Strategies for Success in Heat Detection and Artificial Insemination

Joseph C. Dalton

University of Idaho, 1904 E. Chicago St. Suite AB, Caldwell, ID, USA 83605
Email: jdalton@uidaho.edu

Take Home Messages

- A successful artificial insemination (AI) program must include efficient and accurate heat detection and timely AI relative to ovulation.
- The distribution of first and last standing events of heat occurs with nearly-equal frequency throughout the day and night.
- The duration of heat in dairy cattle is approximately 7 h.
- High producing dairy cows exhibit a shorter duration of heat, resulting in increased difficulty in identifying these cows in heat.
- To maximize conception to AI, insemination should occur at a time that allows for adequate numbers of sperm to be present in the oviducts at the time of ovulation.
- Appropriate semen storage, semen handling, and site of semen deposition in the female reproductive tract are critical factors related to achieving a successful AI program.

What Causes Dairy Cattle To Show Signs Of Heat?

The length of the estrous cycle in dairy cattle is 18 to 24 days, with an average of approximately 21 days. As the nonpregnant cow nears the end of one estrous cycle, the preovulatory follicle (which contains the ovum, or egg), grows larger and produces increasingly more estradiol. Estradiol (a steroid hormone belonging to the estrogen family) plays an important role in reproduction, as it acts on the brain to induce behavioral estrus (heat), and sets into motion the preovulatory surge of luteinizing hormone (LH) from the anterior pituitary gland, which begins the process of ovulation.

The primary sign of heat is when a cow stands to allow a herdmate to mount. Secondary signs include attempting to mount other animals, chin resting,

WCDS Advances in Dairy Technology (2011) Volume 23: 215-229

increased activity, clear mucous discharge from the vulva, and swelling and redness of the vulva.

Environmental conditions may influence whether cows show standing heat. Less estrous behavior occurs when cows are eating or contained in holding pens (Stevenson, 2001). Footing surface is also important, as cows show less activity when housed on slippery concrete or frozen ground (Britt et al., 1986). Lastly, lameness plays a role as animals with foot problems show less mounting and standing activity regardless of whether the problem is structural, subclinical, or clinical (Leonard et al., 1994). Consequently, to enhance the opportunity to identify cows in heat, cows should have healthy feet and legs, access to areas that provide good footing, and few, if any, obstacles to hinder interaction (Nebel, 2003).

■ Heat Detection

A successful AI program must include efficient and accurate heat detection and timely AI relative to ovulation. The failure to detect heat is the most common and costly problem of AI programs and the major limiting factor of reproductive performance on many dairies (Nebel and Jobst, 1998). This is evidenced by the reality that heat detection efficiency (defined as the proportion of possible heat periods detected during a period of time) is less than 50% in many herds. Heat detection accuracy (defined as the proportion of detected heat periods in which cows exhibited low progesterone concentration in milk or blood) varies widely, as 0 to 60% of cows presented for AI did not exhibit low progesterone at the time of AI (Reimers et al., 1985; Nebel et al., 1987).

Progesterone concentration in milk and blood is associated with events of the estrous cycle as concentration is low (~1.0 ng/mL or less) for 2 d prior to heat and remains low for approximately 2 to 3 d after heat (Senger, 1999; Hopkins, 1989; Nebel et al., 1987). Low milk or blood progesterone alone is not an indicator of heat; however, high milk or blood progesterone is considered a confirmation that a cow is not in heat. Consequently, heat detection accuracy may be monitored periodically via blood samples taken from animals identified in heat and receiving AI. A reasonable goal is for 95% of all cows to exhibit low blood progesterone on the day of AI.

■ Importance of Heat Detection Relative to Ovulation

Using HeatWatch (Cow Chips, LLC, Manalapan, NJ, USA) to determine the onset of heat and ultrasonography to detect ovulation, Walker et al. (1996) determined the interval from the onset of standing activity to ovulation to be

approximately 28 h in lactating dairy cattle. The physiological relationship linking ovulation to the onset of standing activity underscores the importance of accurate heat detection: There is a limited window of opportunity in which to maximize conception to AI. Many biological events occur within the limited window of opportunity (Figure 1), including:

- the transport time required of viable sperm from the site of deposition to the site of fertilization
- the functional viable lifespan of sperm and ova
- the timing of ovulation in relation to AI

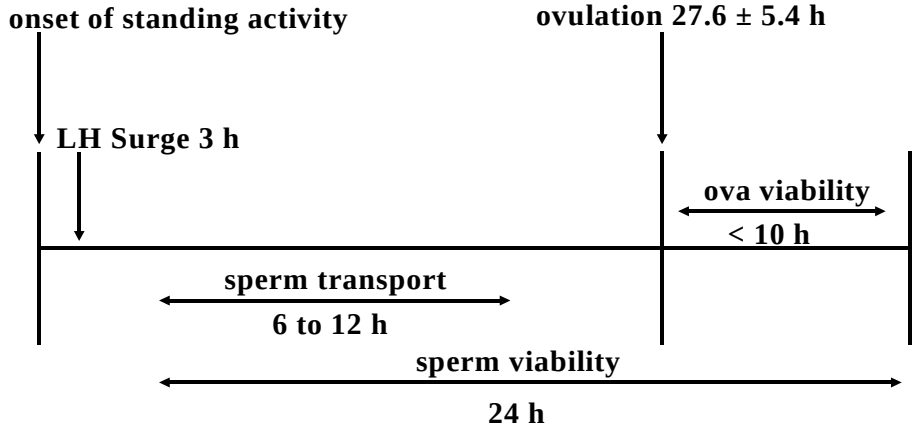

Figure 1. Biological events contributing to the optimization of AI.

The estimated time required for sustained transport of viable sperm to the site of fertilization is between 6 to 12 h (Hawk, 1987; Hunter and Wilmut, 1983; Wilmut and Hunter, 1984). The functional viable lifespan of sperm in the female reproductive tract has been estimated at 24 h (Trimberger, 1948), whereas the optimal window for fertilization of the ovum (egg) is estimated to be less than 10 h (Brackett et al., 1980) (Figure 1).

Dransfield et al. (1998) used HeatWatch on 17 commercial dairies to determine the onset of standing heat relative to the 24-h clock. Dransfield et al. (1998) reported that the distribution of first and last standing events (standing to be mounted by a herdmate) was nearly equal when the 24-h clock was divided into four 6-h periods (Table 1). Xu and coworkers (1998) reported similar results with dairy cattle grazing on pasture in New Zealand.

Table 1. The distribution of first and last standing events of heat as identified by HeatWatch[1].

	Time period			
Event	12:01 AM to 6:00 AM	6:01 AM to 12:00 PM	12:01 PM to 6:00 PM	6:01 PM to 12:00 AM
Onset of first standing event, (%)	24.5	28.4	19.8	27.3
Termination of last standing event, (%)	24.8	27.8	23.4	24.0

[1]2,055 heat periods; Adapted from Dransfield et al. (1998).

The average duration of heat described by Dransfield et al. (1998) using HeatWatch is much shorter than originally reported by Hammond (1927) and Trimberger (1948) (Table 2). Furthermore, Lopez et al. (2004) used HeatWatch and reported that milk yield affected the number of standing events and duration of heat (Table 3). According to Dransfield et al. (1998) and Lopez et al. (2004), the average number of standing events range between 6 to 9 per heat. Typically, each standing event lasts just 2.5 seconds. Consequently, the difficulty in detecting cows in standing heat becomes quite clear. Every 21 days a cow may stand to be mounted by a herdmate for only 15 to 23 seconds! Causing further problems with heat detection is the reality that approximately 20% of heat periods monitored by HeatWatch (Dransfield et al., 1998) were classified as "low intensity and short duration," providing at least a partial explanation of the inability of many producers and technicians to identify greater than 70% of possible heats, as reported on DHI summaries (Nebel, 2003).

Table 2. The duration of heat in dairy cattle.

Reference	Average duration of heat, (h)
Hammond (1927)	19.3
Trimberger (1948)	17.8
Dransfield et al. (1998)	7.1

Table 3. The effect of milk yield on duration of heat and number of standing events in dairy cattle[1].

Item	Low milk (n = 177)	High milk (n = 146)
Milk production, kg/d	33.5 ± 0.3	46.4 ± 0.4
Duration of heat, h	10.9 ± 0.7	6.2 ± 0.5
Number of standing events	8.8 ± 0.6	6.3 ± 0.4

[1]Adapted from Lopez et al. (2004).

The mechanism by which high milk production may alter the expression of heat is not completely understood. Sangsritavong et al. (2002) reported the plane of nutrition necessary for high milk production increases liver blood flow and metabolic clearance of progesterone and estradiol. According to Wiltbank (2003), this would lead to lower circulating concentrations of progesterone and estradiol in high producing cows. Therefore, increased metabolic clearance of estradiol after the onset of heat may contribute to the altered expression of heat in high producing cows, resulting in increased difficulty in identifying these cows in heat.

A variety of automated heat detection systems (HeatWatch, CowChips, LLC, Manalapan, NJ, USA; Select Detect, Select Sires, Inc, Plain City, OH, USA; ai24, Semex Alliance, Guelph, ON, Canada; Heatime, Micro Dairy Logic, Amarillo, TX, USA; Smart Bolus, TenXsys, Eagle, ID, USA) and heat detection aids such as tail paint or chalk, and heat mount detectors (Kamar, Kamar Products, Inc, Zionsville, IN, USA; Bovine Beacon, OmniGlow, LLC, Brownsville, TX, USA; Estrotect, Rockway, Inc, Spring Valley, WI, USA) are available to assist in the identification of cows in heat. Automated heat detection systems and aids play a vital role when used in conjunction with visual observation and appropriate interpretation of records. For example, Peralta et al. (2005) investigated three heat detection methods (HeatWatch; ALPRO activity system, DeLaval, Kansas City, MO, USA; visual observation three times daily) and reported the highest conception rate (CR) occurred with the combination of visual observation and HeatWatch. Consequently, Peralta et al. (2005) concluded that a combination of two heat detection methods (instead of using a single method) appeared to be the best strategy for obtaining more pregnant cows. Regardless of the heat detection aids or automated heat detection system used, producers and technicians should use common sense and search for additional or secondary signs of heat before performing AI.

■ Time of AI

Dalton and coworkers (2001) conducted an experiment to determine the effect of insemination time on fertilization status and embryo quality in single-ovulating cows. All cows were continuously monitored by HeatWatch and received AI with one 0.5-mL straw (25×10^6 sperm) of semen from one of three bulls at 0, 12, or 24 h after the onset of standing heat. Although fertilization rates were greatest in embryos recovered following the 24-h AI treatment (Figure 2), embryo quality declined with increasing intervals after the onset of standing heat, from high quality embryos (0-h AI) to low quality embryos (24-h AI; Dalton et al., 2001).

Embryo quality at the late insemination (24-h AI) may be impaired due to an aging ovum at the time of fertilization (Figures 2 and 3). In this scenario, 24-h AI would result in sperm reaching the site of fertilization at 30 + h after the onset of standing heat, accounting for the time required for sustained sperm transport (6 to 12 h; Hawk, 1987; Hunter and Wilmut, 1983; Wilmut and Hunter, 1984). Consequently, fertilization of an aging ovum would occur, likely leading to lower embryo quality. In contrast, the improved embryo quality associated with 0-h AI suggests that the duration of sperm residence in the female reproductive tract may allow further selection pressure favoring competent sperm, thus optimizing embryo quality at early insemination. Nevertheless, the low fertilization rate associated with 0-h AI is likely due to inadequate sperm longevity (Figures 2 and 3).

Artificial insemination at approximately 12 h after onset of standing heat may provide a compromise between the potentially lower fertilization rate of 0-h AI and the lowered embryo quality (due to increased degenerate embryos) of 24-h AI (Figures 2 and 3). From these data, fertility would be expected to be optimized following the 12-h AI (Figure 3). This agrees with Dransfield et al. (1998), in which the optimal time of AI for lactating cows identified in estrus by HeatWatch was 4 to 16 h after the onset of estrus. This also agrees with Maatje et al. (1997) who described an optimal time of insemination between 6 to 17 h after an increase in locomotive activity as monitored by pedometry. As previously mentioned, conception to AI will likely be maximized when insemination occurs at a time that allows for adequate numbers of sperm to be present in the oviducts at the time of ovulation.

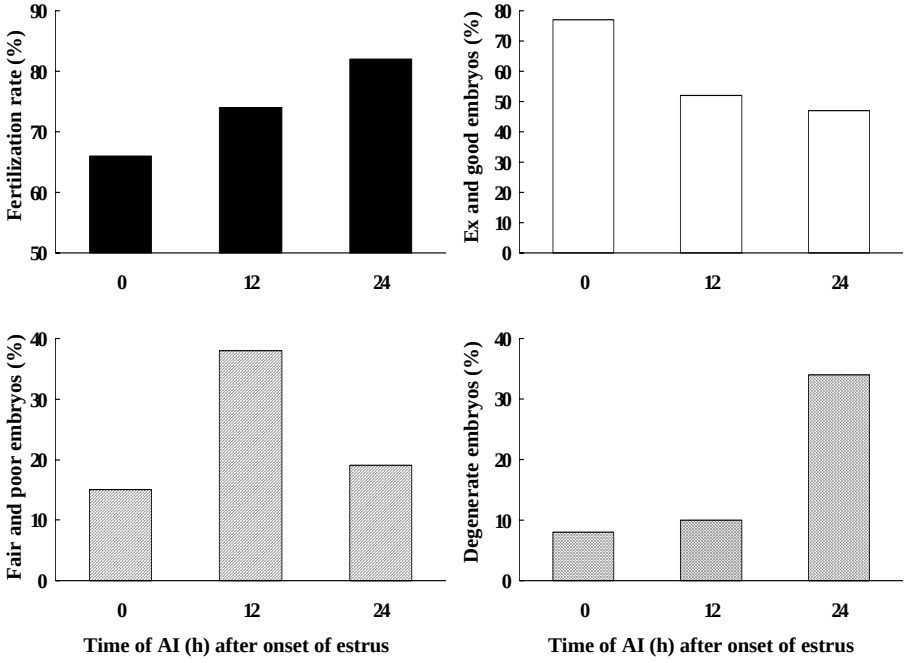

Figure 2. Effect of time of AI after onset of standing heat (as determined by the first standing event in cattle continuously monitored by HeatWatch) on fertilization status and embryo quality. (Adapted from Dalton et al., 2001).

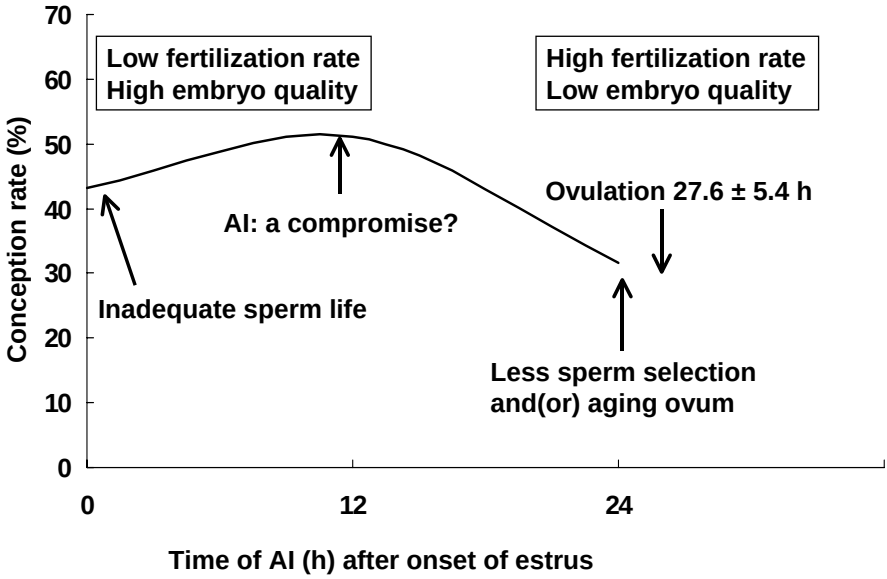

Figure 3. Artificial insemination at 12 h after onset of standing heat appears to be a compromise between the low fertilization rate and high embryo quality of early inseminations and the high fertilization rate and low embryo quality of late inseminations. (Adaptation of data from Dransfield et al., 1998, and Dalton et al., 2001, originally published by Saacke et al., 2000).

In management schemes that allow for visual observation of cows for signs of heat, observation periods should occur at least three times daily (Nebel, 2003). This recommendation stems from classic research (Trimberger and Davis, 1943; Trimberger, 1948) in which visual observation conducted three times daily led to the development of the A.M.-P.M. guideline where: 1) cows observed in heat during the A.M. should receive AI during the P.M., and 2) cows observed in heat during the P.M. should receive AI during the next A.M. Although the onset of heat is usually not known following visual observation three times daily, on average, cows will receive AI approximately 16 h after the onset of heat (12 h after observation + 4 h after initiation of heat, on average). Therefore AI would occur approximately 12 h before ovulation, on average. To reduce the interval from presumptive onset of heat to time of AI, producers and technicians should increase the number of visual observation periods, resulting in a shorter time between observations.

In various management schemes it is unrealistic to expect heat detection and AI to occur more than once daily. In this management strategy AI should occur as soon after detection as possible, as the short lifespan of the ovum must be considered the limiting factor. Heat detection and AI studies,

however, have reported similar fertility results for once-daily AI and AI following the A.M. – P.M. guideline (Foote, 1978; Nebel et al., 1994).

■ Semen Storage and Handling

To attain the maximal potential fertility within straws of frozen semen, the liquid nitrogen tank must be managed properly. The liquid nitrogen tank consists of a "tank within a tank," with insulation under vacuum between the inner and outer tanks. Liquid nitrogen tanks should be stored in a clean, dry area, and preferably on a wood stand to avoid possible corrosion (due to contact with wet or damp concrete). The liquid nitrogen tank should be securely fastened during transportation to avoid tipping the tank over and damaging the tank, both of which usually result in the premature loss of liquid nitrogen.

Regardless of whether the liquid nitrogen tank is stored in an office or transported in a vehicle to a location closer to the cows to be serviced, a detailed inventory of semen should be easily accessible, so that straws may be located and removed from the tank quickly to avoid exposure of semen to ambient temperature. When removing a straw from a liquid nitrogen tank, it is imperative that the technician keep the canister, cane and unused semen straws as low as possible in the neck of the tank. A best management practice is to keep all unused straws below the frost-line in the neck of the tank. Keep in mind that although the temperature of liquid nitrogen is -196°C, there is a temperature gradient in the neck of the tank. For example, a tank with a neck tube that measures 15.24 cm long may have a temperature of -75°C in the middle of the neck (7.62 cm below the top), while the temperature at 2.54 cm below the top may be -15°C. Previous reports have shown that sperm injury (as judged by sperm motility) occurs at temperatures as low as -79°C (Etgen et al., 1957; Bean et al., 1963; DeJarnette et al., 1999). Furthermore, injury to sperm cannot be corrected by returning semen to the liquid nitrogen (Berndtson et al., 1976; Saacke et al., 1978). As would be expected, the temperature in the neck of the tank becomes warmer as the liquid nitrogen level in the tank decreases. Therefore, another best management practice is to monitor the liquid nitrogen level in your tank regularly, and never let the level of liquid nitrogen go below 4 or 5 cm.

Prior to thawing semen for AI, the water bath temperature should be checked with a thermometer. Most semen packaged in 0.25- or 0.5-mL straws should be thawed in water at 35 to 37°C. Research has shown that warm water thawing of semen straws results in greater sperm survival as compared to ice water and air thawing. Generally, ice water and air thawing occurs too slowly, exposing sperm to extended thawing which allows ample time for sperm to be damaged. Warm water thawing rapidly transforms sperm, thus minimizing the time in which sperm may be injured. "Pocket thawing" should only be used for

semen processed and packaged by an AI stud that specifically recommends this procedure.

When numerous cows must be inseminated on a given day, AI technicians routinely thaw multiple straws of semen simultaneously to facilitate AI in a timely manner. In 2004, a field trial was conducted (Dalton et al., 2004) to answer the following questions:

- What is the effect of simultaneous thawing of multiple straws of semen and sequence of insemination (1st, 2nd, 3rd or 4th) on CR?
- Is the CR achieved following AI by professional AI technicians (PAI) and herdsman-inseminators (HI) different?
- What is the effect of elapsed time from initiation of thawing straws of semen to seminal deposition on CR?

Although the average CR differed between PAI and HI (45% vs. 27%, respectively), simultaneous thawing and sequence of insemination (1st, 2nd, 3rd or 4th), and elapsed time from initial thaw to completion of fourth AI had no effect on CR within inseminator group. Nevertheless, a general recommendation as to the number of straws that may be thawed simultaneously detracts from the overall importance of proper semen handling for successful AI. Conception to AI is most likely to be maximized when personnel:

- accurately identify cows in heat
- follow the AI stud's recommendations for thawing semen
- prevent direct straw-to-straw contact during thawing to avoid decreased post-thaw sperm viability as a result of straws freezing together (Brown et al., 1991)
- use appropriate hygienic procedures
- maintain thermal protection of straws during AI gun assembly and transport to the cow
- deposit semen in the uterus of the cow within approximately 10-15 minutes after thawing

Inseminator Skill (Site of Semen Deposition)

Many studies have compared semen deposition near the greater curvature of the uterine horns with conventional deposition into the uterine body. Although Senger et al. (1988), López-Gatius (1996), and Pursley (2004) reported increased fertility when semen was deposited in the uterine horns rather than

the uterine body, Hawk and Tanabe (1986), Williams et al. (1988), and McKenna et al. (1990) found no difference in fertility when comparing uterine body and uterine horn inseminations. Furthermore, Diskin et al. (2004) reported an inseminator and site of semen deposition effect (interaction), with evidence of either an increase, decrease, or no effect of uterine horn deposition on conception to AI for individual inseminators.

Unfortunately, it is not clear why a few studies have shown a fertility advantage following uterine horn insemination while others have not. A possible explanation for the positive effect of uterine horn inseminations may be related to the minimization or elimination of cervical semen deposition. Cervical insemination errors account for approximately 20% of attempted uterine body depositions (Peters et al., 1984). Macpherson (1968) reported that cervical insemination resulted in a 10% decrease in fertility when compared with deposition of semen in the uterine body. To maximize CR, all AI technicians must develop sufficient skill to recognize where the tip of the AI gun is at all times.

■ Timed AI Programs

Timed AI programs provide an organized and efficient approach to administering first AI and the improvement of reproductive performance (Santos, 2007). Conception to AI resulting from these protocols varies. Nevertheless, there are common “compliance” factors among all protocols that must be followed, including accurate cow identification, appropriate drug dosage, time, day, and route of administration, and time of insemination. With the advent of timed AI protocols, more cows are inseminated in a short period of time and, hopefully, more cows will become pregnant earlier in lactation. Nevertheless, it is apparent that some cows will need to be inseminated a second or third time in order to become pregnant. Producers and AI technicians should pay close attention 18 to 24 days after AI to detect cows that return to heat, and (or) implement a follow-up breeding program (Re-synch; Pursley, 2007).

■ Summary

A successful AI program must include efficient and accurate heat detection and timely AI relative to ovulation. Although the average duration of heat in dairy cattle is approximately 7 h, high producing dairy cows exhibit a shorter duration of heat, resulting in increased difficulty in identifying these cows in heat. A variety of automated heat detection systems and heat detection aids are available to assist in the identification of cows in heat. Regardless of the heat detection aids or automated heat detection system used, producers and

technicians should use common sense and search for additional or secondary signs of heat before performing AI. To maximize conception to AI, insemination should occur at a time that allows for adequate numbers of sperm to be present in the oviducts at the time of ovulation. Lastly, appropriate semen storage, semen handling, and site of semen deposition in the female reproductive tract are critical factors related to achieving a successful AI program.

Acknowledgement

Mention of specific products is for informational purposes only, and does not imply endorsement of products over similar products not mentioned.

References

Bean, B.H., B.W. Pickett, and R.C. Martig. 1963. Influence of freezing methods, extenders and storage temperatures on motility and pH of frozen bovine semen. J. Dairy Sci. 46:145-149.

Berndtson, W.E., B.W. Pickett, and C.D. Rugg. 1976. Procedures for field handling of bovine semen in plastic straws. In: Proc. Nat'l. Assn. Anim. Breeders 6th Tech. Conf. AI and Reprod., Columbia, MO, pp. 51-60.

Brackett, B.G., Y.K. Oh, J.F. Evans, and W.J. Donawick. 1980. Fertilization and development of cow ova. Biol. Reprod. 23:189-205.

Britt, J.H.,R.G. Scott, J.D. Armstrong, and M.D. Whitacre. 1986. Determinants of estrous behavior in lactating Holstein cows. J. Dairy. Sci. 69:2195-2202.

Brown, D.W., P.L. Senger, and W.C. Becker. 1991. Effect of group thawing on post-thaw viability of bovine spermatozoa packaged in .5 milliliter French straws. J. Anim. Sci. 69:2303–2309.

Dalton, J.C., S. Nadir, J.H., Bame, M. Noftsinger, R.L. Nebel, and R.G. Saacke. 2001. Effect of time of insemination on number of accessory sperm, fertilization rate, and embryo quality in nonlactating dairy cattle. J. Dairy Sci. 84:2413-2418.

Dalton, J.C., A. Ahmadzadeh, B. Shafii, W.J. Price, and J.M. DeJarnette. 2004. Effect of thawing multiple 0.5-ml semen straws and sequential insemination number on conception rates in dairy cattle. J. Dairy Sci. 87:972-975.

DeJarnette, J.M., D.A. Barnes, and C.E. Marshall. 1999. Effects of pre- and post- thaw thermal insults on viability characteristics of cryopreserved bovine semen. Theriogenology. 53:1225-1238.

Diskin, M.G., J.R. Pursley, D.A. Kenny, J.F. Mee, and J.M. Sreenan. 2004. The effect of deep intrauterine placement of semen on conception rate in dairy cows. J. Dairy Sci. 87:(Suppl. 1):257 (Abstr.).

Dransfield, M.B.G., R.L. Nebel, R.E. Pearson, and L.D. Warnick. 1998. Timing of insemination for dairy cows identified in estrus by a radiotelemetric estrus detection system. J. Dairy Sci. 81:1874-1882.

Etgen, W. M., J.M. Ludwick, H.E. Rickard, E. A. Hess, and F. Ely. 1957. Use of mechanical refrigeration in preservation of bull semen. J. Dairy Sci. 40:774-778.

Foote, R.H. 1978. Time of artificial insemination and fertility in dairy cattle. J. Dairy Sci. 62:355-358.

Hammond, J. 1927. The physiology of reproduction in the cow. University Press, Cambridge, England, pp. 9-49.

Hawk, H.W., and T.Y. Tanabe. 1986. Effect of unilateral cornual insemination upon fertilization rate in superovulating and single-ovulating cattle. J. Anim. Sci. 63:551-560.

Hawk, H.W. 1987. Transport and fate of spermatozoa after insemination of cattle. J. Dairy Sci. 70:1487-1503.

Hopkins, S.M. 1989. Reproductive patterns of cattle. In: Veterinary Endocrinology and Reproduction, Ed. L.E. McDonald, Lea and Fibiger, Philadelphia, PA, pp. 399-415.

Hunter, R.H.F., and I. Wilmut. 1983. The rate of functional sperm transport into the oviducts of mated cows. Anim. Reprod. Sci. 5:167-173.

Leonard, F.C., J. O'Connell, and K. O'Farrell. 1994. Effect of different housing conditions on behaviour and foot lesions in Friesian heifers. Vet. Rec. 134:490-494.

Lopez, H., L.D. Satter, and M.C. Wiltbank. 2004. Relationship between level of milk production and estrous behavior of lactating dairy cows. Anim. Reprod. Sci. 81:209–223.

López-Gatius, F. 1996. Side of gestation in dairy heifers affects subsequent sperm transport and pregnancy rates after deep insemination into one uterine horn. Theriogenology. 45:417-425.

Maatje, K., S.H. Loeffler, and B. Engel. 1997. Predicting optimal time of insemination in cows that show visual signs of estrus by estimating onset of estrus with pedometers. J. Dairy Sci. 80:1098-1105.

Macpherson, J.W. 1968. Semen placement effects on fertility in bovines. J. Dairy Sci. 51:807-808.

McKenna, T., R.W. Lenz, S.E. Fenton, and R.L. Ax. 1990. Nonreturn rates of dairy cattle following uterine body or cornual insemination. J. Dairy Sci. 73:1779-1783.

Nebel, R.L., W.D. Whittier, B.G. Cassell, and J.H. Britt. 1987. Comparison of on-farm and laboratory milk progesterone assays for identifying errors in detection of estrus and diagnosis of pregnancy. J. Dairy Sci. 70:1471-1476.

Nebel, R.L., W.L. Walker, M.L. McGilliard, C.H. Allen, and G.S. Heckman. 1994. Timing of insemination of dairy cows: fixed time once daily versus morning and afternoon. J. Dairy Sci. 77:3185-3191.

Nebel, R.L. and S.M. Jobst, 1998. Evaluation of systematic breeding programs for lactating dairy cows: A review. J. Dairy Sci. 81:1169-1174.

Nebel, R.L. 2003. Components of a successful heat detection program. Adv. Dairy Technol. 15:191-203.

Peralta, O.A., R.E. Pearson, and R.L. Nebel. 2005. Comparison of three estrus detection systems during summer in a large commercial dairy herd. Anim. Reprod. Sci. 87:59-72.

Peters, J.L., P.L. Senger, J.L. Rosenberger, and M.L. O'Connor. 1984. Radiographic evaluation of bovine artificial inseminating technique among professional and herdsman-inseminators using .5- and .25-mL French straws. J. Anim. Sci. 59:1671-1683.

Pursley, J.R. 2004. Deep uterine horn AI improves fertility of lactating dairy cows. J. Dairy Sci. 87:(Suppl. 1):372(Abstr.).

Pursley, J.R. 2007. Strategies and rationale for resynchronization of ovulation in lactating dairy cows. In: Proc. Dairy Cattle Reprod. Council Annual Meeting, Denver, CO, pp. 37-43.

Reimers, T.J., R.D. Smith, and S.K. Newman. 1985. Management factors affecting reproductive performance of dairy cows in the northeast United States. J. Dairy Sci. 68:963-977.

Saacke, R.G., J.A. Lineweaver, and E.P. Aalseth. 1978. Procedures for handling frozen semen. In: Proc. Nat'l. Assn. Anim. Breeders 12th Tech. Conf. AI and Reprod., Columbia, MO, pp. 46-61.

Saacke, R.G., J.C. Dalton, S. Nadir, R.L. Nebel, and J.H. Bame. 2000. Relationship of seminal traits and insemination time to fertilization rate and embryo quality. Anim. Reprod. Sci. 60-61:663-677.

Sangsritavong, S., D.K. Combs, R. Sartori, L.E. Armentano, and M.C. Wiltbank. 2002. High feed intake increases liver blood flow and metabolism of progesterone and estradiol-17β in dairy cattle. J. Dairy Sci. 85:2831-2842.

Santos, J.E.P. 2007. Optimization tips and alternatives for timed insemination at first service. In: Proc. Dairy Cattle Reprod. Council Annual Meeting, Denver, CO, pp. 23-35.

Senger, P.L., W.C. Becker, S.T. Davidge, J.K. Hillers, and J.J. Reeves. 1988. Influence of cornual insemination on conception in dairy cattle. J. Anim. Sci. 66:3010-3016.

Senger, P.L. 1999. The luteal phase of the estrous cycle. In: Pathways to Pregnancy and Parturition, Current Conceptions, Inc., Pullman, WA, pp. 149-166.

Stevenson, J.S. 2001. Reproductive management of dairy cows in high milk-producing herds J. Dairy Sci. 84(E. Suppl.):E128-E143.

Trimberger, G.W. and H.P. Davis. 1943. Conception rate in dairy cattle from artificial insemination at various stages of estrus. Neb. Agric. Exp. Sta. Res. Bull. 129:1-14.

Trimberger, G.W. 1948. Breeding efficiency in dairy cattle from artificial insemination at various intervals before and after ovulation. Neb. Agric. Exp. Sta. Res. Bull. 153:1-26.

Walker, W.L., R.L. Nebel, and M.L. McGilliard. 1996. Time of ovulation relative to mounting activity in dairy cattle. J. Dairy Sci. 79:1555-1561.

Williams, B.L., F.C. Gwazdauskas, W.D. Whittier, R.E. Pearson, and R.L. Nebel. 1988. Impact of site of inseminate deposition and environmental factors that influence reproduction of dairy cattle. J. Dairy Sci. 71:2278-2283.

Wilmut, I. and R.H.F. Hunter. 1984. Sperm transport into the oviducts of heifers mated early in oestrus. Reprod. Nutr. Develop. 24:461-465.

Wiltbank, M.C. 2003. Novel nutritional effects on reproduction. In: Proc. 5th Ann. Intermountain Nutrition Conference, Salt Lake City, UT, pp. 127-140.

Xu, Z. Z., D.J. McKnight, R. Vishwanath, C.J. Pitt, and L.J. Burton. 1998. Estrus detection using radiotelemetry or visual observation and tail painting for dairy cows on pasture. J. Dairy Sci. 81:2890-2896.