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Factors Affecting Conception Rates Following Artificial Insemination or Embryo Transfer in Lactating Holstein Cows

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ABSTRACT

The objective of this study was to evaluate the factors that may affect conception rates (CR) following artificial insemination (AI) or embryo transfer (ET) in lactating Holstein cows. Estrous cycling cows producing 33.1 ± 7.2 kg of milk/d received $PGF_{2\alpha}$ injections and were assigned randomly to 1 of 2 groups (AI or ET). Cows detected in estrus (n = 387) between 48 and 96 h after the $PGF_{2\alpha}$ injection received AI (n = 227) 12 h after detection of estrus or ET (n = 160) 6 to 8 d later (1 fresh embryo, grade 1 or 2, produced from nonlactating cows). Pregnancy was diagnosed at 28 and 42 d after estrus, and embryonic loss occurred when a cow was pregnant on d 28 but not pregnant on d 42. Ovulation, conception, and embryonic loss were analyzed by a logistic model to evaluate the effects of covariates [days in milk (DIM), milk yield, body temperature (BT) at d 7 and 14 post-AI, and serum concentration of progesterone (P4) at d 7 and 14 post-AI] on the probability of success. The first analysis included all cows that were detected in estrus. The CR of AI and ET were different on d 28 (AI, 32.6% vs. ET, 49.4%) and 42 (AI, 29.1% vs. ET, 38.8%) and were negatively influenced by high BT (d 7) and DIM. The second analysis included only cows with a corpus luteum on d 7. Ovulation rate was 84.8% and was only negatively affected by DIM. Conception rates of AI and ET were different on d 28 (AI, 37.9% vs. ET, 59.4%) and 42 (AI, 33.8% vs. ET, 46.6%) and were negatively influenced by high BT (d 7). The third analysis included only ovulating cows that were 7 d postestrus. Conception rates of AI and ET were different on d 28 (AI, 37.5% vs. ET, 63.2%) and 42 (AI, 31.7% vs. ET, 51.7%) and were negatively influenced by high BT (d 7). There was a positive effect of serum concentration of P4 and a negative effect of milk production on the probability of conception for the AI group but not for the ET group. The fourth analysis was embryonic loss (AI, 10.8% vs. ET, 21.5%). The transfer of fresh embryos is an important tool to increase the probability of conception of lactating Holstein cows because it can bypass the negative effects of milk production and low P4 on the early embryo. The superiority of ET vs. AI is more evident in high-producing cows. High BT measured on d 7 had a negative effect on CR and embryonic retention.
Key words: embryo transfer, artificial insemination, conception rate, dairy cow

INTRODUCTION

Reproductive efficiency in lactating dairy cows has declined and has been associated with the increase in average milk production (Lucy, 2001; Washburn et al., 2002). Lactating dairy cows have increased metabolism because of milk production that decreases the ability to dissipate heat (Kadzere et al., 2002). The negative effects of high milk production on oocyte quality, fertilization, and early embryonic development are exacerbated by heat stress (Hansen and Arechiga, 1999). Oocytes and embryos at early stages are extremely sensitive to heat stress. High environmental temperatures reduce the rate at which embryos progress during the development period (Ealy et al., 1993). Embryos 3 d after conception and older are less sensitive to heat stress (Ealy et al., 1993; Hansen and Arechiga, 1999). Sartori et al. (2002) reported that lactating cows had a greater increase in body temperature (**BT**) in response to an increase in environmental temperature than heifers and that the reproductive performance of heifers did not change during the summer. They recovered embryos and oocytes from Holstein cows and heifers during the summer, 6 d after observation of estrus, and verified that the fertilization rate was greater in heifers than in lactating cows. The percentage of good quality embryos was also greater in heifers than in lactating cows. Differences in fertilization rate between lactating and nonlactating cows in winter were not found but the percentage of good quality embryos was greater in nonlactating cows than in lactating cows.

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Progesterone controls LH pulsatility (Bergfeld et al., 1996), follicular dynamics (Stock and Fortune, 1993), the uterine environment (Thatcher et al., 2001; Green et al., 2005), and embryonic development (Mann and Lamming, 2001). High-producing dairy cows have lower plasma concentrations of progesterone (**P4**; Vasconcelos et al., 1999), perhaps because of the increase in DMI, which increases liver blood flow and the metabolic clearance rate of P4 (Sangsritavong et al., 2002; Vasconcelos et al., 2003). There is a positive correlation between DMI and milk production in lactating dairy cows (Harrison et al., 1990) and an inverse relationship between feed intake and peripheral plasma concentrations of P4 (Vasconcelos et al., 2003). Stronge et al. (2005) demonstrated that low P4 between d 5 and 7 after AI was associated with low fertility in dairy cows. Mann et al. (2006) observed that P4 supplementation 5 d after AI resulted in better embryonic development. These data indicate that the concentration of P4 is important during the first days after insemination and it may be one of the factors that determines the success or failure of pregnancy in lactating dairy cows.

Embryo transfer has been used as a method to attenuate some detrimental effects of heat stress on fertility (Putney et al., 1989; Drost et al., 1999). Putney et al. (1989) transferred fresh embryos produced in heifers to lactating Holstein cows in the summer and observed that there was an increase in conception after embryo transfer (**ET**) relative to AI when pregnancy was diagnosed within 45 d. Drost et al. (1999) compared frozen-thawed embryos produced in vivo with AI during the summer. Ovulation was determined by the presence of CL on cows that received an embryo and by concentrations of P4 on d 7 in AI cows. There was an increase in conception rate (**CR**) when embryos were used. No differences were detected by Sartori et al. (2006) for CR after AI or after ET.

The hypothesis for this study was that the transfer of fresh embryos would increase the CR of lactating Holstein cows because the embryo bypasses the deleterious effects caused by milk production and heat stress during the first 7 d after insemination. Objectives of this study were to evaluate factors that may alter CR after AI or ET in lactating Holstein cows throughout 1 yr.

MATERIALS AND METHODS

Location and Period

The experiment was conducted from October 2003 to September 2004 at a commercial dairy farm located in Descalvado, São Paulo State, Brazil; latitude S 21°54′05″, longitude W 47°37′26″, and altitude 648 m.

Animals

Multiparous lactating Holstein cows (n = 1,025) at 55 to 547 DIM and 33.1 ± 7.2 kg of milk production per day were used for 12 mo from a herd consisting of 1,200 lactating cows. Animals were maintained in a free-stall barn with access to an adjoining sod-based area. Barns were cooled by intermittent sprinkling and forced ventilation to minimize the effects of heat stress. Cows were fed ad libitum with a TMR based on corn silage, fresh coast-cross grass (*Cynodon dactylon*), corn, cottonseed, soybean flour, minerals, and vitamins balanced to meet nutritional requirements for lactating dairy cows (NRC, 2001). Cows were milked 3 times a day in a side-by-side milking system. Daily milk yield for each cow was recorded automatically.

Reproductive Management, AI, and ET

Nonpregnant cows with more than 55 DIM were evaluated monthly. When a corpus luteum (**CL**) was palpated per rectum, cows were treated with $PGF_{2\alpha}$ (500 μg of cloprostenol, i.m.; Ciosin, Schering-Plough, Cotia, SP, Brazil), blocked by DIM and number of previous AI, and randomly assigned to receive AI or ET (Figure 1). Estrus was detected by visual observation twice daily. Cows that were standing to be mounted by other females were considered in estrus. Cows that showed estrus 48 to 96 h after $PGF_{2\alpha}$ injection received AI 12 h after detected estrus (AI group) or 1 embryo 6, 7, or 8 d after detected estrus (ET group).

Artificial insemination was performed by a single experienced technician 12 h after estrus detection. Semen from 4 sires was used. On d 7, the AI cows were evaluated by ultrasound (Aloka SSD, 500 V equipped with a 5-MHz linear transrectal probe; Aloka Co. Ltd., Tokyo, Japan) to determine the presence of a CL.

Nonlactating Holstein cows served as embryo donors (n = 10). Donors received a P4-releasing intravaginal device (CIDR; 1.9 g P4; Pfizer, Paulina, SP, Brazil) and 2 mg, i.m., of estradiol benzoate 1 mg/mL; Index, Sao Paulo, SP, Brazil) on d –8 (8 d before AI). Superstimulatory treatments were initiated on d –4, with decreasing doses of FSH (Pluset, Calier, Brazil) given twice daily, i.m., over 4 d, totaling 350 IU. Donors received $PGF_{2\alpha}$ treatment on the morning of d –2 and the P4 devices were removed on the morning of d –1. Donors also received 200 μg of GnRH i.m. (Fertagyl, Intervet, Sao Paulo, SP, Brazil) on the morning of d 0 and were inseminated without estrus detection 12 and 24 h later. Semen from 4 sires was used (same sires as the AI group). Collection of ova or embryos was performed nonsurgically on d 7. Embryos were evaluated under a stereomicroscope and placed in TQC holding solution (Nutricell, Campinas, SP, Brazil) and maintained at ambient tem-