



Investigation of Relation between the Ovulation Confirmation and Conception Rate in Dairy Cattle

Hyun-Joo Lim[†] and Ho-Beak Yoon

Dairy Science Division, National Institute of Animal Science, RDA, Cheonan 330-801, Korea

Abstract

Ensuring timely ovulation concerning the service is valuable. A satisfactory conception rate can be achieved by making sure that ovulation occurs within 7-18 hours after artificial insemination (AI). Delayed ovulation is one of the disturbances commonly encountered in repeat breeding animals. Although demanding research, many studies have not been conducted. Therefore, we aimed to examine the relation between ovulation confirmation and conception rate in dairy cattle. The research findings showed that the signs of true estrus were bred 12 hours after the onset of estrus by AI in cattle. Also, the performance of AI on ovulation was confirmed by the presence of fluctuant Graafian follicles through rectal palpation. From the results, we confirmed that cow encountered delayed ovulation were bred again. The Conception rate in cows with confirmed ovulation was 51.9%, while for those without confirmed ovulation were 33.3%. In conclusion, the results indicate that ovulation confirmation will likely increase conception rate.

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INTRODUCTION

Over the past years, the productivity of the dairy industry worldwide has been progressing through genetic improvements, by artificial insemination and embryo transfer, and improvements in feeding and housing of cows. As a result, the mean 305-day milk production per cow has increased prominently from 6,176 to 10,289 kg from 1990 to 2015 (Dairy Statistics Yearbook, 2015). On the other hand, the average calving interval of dairy cows has increased from 439.5 days to 454.4 days from 2005 to 2016 (DHI Annual Report in Korea, 2005; 2016). This decline in the reproductive performance of dairy cows has been detected not only in Korea but also worldwide (Lucy, 2001; López-Gatius, 2003; Reimers et al., 2003; Van Eerdenburg et al., 1985; Van Vliet and Van Eerdenburg., 1996; Butler and Smith., 1989; Royal et al., 2000; Butler, 1998). Recently, it has been explained that one of the main reasons for this reduction in reproduction is the weakened and shortened estrous and estrous signs (Yoshida and Nakao et al., 2005; Washburn et al., 2002). These factors might give rise to failed artificial insemination (AI) or missing of the optimal time of insemination, leading to a decline in the conception rate and a consequential increase in the calving interval.

Variation in the proportion and the error of estrus detection, may lead to inappropriate timing and poor success rates of insemination. Delayed ovulation is responsible for a high percentage of the so-called 'optimum time of insemination' failures to conceive.

Ovulation occurs about 30 hours after the onset of estrus, that is, after the end of behavioural estrus. Various factors can affect

[†] Correspondence: Hyun-Joo Lim (ORCID: 0000-0001-7059-1553)
Phone: +82-41-580-3385, Fax: +82-41-580-3419
E-mail: limhj0511@korea.kr

the actual time of ovulation concerning the estradiol peak (maximal estrus signs). Compromised luteal function due to metabolic shortages and excessive metabolic rate, deficiency of manganese (Mn), or the effects of high ambient temperature (heat stress) can lead to delay in ovulation (Kreplin and Yaremco, 1992; Corah, 1996). Also, delayed ovulations have been related to asynchrony between estrus and ovulation, asynchrony of LH peak and ovulation, or incapacity for LH release (Duchens et al., 1994; Lee et al., 1983; Duchens et al., 1995).

With the comparatively short survival time of frozen semen, the success of artificial insemination (AI) is very much reliant on the appropriate timing of insemination about the time of ovulation. The fertilization rate of the oocyte decreases significantly 8 to 12 hours, post ovulation, while insemination 25 to 40 hours before ovulation is associated with significant reduction in conception rate.

Ensuring timely ovulation concerning the service is valuable. By making sure that ovulation takes place within 7-18 hours after AI, a acceptable conception rate can be achieved (Schels and Mostafawi, 1978). Therefore, this study investigated the relationship between ovulation confirmation and conception rate on dairy cattle.

MATERIALS AND METHODS

One hundred sixty two (162) dairy cattle were used in the study. The cows were kept in free stalls in a cowhouse, with 15-20 animals per straw-bedded pen. The average age of observed cows was 4.6 ± 1.8 years old and parity was from 0 to six. The selected cows were checked for signs of estrus twice daily, once in the morning and once in the evening. Also, the cow must express behavior and physiological changes. The cows were considered to be in estrus when they stood for mounting by another cow (behavioral changes). Then we were observing the physical changes in the reproductive tract (vulvar edema and mucus discharge). Estrus was diagnosed by the absence of a corpus luteum and the presence of a large fluctuating follicle on rectal palpation. Cows expressing signs of true estrus were inseminated 12 hours after the onset of standing estrus. AI and its schedules were conducted and recorded by an inseminator. The AI was performed using frozen-thawed semen in a 0.25 ml straw containing as a minimum of 10 million motile spermatozoa from a single bull with proven fertility. Frozen semen was thawed at 37°C water bath for 30 sec and then placed into the device for AI. The AI process was conducted as follows. First, the vulva was sterilized with an antiseptic solution. Second, the device for AI (semen injector) was inserted into the vagina, and the tip of the semen injector was inserted into the external uterine orifice. Then, the instrument was held in position with one hand. Third, the other hand was inserted into the rectum to fold the cervix. The tip of the semen injector was guided into the uterus through the cervical canal by manipulating the cervix, through the rectal wall, with the hand inserted in the rectum. After that, the cows were inseminated by semen deposition into the uterine body. Afterwards, the ovary was checked using rectal palpation or ultrasonic examination a day after heat to confirm if ovulation has occurred. According to ovulation, the cow was divided into two groups consisting of ovulation group and ovulation delay group. A second insemination is performed if ovulation did not happen. If ovulation has not occurred within 24h after AI, AI should be reperfomed. If the cows did not ovulate until 48 h after AI, further observations of ovulation and estrous signs were not performed. Pregnancy diagnosis was confirmed 60 days post-breeding based on rectal palpation or ultrasonic examination (Figure 1). Transrectal ultrasonographic observations were carried out using a B-Mode ultrasound scanner (MyLab™OneVET, esaote) equipped with a 5.0-MHz linear array probe.

Data were subjected to analysis of variance (ANOVA) using the Statistical Analysis System (SAS Institute, Cary, NC, USA).

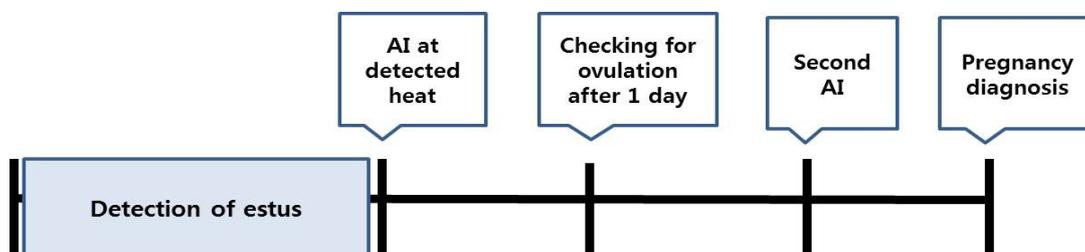


Figure 1. Scheme of reproduction

RESULT

Figure 2 presents an image sequence of ovulation in dairy cattle with natural estrus. Ovulation was checked on the basis of the following findings: the follicles were indistinct or indefinite because of the disappearance of the follicular fluid, usually revealing a depression, as decided on palpation at the former site of the follicle, and a clear decrease in the follicle diameter by rectal examination and transrectal ultrasonography. The pre-ovulatory follicles were observed at the onset of estrus (A). Ovulation occurred after detection of estrus with apparent follicle disappearance (B). Delayed ovulation was observed with evident follicle appearance after the onset of estrus (C).

Ovulation group for the dairy cattle herd represents 83% of the total dairy cattle (162). Ovulation delay group for the dairy cattle herd represents 17% of the total dairy cattle. Table 1 presents the conception rate of cows about ovulation confirmation. The pregnancy rate of the initial AI was 33.33%, while the pregnancy rate for the second insemination was 51.85%.

Table 2 shows the result of analysis of variance. The data would yield a *p*-value of 0.0686, which is not significant at the 0.05 level. But value of *P* was considered to be statistically close.

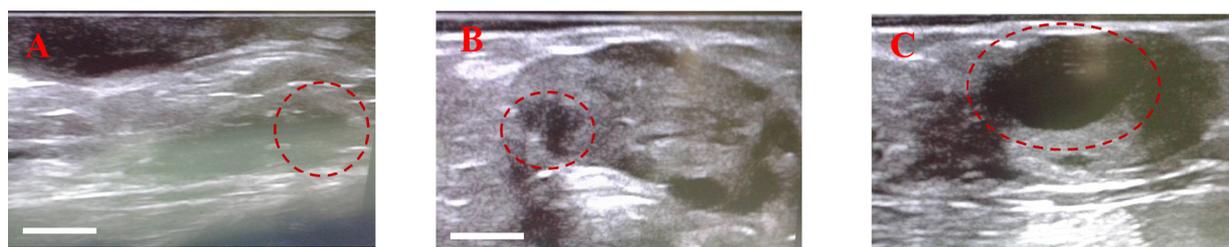


Figure 2. Ovary ultrasound images. (A) A large pre-ovulatory follicle in estrus. Note the clear spherical shape indicating that ovulation is not that imminent; (B) After ovulation induction. Note the rupture of the follicle wall; (C) Delayed ovulation. Note the intact of the follicle wall. Scale bars = 1 cm.

Table 1. Cow's reproduction according to services times by ovulation confirmation

| | Services times | |
|-----------------------|----------------|----------------|
| | First service | Second service |
| Number of cows, n | 135 | 27 |
| Conception rate(%), n | 33.33% (45) | 51.85% (14) |

Table 2. Analysis of variance

| Source | DF | SS | MS | <i>p</i> -value |
|--------|-----|--------|-------|-----------------------|
| Group | 1 | 0.772 | 0.772 | 0.0686 ^{n.s} |
| Error | 160 | 36.741 | 0.230 | |
| Total | 161 | 37.512 | | |

* DF : the degrees of freedom

SS : the sum of squares

MS : the mean sum of squares

DISCUSSION

The timing of insemination is vital for a successful AI. To achieve maximum fertility, the sperm must reside in the female reproductive tract for a maximum period before ovulation for sperm transport to the ampullary (Senger, 1999). Also, bovine sperm and oocytes have limited life spans, which limit the amount of time before or after ovulation that insemination can lead to

conception. Artificial insemination can only be successful if the sperm is deposited in the female reproductive organ at an appropriate time about ovulation (Nalbandov and Casida, 1942). Saacke et al. (2000) examined embryo quality about the time of insemination and found a shift from high-quality embryos accomplished by inseminations at the onset of estrus to low-quality embryos attributing to inseminations at 24h following onset of estrus. Therefore, the timing of insemination is a balance between the time of ovulation, the time needed for sperm transport and capacitation, and longevity of sperm and oocyte.

Delayed ovulation is one of the leading causes of repeat breeding in cattle. Repeated insemination at 6 hours interval from onset of behavioural estrus to spontaneous ovulation yielded a higher pregnancy rate in repeat breeding cattle as compared to control (Singh et al., 2005). Delayed ovulators have a longer interval from onset of estrus to ovulation (Bage et al., 2002; Singh et al., 2005). Therefore the importance of timing of insemination on ovulation has repeatedly been accentuated to ensure fertilization (Rodriguez-Martinez, 2001). According to Trimberger and Davis (1943), the optimum time for AI is from the middle of estrus up to six hours after the end of estrus.

This study was conducted to evaluate the usefulness of ovulation confirmation. Environmental and stress factors promote ovulation failure, which is regarded to be a major factor of infertility in dairy cattle. For instance, the incidence of ovarian dysfunction in high producing dairy cattle has recently increased alongside milk production (Wiltbank et al, 2002; Dobson et al., 2003, Hunter, 2003). As the ovulation state changes, practical reproductive management of a dairy herd must evolve to adapt to the new technologies. Judgement of ovulation is recommended 24 h after AI, and if ovulation has not happened by then, conducting AI again could ameliorate the chances of conception.

In this study, we conducted to confirm the ovulation after the first insemination. Numerous inseminations were selected for ovulation confirmation. A second insemination was performed for ovulation that did not occur. It may be concluded that repeated inseminations in dairy cattle with delayed ovulation are useful by checking ovulation during estrus.

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REFERENCES

- Bage R, Gustafsson H, Larsson B, Forsberg M and Rodriguez-Martinez H. 2002. Repeat breeding in dairy heifers: follicular dynamics and estrous cycle characteristics in relation to sexual hormone patterns. *Theriogenology* 57:2257-2269.
- Corah L. 1996. Trace mineral requirements of grazing cattle. *Anim. Feed Sci. Tech.* 59:61:70.
- Duchens M, Forsberg M, Edqvist L, Gustafsson H and Rodríguez-Martínez H. 1994. Effect of induced suprabasal progesterone levels around estrus on plasma concentrations of progesterone, estradiol-17beta and LH in heifers. *Theriogenology* 42:1159-1169.
- Duchens M, Maciel M, Gustafsson H, Forsberg M, Rodríguez-Martínez H and Edqvist L. 1995. Influence of perioestrous suprabasal progesterone levels on cycle length, oestrous behaviour and ovulation in heifers. *Anim. Reprod. Sci.* 37:95-108.
- Kreplin C and Yaremicio B. 1992. Effects of nutrition on beef cow reproduction. *Agdex.* 420/51:1.
- Lee CN, Maurice E, Ax RL, Pennington JA, Hoffman WF and Brown MD. 1983. Efficacy of gonadotrophin-releasing hormone administered at the time of artificial insemination of heifers and postpartum and repeat breeder dairy cows. *Am. J. Vet. Res.* 44:2160-2163.
- Nalbandov A and Casida LE. 1942. Ovulation and its relation to estrus in cows. *J. Anim. Sci.* 1:189-198.
- Rodríguez-Martínez H. 2001. Oviduct function in cows and pigs: With special reference to sperm capacitation. *Asian-Aust. J.*

- Anim. Sci. 14:28 - 37.
- Saacke RG, Dalton JC, Nadir S, Nebel RL and Bame JH. 2000. Relationship of seminal traits and insemination time to fertilization rate and embryo quality. Anim. Prod. Sci. 60:663-677.
- Senger PL. 1999. Pathways to Pregnancy and Parturition. 1st rev. ed. Current Conceptions Inc., Pullman, WA. 116-128.
- Singh B, Savavia F, Bage R and Rodríguez-Martínez H. 2005. Pregnancy rates in repeat-breeder heifers following multiple artificial inseminations during spontaneous oestrus. Acta Vet Scand 46:1-12.
- Trimberger GW, and Davis HP. 1943. Conception rate in dairy cattle by artificial insemination at various stages of estrus. Nebraska Agric. Exp. Stn. Res. Bull. 129:1-14.
- Dairy Statistics Yearbook. Korea Dairy Committee. 2015. pp101. (In Korean)
- DHI Annual Report in Korea. NongHyup Agribusiness Group Inc. 2005; 2016. (In Korean)
- Lucy MC. 2001. Reproductive loss in high-producing dairy cattle: where will it end?. J Dairy Sci. 84:1277-1293.
- López-Gatius F. 2003. Is fertility delining in dairy cattle? A RETROSECTIVE STUDY IN NORTHEASTERN Sparin. Theriogenology. 60:89-99.
- Reimers TJ, Smith RC, Newman SK. 1985. Management factors affecting reproductive performance of dairy cows in the northeastern United States. J Dairy Sci. 68:963-972.
- Van Eerdenburg FJCM, Loeffler HSH, van Vliet JH. 1996. Detection of oestrus in dairy cows: a new approach to an old problem. Vet Q. 18:52-54.
- Van Vliet JH, Van Eerdenburg FJCM. 1996. Sexual activities and oestrus detection in lactating Holstein cows. Appl Anim Behav Sci. 50:57-69.
- Butler WR, Smith RC. 1989. Interrelationships between energy balance and postpartum reproductive function in dairy cattle. J Dairy Sci. 72: 767-783.
- Royal MD, Darwash AO, Flint APF, Webb R, Woollians JA, Lamming GE. 2000. Declining fertility in dairy cattle: changes in traditional and endocrine parameters of fertility. Anim Sci. 70:487-501.
- Butler WR. 1998. Review: effect of proein nutrition on ovarian and uterine physiology in dairy cattle. J Dair Sci. 81:2533-2539.
- Yoshida C, Nakao T. 2005. Some characteristics of primary and secondary oestrous sings in high-producing dairy cows. Reprod Domest Anim. 40:150-155.
- Washburn SP, Silvia WJ, Brown CH, McDaniel BT, McAllister AJ. 2002. Trends in reproductive performance in Southeastern Holsein and Jersey DHI herds. J Dairy Sci. 85:244-251.
- Schels HF, Mostafawi D. 1978. The effect of GnRH on the pregnancy rate of artificially inseminated cows. Vet. Rec. 103:31.
- Wiltbank MC, Gümen A, Sartori R. 2002. Physiological classification of anovulatory conditions in cattle. Theriogenology. 57:21-52.
- Dobson H, Ghuman S, Prabhakar S, Smith R. 2003. A conceptual model of the influence of stress on female reproduction. Reproduction. 125:151-63.
- Hunter RHF. 2003. Physiology of the Graafian Follicle and Ovulation. Cambridge: Cambridge University Press.