarly, heifers that have reached puberty prior to the initiation of the TAI protocol have greater pregnancy than heifers that failed to reach puberty.[15]

Nutritional status is closely related to incidence of postpartum anestrous in beef cows and puberty achievement in beef heifers. In beef cows, the two main factors that affect pregnancy success of TAI programs are body condition score (BCS) and days postpartum. A retrospective analysis of several studies[13] indicated that the greatest pregnancy rates to TAI were in mature cows with extended days postpartum (> 72 days) and greater than 5 BCS. Interestingly, pregnancy rate was similar for cows with days postpartum greater than 72 days and BCS lesser than 5, and cows with days postpartum lesser than 72 days and BCS greater than 5. However, in both cases pregnancy rate was decreased significantly compared to cows with having greater than 72 days postpartum and greater than 5 BCS. Highlighting the importance of coupling both days postpartum and BCS for improved TAI pregnancy success.

In heifers, attainment of puberty is dependent on both age and body weight (BW).[15,16] Heifers are usually developed to a target weight, reaching between 55 or 65% of mature BW. Several studies have compared both targeted BW, as reviewed by Perry.[15] When heifers are developed to 55% compared with 65% of mature BW, no difference between developmental weights was detected in percentage of heifers reaching puberty at 12 months of age or yearling pregnancy rates after an 80-d breeding season.[17] However, more heifers developed to 65% of mature BW were pregnant during the first 45 d of the breeding season compared with heifers developed to 55% of mature BW.[18] The development target had carry-over effects, where a difference was observed in postpartum interval with heifers developed to 55% of mature BW taking longer to reinitiate postpartum estrous cycles after calving compared with heifers developed to 65% of mature BW.[17] This is of particular importance, since heifers that calve early during their first calving season will wean heavier calves for up to 6 calving seasons and have increased longevity compared to heifers that calve later in their first calving season.[19]

When considering pregnancy success of TAI programs much attention is focused on the female; however, recent studies have demonstrated the impact of sire on pregnancy success through effects on pregnancy loss. Variation on TAI pregnancy rates of sires has been reported from 35% to 55%, for *Bos taurus* sires that had semen collected, frozen and successfully passed all pre-freezing and post-thawed quality tests.[20] For that same study, sires were classified by pregnancy loss as either high with a mean of 7.25% or low with a mean of 3.93%. Paternal genetics play an important role in placenta formation and appear to be critical during later stages of embryonic development.[21,22] Although selection and classification of sires based on TAI pregnancy success is possible, this task will fall largely on cattle genetic selection and semen industry.

When females exhibit estrus prior to TAI, fertility and pregnancy is enhanced. Concentration of estradiol increases prior to estrus behavior and the initiation of standing estrus, as well as the process of ovulation[23], it also plays a role in changing the uterine environment to receive the embryo and maintain early embryonic development.[23,24] A meta-analysis with over 10,000 females and across several estrous synchronization protocols indicated a 27% increase in TAI pregnancy rate when females were detected in estrus prior to insemination.[25] This study also indicated that BCS and cyclicity status prior to initiation of the TAI protocol impacted estrus response. Cows with BCS > 4 had increased estrus expression compared to cows with < 4 BCS, while cows in anestrous had greater estrus expression compared to estrus-cycling cows.[25] Estrus-cycling cows could be at any stage of the estrous cycle at the onset of the TAI synchronization protocol and may not respond as well as anestrous cows[26], due to a variable response to the first GnRH injection[27,28], which will impact synchronization rate and, therefore, expression of estrus.[25]

Cattle temperament has also been shown to impact overall performance, fertility and pregnancy success of TAI. Adequate handling facilities, personnel training on proper low-stress handling techniques and increased exposure of animals to handling have been shown to reduce the physiological response to stress and improve temperament.[29] In addition, cows with adequate temperament have increased pregnancy rates, calving rate and tend to have increased kg of calf weaned per cow exposed.[30] Furthermore, *Bos taurus* beef heifers with adequate temperament had a greater pregnancy rate when enrolled in a TAI protocol (Figure 2)[31]. In that study, exposure to handling during the TAI protocol reduced concentration of cortisol of heifers, independent of temperament type, indicating that heifers can be quickly acclimated to frequent handling.[31]

Current TAI protocols for beef females

Since the first attempts to synchronize estrus with a single injection of PGF[32], extensive research has been conducted and great advancements in the control of the estrous cycle and synchronization of ovulation of beef females has been achieved.[33] Over the past fifty years, several different protocols for TAI have been developed for beef cows in the U.S. This abundance of TAI protocols, however, generated confusion among beef producers, especially since little consistency existed on nomenclature of protocols and products used. In 2002 a group of beef cattle reproductive biologists from different universities across the U.S. created the Beef Reproduction Task Force (BRTF) in an effort to combine expertise, improve understanding of the physiological processes of the estrous cycle, and educate producers and veterinarians regarding the procedures available to manipulate the estrous cycle and synchronization of ovulation.[34] The BRTF also created a short list of recommended estrous synchronization protocols for beef cows and heifers, which was developed based on results of peer-reviewed and published research as well as field data collected by AI and genetic companies. Since then, this protocol list has been reviewed, updated and published annually on the BRTF website (www.beefrepro.org) and all major bovine AI sire catalogs (Figures 3 and 4). Recently, two new protocol sheets have been added by the BRTF and include a list of protocols for AI with detection of estrus and when using natural service, as well as protocols for the use of sexed semen (Figure 5). When selecting an estrous synchronization and TAI protocol for beef females, using the BRTF protocol sheets is highly recommended.

Protocols that combine estrus detection and TAI. Typically, these protocols rely on a longer interval between luteolysis (PGF injection and progestin device removal) and TAI, which allow for more females to express estrus before insemination. In such protocols, females that express estrus can be inseminated approximately 12 hours from the onset or detection of estrus, or inseminated in two separated TAI events, a strategy that is commonly referred as split-time AI (STAI).[35] Overall pregnancy rates for cows have not been improved when using STAI[35,36], and results in heifers have been mixed.[36,37] Advantages of estrus detection and TAI, and STAI protocols include the increased proportion of females that are inseminated following estrus expression, and the ability to perform TAI without GnRH injection at the time of AI, which can reduce costs associated with the protocols.[35,36,37] The main disadvantage of estrus detection and TAI, and STAI protocols is the increased labor associated with estrus detection and extra cattle handling, which may increase the costs associated with the protocol. In addition, when performing estrus detection, the use of estrus detection aids is recommended to improve estrus detection rate.

Protocols for TAI. The primary advantage of utilizing protocols that rely solely on TAI is the optimization of labor by eliminating estrus detection. For *Bos taurus* beef cows these protocols tend to be short with approximately 8 to 10 days in duration, including the 7-day CO-

Synch+CIDR[38] and the 5-day CO-Synch+CIDR[39]. Comparison between these two protocols have shown either an advantage in pregnancy rates of the 5-day program[39,40] or similar pregnancy rates between protocols.[13,41]

For beef heifers, TAI protocols can be separated into short- or long-term protocols. Similarly to cows, the two most used short-term protocols are the 7-day CO-Synch+CIDR[42] and the 5-day CO-Synch+CIDR[39]. While the two most used long-term protocols are the MGA-PG & TAI and the 14-day CIDR-PG & TAI. [43,44] Reports indicate an increase in synchrony of estrus for long-term TAI protocols in comparison to short-term protocols in beef heifers; however, pregnancy success is similar among protocols.[45]

Protocols for pre-synchronization and TAI. More recently a focus has been set on improving response of the first GnRH injection and consequently ovulation of dominant follicles at the initiation of TAI protocols, improving synchronization of the subsequent follicular waves for both cows and heifers.[46] The most successful pre-synchronization protocols use a combination of PGF injection and prolonged exposure to progesterone to increase ovulation response to first GnRH, improve synchrony and increase estrus response prior to TAI in heifers[47] and in cows[48]. The protocol for pre-synchronization utilizing both PGF and the progestin intravaginal insert became commonly known as the 7&7 Synch.[48]

In cows, reports of pregnancy success of the 7&7 Synch in comparison to other TAI protocols have indicated either an improvement[49] or similar results[50,51]. However, in heifers reports of pregnancy success of the 7&7 Synch indicate an improvement in comparison to the 7-day CO-Synch+CIDR protocol.[52]

Protocols for utilization of sexed semen. Pregnancy success of TAI protocols utilizing sexed semen is typically between 10 and 20% lower than those of conventional semen[53], mainly due to the premature onset of sperm capacitation and reduced sperm lifespan in the female reproductive tract[54]. Sexed semen can be used with any TAI protocol for cows and heifers; however, insemination with sexed semen is more successful when performed on females that have expressed estrus.[55,56,57] In addition, a delayed insemination between 16 to 22 hours following onset of estrus is recommended when performing AI based on detection of estrus and with sexed semen.[54,55,56,57].

Pre-synchronization protocols have overall greater estrus response, as previously discussed, and can be used strategically with sexed semen insemination of females that express estrus with improved pregnancy success in cows[49] and heifers[57].

Considerations for Implementing TAI in beef cow-calf operations

A common strategy to implement estrous synchronization and TAI in beef cow-calf operations is to start by enrolling only heifers. This allows for producers to familiarize themselves with the TAI procedures, while increasing the proportion of heifers that become pregnant early and will calve early, resulting in beneficial long-term effects on weaning weights of the subsequent offspring and their own longevity in the herd.[19] Another strategy is to utilize synchronization protocols for beef cows in combination with natural service and increase proportion of cows pregnant early in the breeding season during one or two years and then enroll cows into a TAI program. This will allow for familiarization with synchronization procedures, while helping increase mean calving date and consequently increase days postpartum and improve BCS prior to the following breeding season, which are essential for TAI pregnancy success.[13] In addition, TAI can be implemented in operations that previously relied only on natural service by enrolling heifers into TAI and then forming groups of cows based on their calving dates and enrolling them to TAI, ultimately creating groups of early calving cows that will be exposed to TAI first and groups of late calving cows that are exposed to TAI later in the breeding season. This approach has been previously described in detail by Lamb and Mercadante[33] and allows for the gradual increase in the proportion of cows that become pregnant early in the breeding season, will calve early into the subsequent calving season and will be eligible to enroll into the early TAI group during the following breeding season.

There are several factors that can impact the results of TAI programs, some of which were discussed previously. Pregnancy success varies among different TAI protocols; however, when utilizing protocols that have been established through intense research and tested in thousands of females under different management conditions, the variation in pregnancy success is significantly reduced. Other factors such as BCS, days postpartum, cyclicity status and even cattle temperament become more important for improved TAI pregnancy success.

An important consideration when adopting a TAI breeding program is the need for a long-term commitment with gradual improvements in TAI results. In 2013 we participated in a breeding program that enrolled over 1,500 mature beef cows in 8 locations in South Dakota (Lamb and Mercadante, data not published). Within location, cows were enrolled using the 7-day CO-Synch+CIDR protocol, were inseminated to the same sires and by the same AI technicians, and pregnancy diagnosis was performed by ultrasonography 35 days post-TAI (Figure 6). Pregnancy rates to TAI varied from 44.4% to 65.8%, with only three locations achieving TAI pregnancy rates above 55%. Those three locations had previously utilized TAI for at least 5 years, while the other locations were using TAI for the first time or infrequently and not consistently. In fact, the location with the greatest TAI pregnancy rate (65.8%) had used TAI for the previous 7 years. Prolonged use of TAI results in a greater proportion of cows becoming pregnant early in the breeding season, which is intensified over multiple years. The mean days postpartum on the day of AI for the location with the greatest pregnancy success was 87 days with a standard deviation of 5.6 days, whereas for the location with the poorest pregnancy rate the mean days postpartum was 70 days with a standard deviation of 16.9 days. This larger variation in days postpartum affects BCS at TAI, both of which are important to pregnancy success.[13] In addition to the improvements in days postpartum and BCS, long-term exposure to TAI can decrease the stress response of cattle from handling[31] resulting in improved pregnancy results.

Summary

Estrous synchronization and TAI remain an important tool to help beef cow-calf producers achieve improved reproductive efficiency, increased weaning weights and greater net returns. However, adoption of TAI by beef cow-calf producers has been slow when compared to the U.S. dairy industry and other major beef producing countries, such as Brazil. Advancements in the understanding of the bovine estrous cycle have made possible the development of estrous synchronization programs that have great synchronization rates and deliver consistent pregnancy rates above 50%. Although much attention is focused on protocol success, pregnancy success is similar among protocols, while other factors such as BCS and days postpartum remain of more importance for TAI pregnancy success. Lastly, when adopting TAI programs, a long-term approach must be considered to ensure achievement of greater pregnancy and overall program success.

Figures

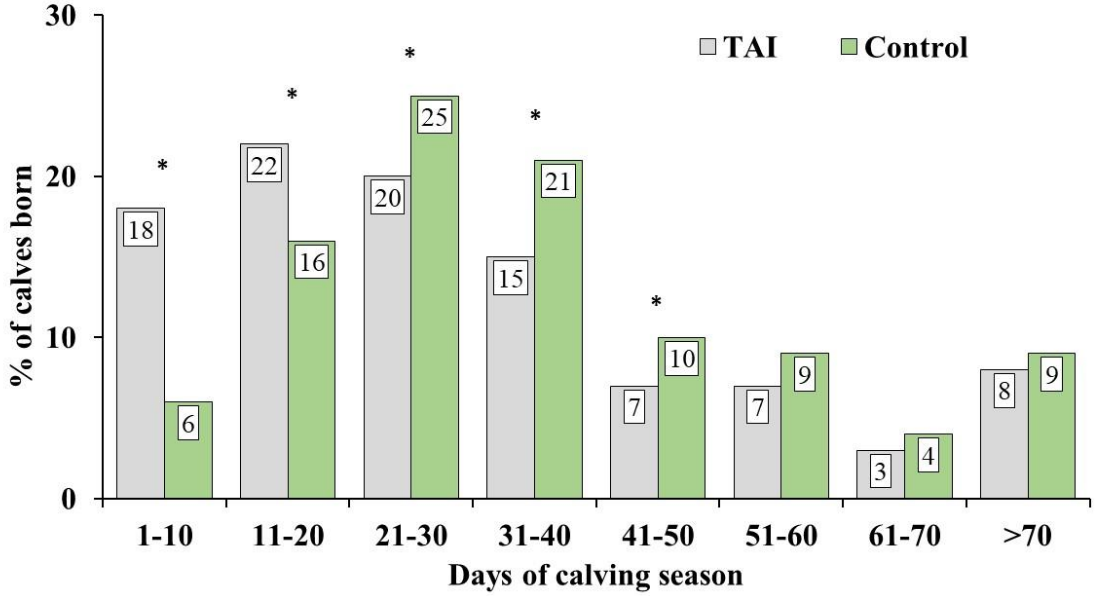


Figure 1. Calving distribution of cows enrolled in a breeding season with (TAI) or without (Control) estrous synchronization. Adapted from Rodgers et al., 2012.[3] *Within 10-d interval treatments differ ($P < 0.05$).

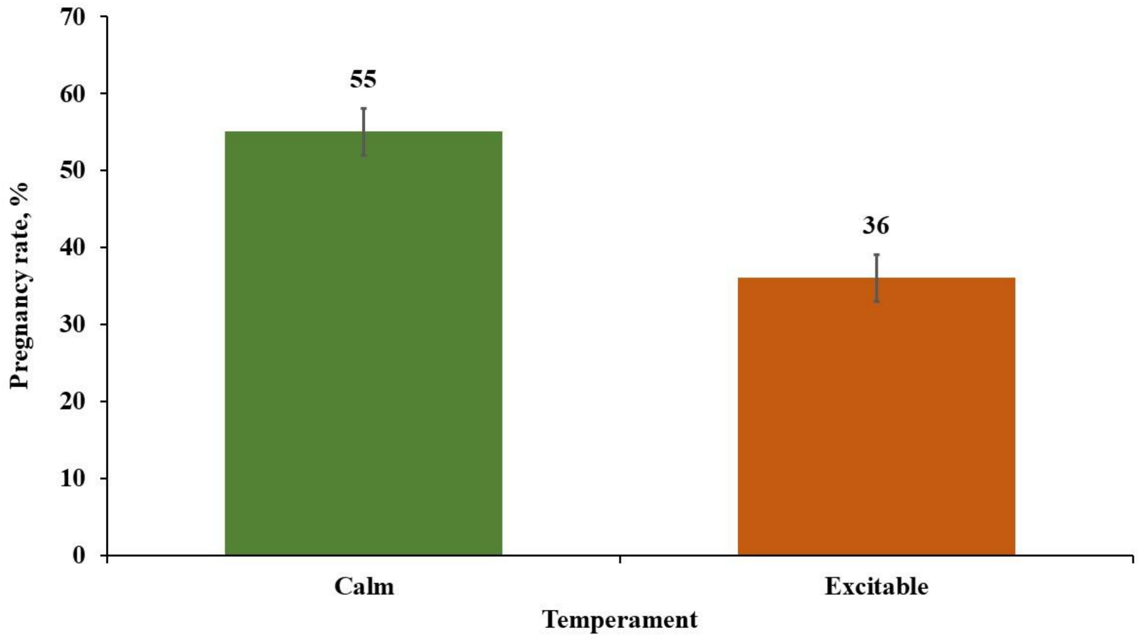


Figure 2. Pregnancy rate of heifers enrolled in TAI according to temperament type. Adapted from Dias et al., 2022.[31] Effect of temperament $P < 0.05$.

Figure 3. Beef Reproduction Task Force list of estrous synchronization protocols for beef cows.

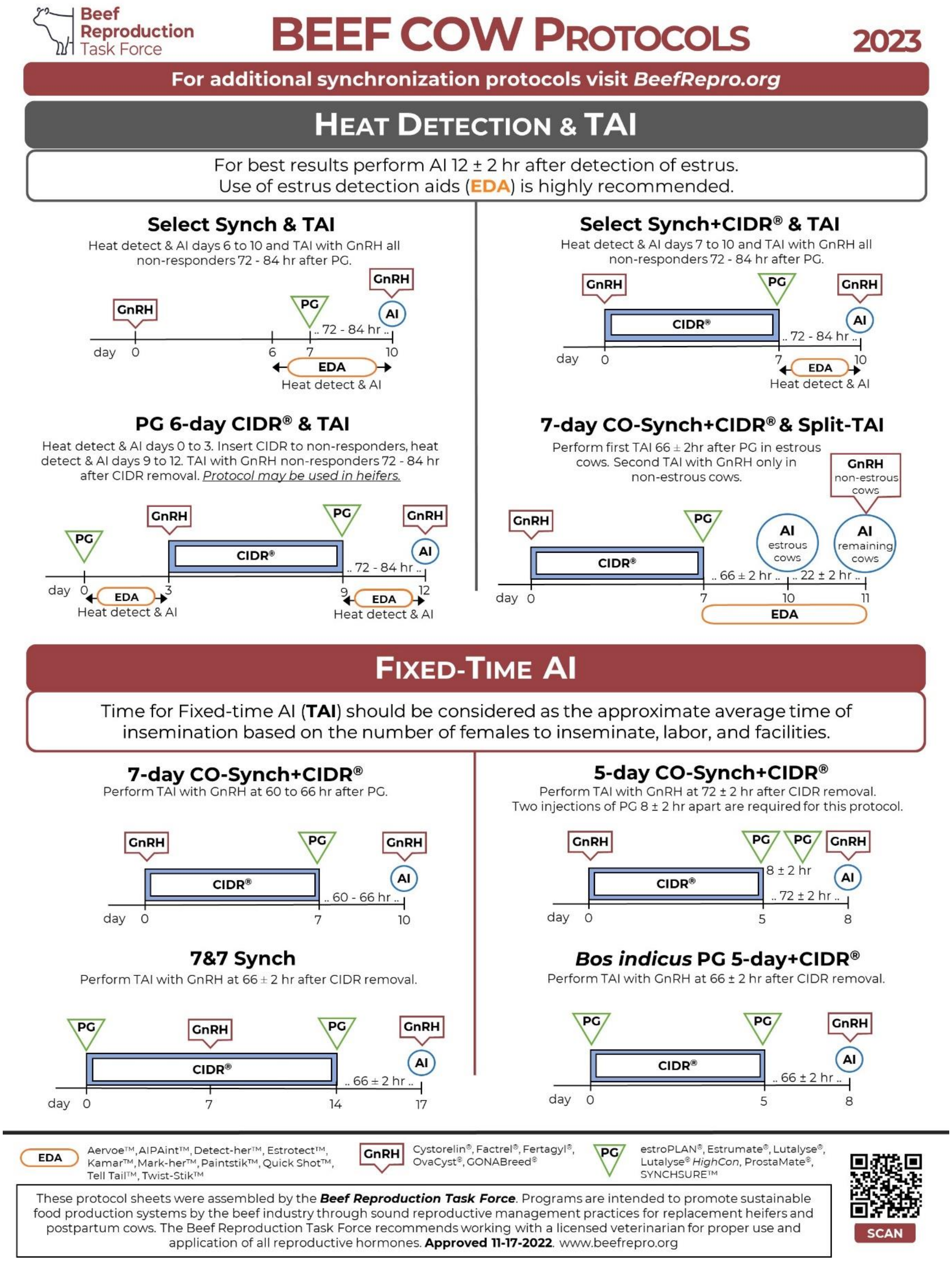


Figure 4. Beef Reproduction Task Force list of estrous synchronization protocols for beef heifers.

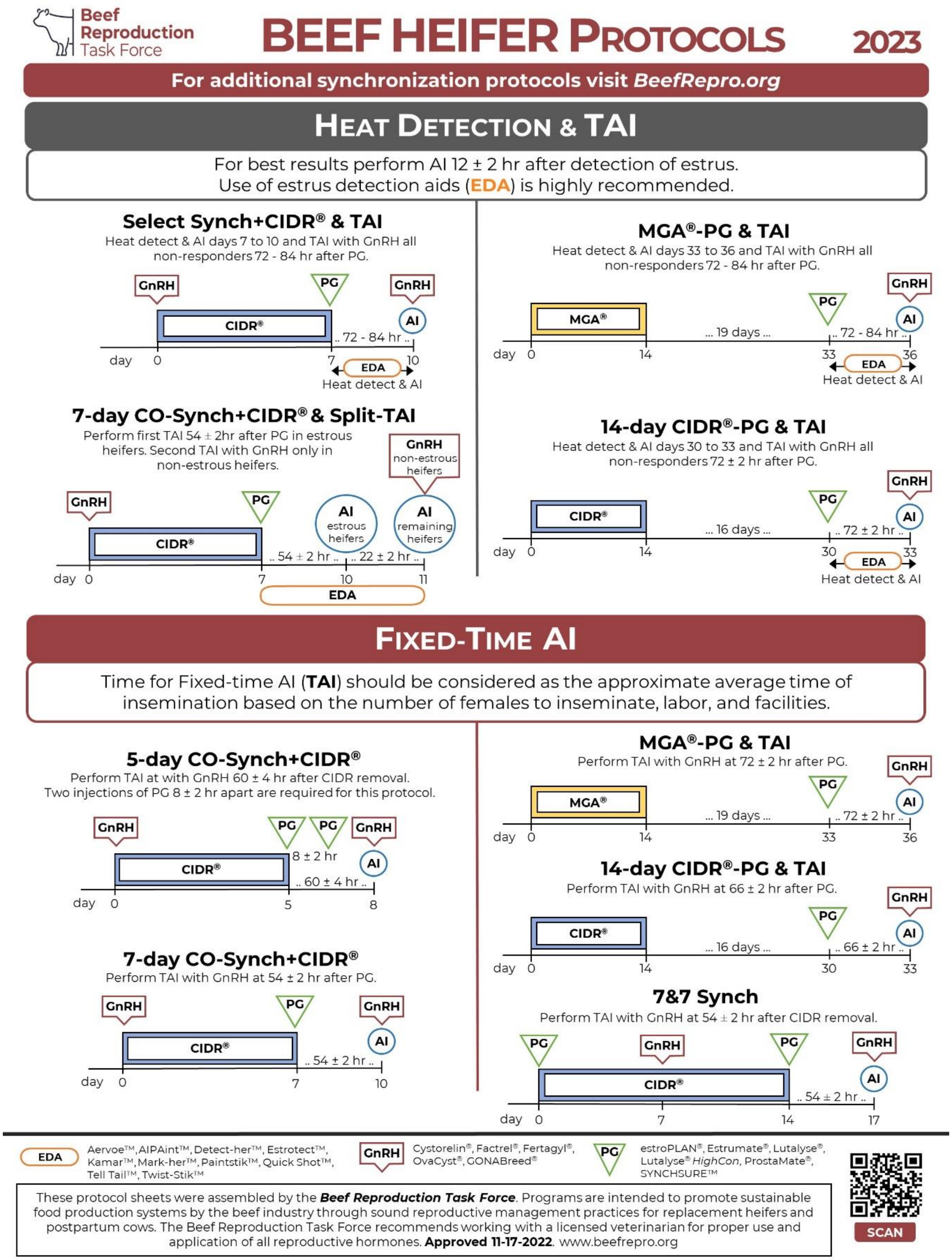
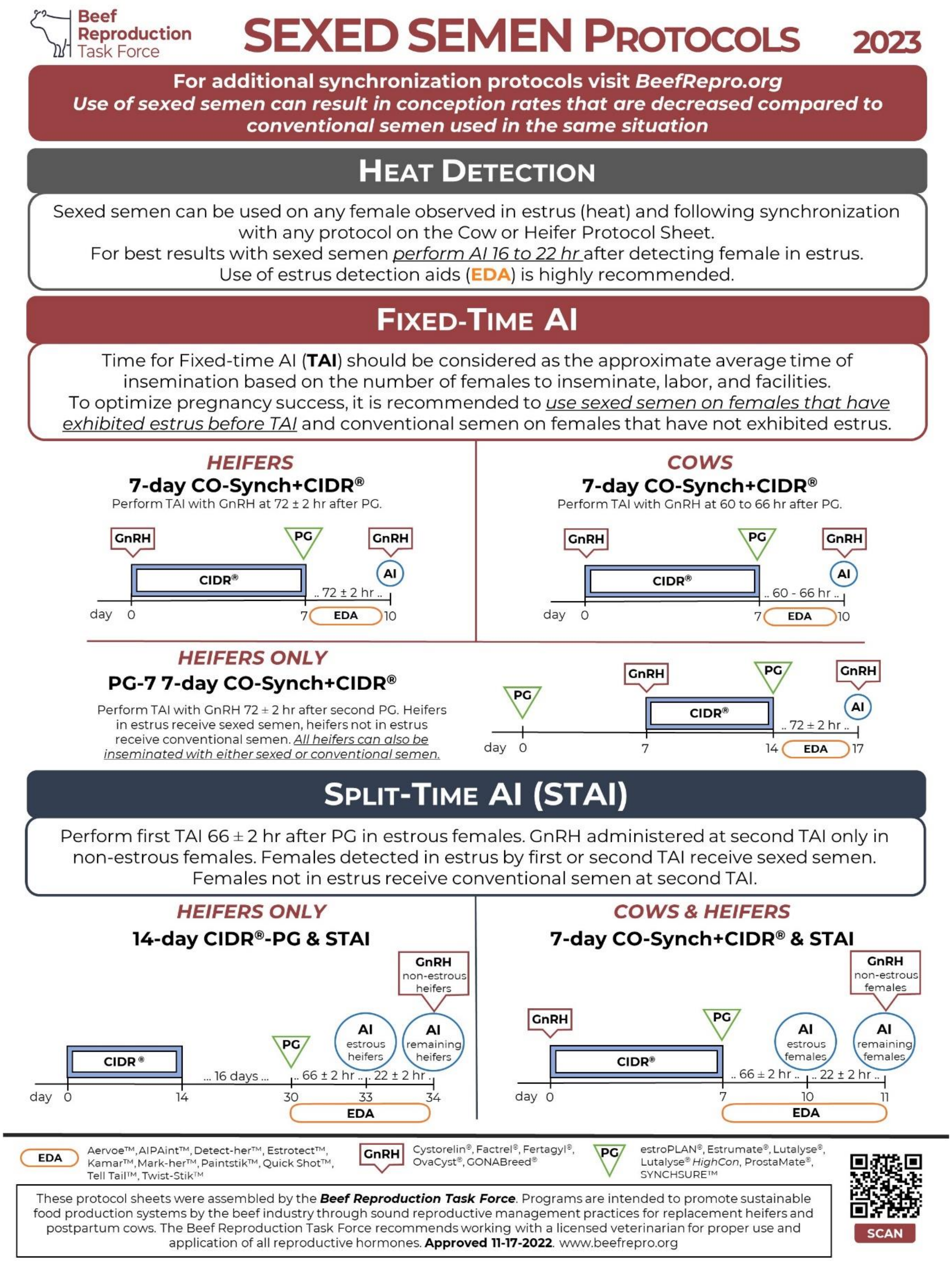


Figure 5. Beef Reproduction Task Force list of estrous synchronization protocols for beef cows and heifers when using sexed-semen.



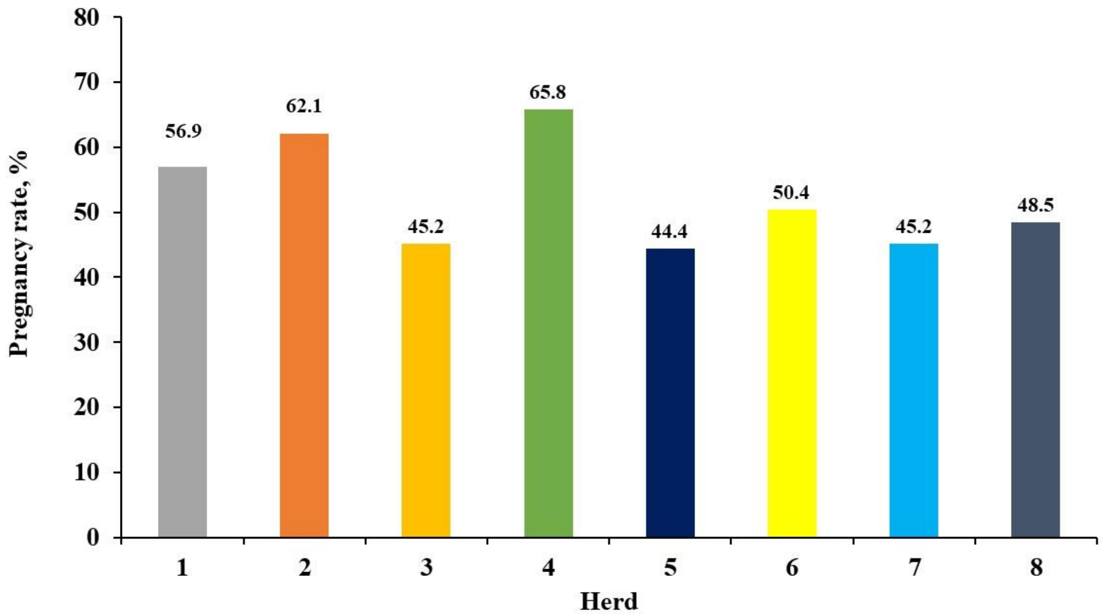


Figure 6. Pregnancy rate of beef cows enrolled in the 7-day-CO-synch+CIDR TAI protocol by location (Lamb and Mercadante, data not published).

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