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Reducing the duration between gonadotropin-releasing hormone (GnRH) and prostaglandin $F_{2\alpha}$ treatment in the Ovsynch protocol to 6 days improved ovulation to second GnRH treatment, but inclined to reduce fertility

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ABSTRACT

The aim of this study was to test Ovsynch (OVS) versus modified OVS (decreasing the interval between first GnRH and $PGF_{2\alpha}$ to 6 d) protocols on pregnancy per artificial insemination (AI) and OVS outcomes in cyclic dairy cows. Cyclic cows (n = 920) were assigned to 1 of 2 groups: the OVS7 group (n = 459) received the OVS protocol [GnRH treatment, $PGF_{2\alpha}$ treatment 7 d later, a second GnRH (GnRH2) treatment 56 h later, and timed AI (TAI) 16 to 18 h after the GnRH2 treatment], and the OVS6 group (n = 461) received a modified OVS protocol, in which the interval between the first GnRH and $PGF_{2\alpha}$ was decreased to 6 d (GnRH treatment, $PGF_{2\alpha}$ treatment 6 d later, GnRH2 treatment 56 h later, and TAI 16 to 18 h after the GnRH2 treatment). The response to the first GnRH of OVS was similar between OVS7 (54.5%, 250/459) and OVS6 (54.2%, 250/461) groups. The ovulatory response to GnRH2 of OVS was higher in OVS6 (91.3%, 421/461) than OVS7 (84.5%, 388/459). The follicle size (mean ± standard error of the mean) at the time of TAI was smaller in OVS6 (15.23 ± 0.11 mm) than OVS7 (16.04 ± 0.11 mm). When all cows were evaluated, the pregnancy per AI at 31 d tended to be lower in OVS6 (38.0%, 175/461) than in OVS7 (43.8%, 201/459). Moreover, the pregnancy per AI at 31 d was lower in OVS6 (40.9%, 172/421) compared with OVS7 (50.3%, 195/388) in synchronized cows. In conclusion, although the modified OVS protocol decreased the follicle size at the time of AI and increased the ovulatory response to GnRH2 of OVS, it unexpectedly reduced fertility in cyclic lactating dairy cows.

Key words: cyclic cow, gonadotropin-releasing hormone, Ovsynch, prostaglandin $F_{2\alpha}$

INTRODUCTION

Milk yield and fertility are important factors that affect the productivity and profitability of dairy herds. A high milk yield results in decreasing the duration of estrus, ovulation without signs of estrus, and anovulatory conditions that are directly related to fertility (Wiltbank et al., 2006, 2008). Thus, the basic reproductive management tool, estrus detection followed by AI, has lost its effectiveness in many dairies, and synchronization protocols, including the use of hormones that regulate and control estrous cycles and ovulation, have become common in practice (Pursley et al., 1995; Rabiee et al., 2005; Souza et al., 2008). The Ovsynch (**OVS**) protocol is an effective worldwide timed AI (**TAI**) program that includes administrations of GnRH 7 d before and 48 h after $PGF_{2\alpha}$ and TAI 16 to 18 h after the second administration of GnRH (Pursley et al., 1995). After the development and common usage of the OVS protocol, many studies have focused on improving the pregnancy rates achieved with OVS, with modifications intending to optimize outcomes of the OVS protocol. These attempts include many different modifications of the OVS protocol, such as performing TAI at the time of second GnRH (Cosynch; Small et al., 2001), using double $PGF_{2\alpha}$ administrations before the protocol (Presynch; Moreira et al., 2001), replacing the second GnRH administration with estradiol (Heatsynch; Pancarci et al., 2002; Stevenson et al., 2004), supplementing an exogenous progesterone source between the first GnRH and $PGF_{2\alpha}$ treatments (El-Zarkouny et al., 2004; Stevenson et al., 2006), and replacing the first GnRH treatment of OVS with human chorionic gonadotropin (Keskin et al., 2010).

The OVS protocol is a TAI program that is able to synchronize new follicular wave emergence in cows that respond to the first GnRH of the protocol and ovulation of the dominant follicle from the new wave. The cycle of development of a dominant follicle in the course of a follicular wave goes through a selection, dominance,

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and atresia or ovulation phase (Matton et al., 1981). The duration between selection and ovulation or atresia phases is defined as the dominance period of the dominant follicle. Previous studies reported that pregnancy (Mihm et al., 1994; Bleach et al., 2004) and embryo quality (Cerri et al., 2009) were inversely related to the length of follicle dominance. For example, the prolonged dominance of the ovulatory follicle is associated with decreased fertility, which is reduced as the duration of dominance increases from 4 to 8 d (Mihm et al., 1994). Similarly, Bleach et al. (2004) demonstrated that cows that had an interval from follicular emergence to estrus that was 1 d shorter were more likely to become pregnant. Because previous studies (Mihm et al., 1994; Bleach et al., 2004; Cerri et al., 2009) indicated that the extended period of dominance of the ovulatory follicle might have a negative effect on pregnancy, reducing the interval from follicular emergence to TAI in an OVS protocol might be considered as one of the modifications that need to be evaluated. Likewise, Santos et al. (2010) evaluated reducing the interval between GnRH and $PGF_{2\alpha}$ to 5 d in a Cosynch protocol (5-d TAI protocol) in dairy cows, and achieved improved fertility with a shorter duration of follicle dominance. However, the 5-d TAI protocol requires a double dose of $PGF_{2\alpha}$ (at least 7 h apart; Kasimanickam et al., 2009; Santos et al., 2010) because the new corpus luteum (**CL**) induced by the first GnRH of the protocol is not responsive to $PGF_{2\alpha}$ during the first 5 d after ovulation (Tsai and Wiltbank, 1998; Miyamoto et al., 2005). A recent study (Ribeiro et al., 2012) that aimed to overcome this impractical issue of the 5-d TAI program reported that using a double dose of $PGF_{2\alpha}$ as a single administration (1 mg per cow) on d 5 resulted in inadequate CL regression, whereas dividing the same dose into 2 administrations 24 h apart (0.5 mg/cow at each) resulted in success.

Thus, the current study aimed to evaluate a potentially more practical modification of the OVS protocol by reducing the interval between the first GnRH and $PGF_{2\alpha}$ to 6 d (6-d TAI program), in which only 1 dose of $PGF_{2\alpha}$ administration should be sufficient, and compare all OVS outcomes (preovulatory follicle size, responses to $PGF_{2\alpha}$, and GnRH administrations and pregnancy rates) in OVS with a modified OVS protocol in cyclic lactating dairy cows.

MATERIALS AND METHODS

Animals

Cyclic lactating dairy cows (n = 920) from a commercial dairy farm located in the South Marmara region (Bursa, Turkey) were enrolled in the present study. The cows were housed in freestall barns with self-catching headlocks, and all barns had fans and sprinklers that were activated during the hotter months of the year. All cows were grouped according to their milk production and milked 3 times daily at approximately 8-h intervals. The mean milk production of the herd was 9,880 ± 69.7 kg (305 d) and 32.4 kg/d per cow. The cows had free access to water and were fed complete mixed rations according to the NRC (2001) recommendations. The daily milk yield, reproductive health, and management records for each cow were collected from Alpro 2000 software (DeLaval, Tumba, Sweden). The average milk production for each cow was recorded from the 7 d before and after AI. Body condition scores were determined for all cows at the beginning of the study using a 5-point (1 = thin to 5 = fat) scoring system (Ferguson et al., 1994). All protocols involving cows used in this research were approved by the Lalahan Livestock Central Research Institute Animal Care Committee (Lalahan, Ankara, Turkey).

Treatment Groups

After determination of the cyclicity of the cows by ultrasonographic examination, cyclic cows (n = 920) were randomly assigned to 1 of 2 groups. In the **OVS7** group (n = 459), the cows received the OVS protocol: a GnRH treatment (**GnRH1**; buserelin acetate, 10 μg, i.m.; Receptal; Intervet, Istanbul, Turkey) followed by $PGF_{2\alpha}$ (cloprostenol, 500 μg, i.m.; Estrumate; CevaDIF, Istanbul, Turkey) 7 d later. A second GnRH treatment (**GnRH2**) was administered 56 h after $PGF_{2\alpha}$, and all cows were inseminated at a fixed time (TAI) 16 to 18 h after GnRH2 treatment. In the **OVS6** group (n = 461), the cows received a modified OVS protocol (6-d TAI program) in which $PGF_{2\alpha}$ was administered 6 d after GnRH1. The GnRH2 administration was performed 56 h after $PGF_{2\alpha}$, and all cows were inseminated at TAI 16 to 18 h after GnRH2, similar to the OVS7 group.

Ultrasonographic Examinations

All transrectal ultrasonographic evaluations in this study were performed with a Honda HS 2000 ultrasound scanner equipped with a 7.5-MHz transducer (Honda, Tokyo, Japan). Ultrasonographic examinations were performed at the time of GnRH1 administration on d 0 for OVS7 and d 1 for OVS6 (to determine the size of the largest follicle and presence of the CL in the ovaries), at the time of $PGF_{2\alpha}$ administration on d 7 (to determine ovulation in response to GnRH1), at the time of TAI on d 10 (to determine the presence and size of the ovulatory follicle), and 7 d after TAI on d 17 (to determine ovulation in response to GnRH2).