

TIMING OF INSEMINATION IN BEEF HEIFERS

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Timing of insemination is crucial to successful AI. Semen must be present in the female reproductive tract for sufficient time in advance of ovulation to allow for sperm transport and capacitation to occur. In addition, bovine sperm and oocytes have finite life spans which limit the amount of time before or after ovulation that insemination can result in conception.

Artificial insemination can only be successful if semen is deposited in the female at the appropriate time relative to ovulation (Nalbandov and Casida, 1942). In order for a population of sperm to reach the oviducts that is sizable enough to cause fertilization, a transport time of at least 6 h is needed with sperm numbers steadily increasing thereafter from 8 to 18 h (Thibault, 1973; Wilmut and Hunter, 1984; Hawk, 1987; Dransfield et al., 1998). For maximum fertility to be achieved, spermatozoa must reside in the female reproductive tract for a minimum period of time (Senger, 1999). This phenomenon, termed capacitation, endows the sperm with the ability to undergo the acrosome reaction, exhibit hyperactivated motility and fuse with the oocyte (Yanagimachi, 1981; Bedford, 1983; Herz et al., 1985). In the bovine, the time required *in vivo* for capacitation of ejaculated sperm has been estimated at 6 h based on first penetration of ova after insemination (Hunter, 1980; Herz et al., 1985). Sperm have been found to retain their fertility in the female bovine reproductive tract from 28-50 h (Hamner, 1973; Dukelow and Riegler, 1974). Saacke et al. (2000) examined embryo quality in relation to time of insemination and found a shift from high quality embryos achieved by inseminations at onset of estrus to low quality embryos resulting from inseminations at 24 h following estrus onset. Success in breeding early appeared to be limited by sperm life leading to fertilization failure and breeding late was limited by declining embryonic quality (Saacke et al., 2000). Therefore, timing of insemination is a balance among time of ovulation, time needed for sperm transport and capacitation, and longevity of sperm and oocyte.

Estrous behavior, especially standing to be mounted, is easily observed in cattle. This is facilitated by cows exhibiting homosexual behavior so males do not have to be present to identify cows in estrus. Using initiation or cessation of estrous behavior as a marker for timing of insemination has been practiced for many years. In the beef cow, ovulation occurs 31.1 ± 0.6 hours after the onset of estrus. (White et al., 2002) compared to 27.6 ± 0.6 hours in the dairy cow (Walker et al., 1996). Therefore, timing of artificial insemination has been based on a recommended window of time relative to observed estrus.

The A.M. – P.M. Rule

Early studies found a narrow window of time for acceptable pregnancy rates to AI. Trimberger and Davis, (1943) found the optimum time for AI to be from the middle of estrus up to six h after the end of estrus (Figure 1). Poor conception rates occurred at the start of estrus and 12 h after the end of estrus. Trimberger, (1948) concluded that best rates of conception for dairy heifers (n = 46) occurred in an 18 h window that began 1.5 h after onset of estrus and ended 6 h before ovulation. A similar 18 h window was found for dairy cows (n = 86). This window began 4.5 h after onset of estrus and ended 6 h before ovulation. These findings resulted in the development of the a.m.-p.m. rule for the timing of AI. Barrett and Casida, (1946) collected data from 3,841 inseminations of dairy cattle and grouped them into blocks of time based on when they were bred in relation to onset of estrus determined by visual observation. Average pregnancy rate was 52.4% with the worst pregnancy rates occurring after the estimated time of ovulation. Aschbacher et al., (1956), bred 50 dairy cattle by AI at three different intervals during the same estrus to determine optimum time for insemination. Time of breeding on pregnancy rate was not significant with only 50 cows; however, higher pregnancy rates occurred when inseminations were done before the expected time of ovulation. Early research was most consistent with the idea that cows should not be bred after ovulation and did not convincingly show an optimal time for AI (Wiltbank et al., 2000).

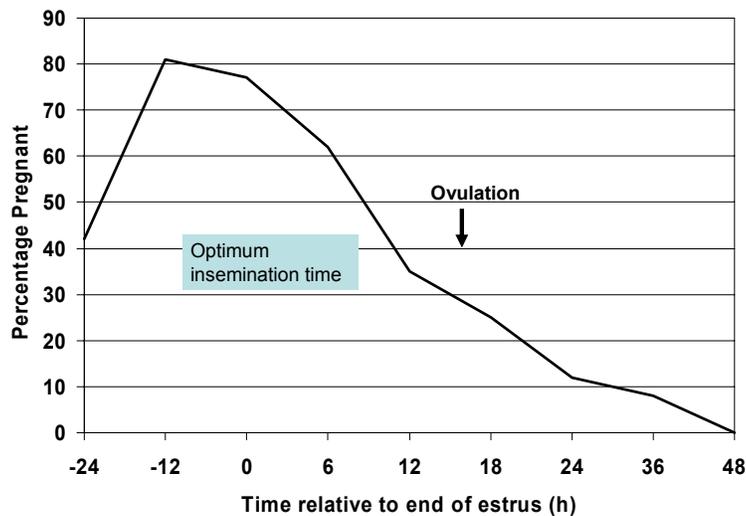


Figure 1. Optimum time for AI in dairy cows relative to estrus and ovulation. Based on Trimberger and Davis, 1943; Adapted from Hafez, 1974

Can timing be improved over the a.m.-p.m. rule?

For over 60 years, the a.m.-p.m. rule served the AI industry well as a system for determining timing of artificial insemination. During the same time period, the AI, beef and dairy industries have experienced tremendous change. Dairy farms of the 40's-early 70's had relatively small herds that often had access to dirt lots or grass pastures. Ample family labor was available to "heat check" the few cows in estrus on any given day. Dairy cows gave significantly less milk. Beef heifers usually did not calve before 36 months of

age. Growth rates, milking ability and beef cow size varied during the 1940-1980 time period. However, in general, these production measures were considerably lower than they are today. Dramatic changes in the AI industry in semen evaluation, extension, packaging and handling are a far cry from the chilled, extended, fresh semen of the 1940's. The advent of estrous synchronizations systems resulted in the ability to have several hundred animals in estrus and ready to be inseminated on a single day. The steady decline in AI pregnancy rates in dairy cattle along with the need to inseminate large numbers of beef cows or heifers in a single day prompted researchers to revisit the question of timing of AI and the a.m.-p.m. rule.

Characteristics of estrus

Understanding the basic characteristics of standing estrus – duration, mounting activity and mounting intensity was limited by the need to conduct around the clock estrus detection coupled with human error associated with identification of animals. Cattle observed in estrus visually in the morning could be in early, mid, or late estrus. Studies using pedometers, and more recently the HeatWatch® (HW) system have been able to provide around the clock detection for estrus in cattle (Maatje et al., 1997; Dransfield et al., 1998). HeatWatch® enabled researchers to collect information on characteristics of estrous activity and more accurately identify time from onset of estrus to insemination

In studies using the HW system on dairy cattle, duration of estrus averaged seven to ten hours, and mounting activity averaged 8-14 times per cow (Walker et al., 1996; Dransfield et al., 1998). In contrast, duration of estrus and mounting activity is greater in beef cattle. In beef heifers, estrus averaged 14 h in duration with 40 to 50 (range 3 to 100+) standing events per heifer (Stevenson et al., 1996, Dorsey, 2005). The duration of estrus for beef cows ranged from 13.9 h to 17.6 h (White et al., 2002). Mounting intensity even in beef heifers was only 2 to 4 mounts per hour which may explain some of the limitations in visual estrous detection (Dorsey, 2005). In synchronized heifers, visual estrous detection was delayed in identifying heifers in estrus by 8 h compared to HeatWatch (Dorsey, 2005). Therefore, twice daily visual estrous detection cannot accurately identify the onset of estrus.

Time of day of estrus onset

It has been reported that onset of estrus occurs more frequently at night in dairy cattle, thus by inseminating at mid morning cows have been in estrus between 12 to 18 h, yielding the highest probability for conception (Hurnik et al., 1975; Nebel et al., 1994). Recently, we determined that onset of estrus occurred in the early morning in a majority of heifers synchronized by CIDR-PG or MGA-PG when average time of PG injections was 1200 (Figure 2). Average time of estrus onset was 0836 and 1054 for heifers bred in spring and fall, respectively. More information effect of estrous synchronization method on the time of day of onset of estrus in beef cattle is needed, but clearly time of day of estrus onset could influence timing of mass insemination.

Once daily AI

Several studies have been performed to determine the effectiveness of breeding once per day versus the a.m./p.m. rule. Nebel et al, (1994) collected results from 3,659 inseminations performed once daily and 3,581 inseminations performed using the

a.m./p.m. rule. Non-return rate results were not significantly different at 60 d with 64.6% using once daily inseminations and 65.6% using the a.m./p.m. rule. Graves et al, (1997) conducted a study using 337 Jersey cows and heifers using once daily AI versus the a.m./p.m. rule and produced conception rates of 57.6% and 60.5% which were not significantly different. However, slightly lower conception rates occurred in cows detected in estrus in the morning and bred later the same morning compared with cows that were not bred until 12 h after estrus onset. Once-daily insemination of dairy heifers after estrous synchronization produced pregnancy rates 62% similar to heifers bred by the a.m./p.m. rule. Artificial inseminations performed once daily in dairy cattle have proven to be as effective under certain conditions as the a.m./p.m. rule. Under routine field conditions of less frequent observation for estrus, cows can be submitted for AI shortly after detection of estrus with nearly optimal results with respect to timing (Nebel et al., 1994).

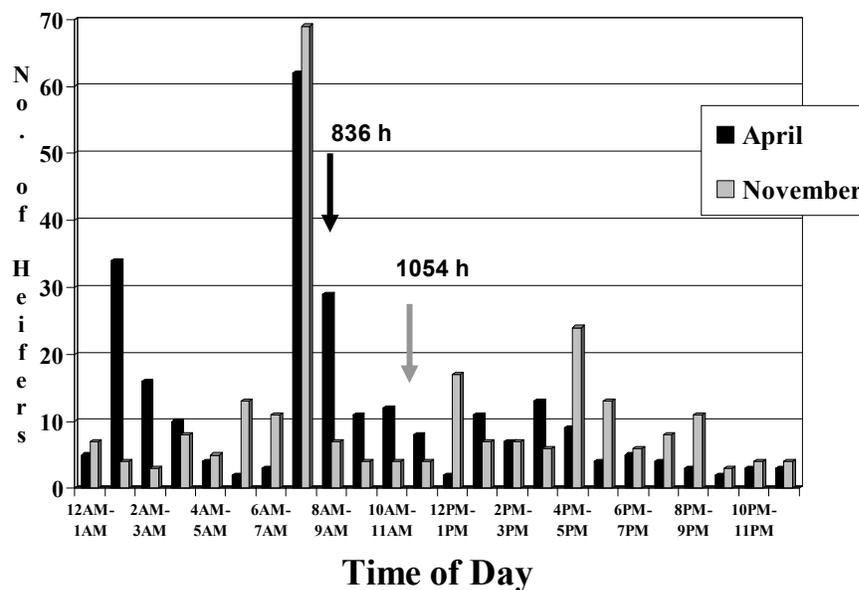


Figure 2. Numbers of heifers in estrus by time of day by season of experiment. Effect of season ($P < 0.05$). (Both HeatWatch[®] and visually observed heifers).

Recommended Timing of Insemination - Has it changed?

Recent studies in cattle indicate that the optimum time for AI relative to the onset of estrus may have shifted in dairy cattle. Maatje et al., (1997) used pedometers on 121 Holstein cows and heifers to predict the optimal time of insemination. Highest chances for conception occurred 6 to 17 h after pedometer activity heightened which coincides with estrus onset. From the data, it was determined that 11.8 h after elevated pedometer activity was the optimum time for AI. Dransfield et al., (1998) studied the interval from onset of estrus onset to AI on 2,661 breedings with dairy cattle. Using the HW system, continuous estrous surveillance was conducted. Optimum conception rates (46-51%) occurred from 4-16 h after estrus onset. Mathematical modeling to predict the optimal time for AI using

activity pedometers and visual signs of estrus, estimated 11.8 h from onset (Maatje et al., 1997), which coincides with the approximate midpoint of 4 to 16 h as optimum using HW (Dransfield et al., 1998). Together, these data would suggest that breeding on the a.m./p.m. rule would result in some cows being inseminated later than optimum especially if visual estrus detection is the primary method for determining onset of estrus.

The window of opportunity for insemination may be wider or later in beef cattle than in dairy cattle. Information available on the duration of elapsed time required from estrus detection to insemination to achieve optimal fertility in beef cattle is sparse and inconclusive (Laster, 1974; Fields et al., 1975; Robbins et al., 1978). Robbins et al., (1978), determined calving rates of 2,091 beef cows from first inseminations at various intervals from estrus. Visual detection of estrus was conducted between dawn and noon, and then from 3-4 pm until dark. All cows were bred the following day after being observed in estrus resulting in insemination times of 12 to 29 h after estrus observation. Optimal calving rates occurred when inseminations occurred between 16 and 25 h after estrus observation with fertility not being lowered precipitously when inseminations were made 4 h earlier or 4 h later (Robbins et al., 1978).

Recent work by our research group indicates that wide range in time of insemination relative to the onset of estrus exists in synchronized heifers. Heifers were synchronized by CIDR-PG or MGA-PG. Estrus detection was performed by the HeatWatch® system. A total of 506 heifers were identified in estrus across synchronization methods with no effect of method ($P > 0.1$) on percentage of heifers in estrus, time of insemination relative to onset of estrus, or AI pregnancy rates. Data on effect of time of insemination from onset of estrus was combined and analyzed. Pregnancy rate was not effected ($P > 0.1$) by time of insemination relative to estrus onset and averaged 60.9%. Regression analysis revealed a significant quadratic component to the formula describing the relationship of time of insemination on percentage pregnant (Figure 3).

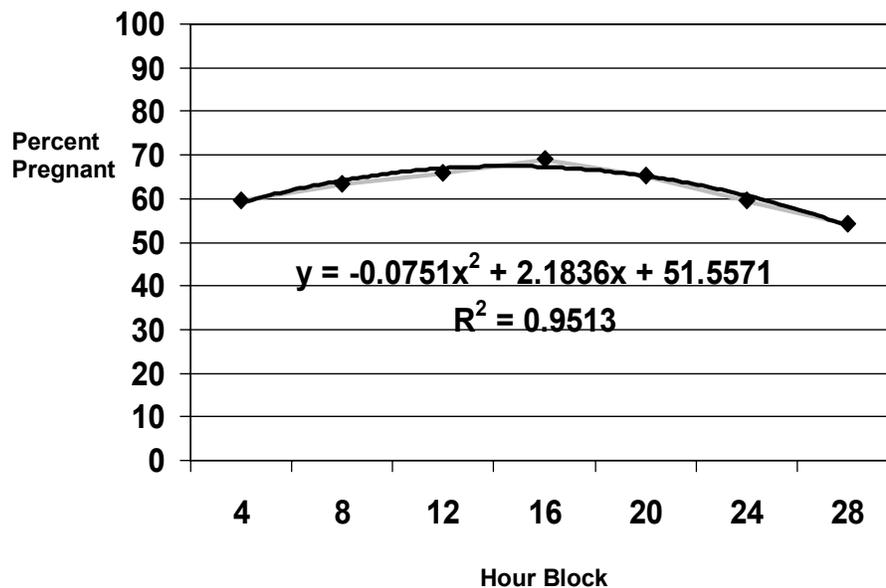


Figure 3. Regression of timing of insemination relative to the onset of estrus (4 h blocks) on pregnancy rate.

The quadratic equation is significant ($P < 0.002$). We concluded that pregnancy rates were not impacted by inseminating heifers between 4 and 24 hours after onset of estrus. The regression equation may indicate a slight peak at 16 hours after onset of estrus, but one must be careful not to over interpret this graph.

Summary

Current research indicates that a wide window exists for time of insemination relative to the onset of estrus in beef heifers when high fertility AI bulls are used. Once-daily AI of heifers may be an option for producers to consider. In addition, there may be greater latitude in timing of insemination when breeding large (>200) groups of synchronized heifers. Because there is variation among AI bulls in fertility when semen is deposited at different times relative to onset estrus (See Dalton in these proceedings), careful selection of bulls is required. Limited information on beef cows indicates that insemination between 16 and 20 hours after estrus may increase AI pregnancy rates; however, more information on timing of AI in postpartum beef cows is needed.

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