MJAVL
Manas Journal of Agriculture Veterinary and Life Sciences
ISSN 1694-7932 | e-ISSN 1694-7932
Volume 12 (Issue II) (2022) Pages 98-105 https://doi.org/10.53518/mjavl.1165972

Effect of Season on Ovulatory Response and Reproductive Performance in Noncyclic Lactating Dairy Cows Synchronized with Ovsynch

Barış GÜNER[1a*], Ebru KARAKAYA BİLEN[2b]

[1]Balıkesir University, Faculty of Veterinary, Department of Obstetrics and Gynecology, Balıkesir, TÜRKİYE
[2]Siirt University, Faculty of Veterinary, Department of Obstetrics and Gynecology, Siirt, TÜRKİYE

[a]*https://orcid.org/ 0000-0001-6414-6752*, [b]*https://orcid.org/ 0000-0003-4837-1858*
*Corresponding author: baris.guner@balikesir.edu.tr

ABSTRACT

The aim of this study was to compare the ovulatory response and reproductive performance after Ovsynch protocol in noncyclic dairy cows during the warm (n = 43) and cool seasons (n = 70). Noncyclic cows (n=113) received Ovsynch protocol; GnRH1 (d 0); $PGF_{2\alpha}$ (d 7); GnRH2 (d 9); FTAI (d 10). Ultrasonographic examinations were performed to determine the preovulatory follicle size (d 0, d 10) and ovulatory response to the GnRH1 (d 7) and GnRH2 (d 17). Follicle size at the onset of Ovsynch was not different in cool season (20.8 ± 0.9 mm) compared to that in warm season (19.1 ± 1.1 mm, P > 0.05). The percentage of small-sized follicles at the onset of Ovsynch was higher (P < 0.05) in warm season (37.2%) than cool season (18.6%). However, follicle size at FTAI was similar (P > 0.05) between warm (15.3 ± 0.3 mm) and cool (15.8 ± 0.3) seasons. Ovulatory response to the GnRH1 and the GnRH2 of Ovsynch were not different during the warm (90.7%, 83.7%) and cool seasons (81.4%, 87.1%), respectively (P > 0.05). Pregnancy rate was also similar (P > 0.05) during the warm (34.9%) and cool seasons (35.7%). Cows that had large-sized follicles at the onset of Ovsynch had a tendency (P = 0.08) for lower pregnancy rate in warm season (12.5%) compared to cool season (52.6%). It was concluded that season did not affect the ovulatory response to the first GnRH and pregnancy rate in noncyclic dairy cows that were synchronized with Ovsynch.

ARTICLE INFO

Research article
Received: 23.08.2022
Accepted: 06.10.2022

Keywords:
Cool, Noncyclic, Ovsynch, Warm, Season

INTRODUCTION

Global warming causes prolongation of summer periods and heat stress is one of the major reasons for economical losses in the global dairy industry (Nanas et al. 2021). Heat stress in the warm season not only impairs the general physiology, welfare, and production but also leads to a reduction in reproductive efficiency (De Rensis et al. 2015, Thorton et al 2021). Holstein cows are more susceptible to heat stress due to higher milk yield compared to other breeds such as Jersey and Brown Swiss (El-Tarabany and El-Tarabany 2015; Thorton et al. 2021). Besides, as in higher milk yield, heat stress negatively contributes the low fertility (20 to 30%) by increasing the incidence of anovulatory or noncyclicity condition and disruptions in cyclic activity in cows (De Rensis et al. 2017; Santos et al. 2016; Schüller et al. 2014). Previous studies reported that incidence of noncyclicty ranged from 18 to 29% (Bisinotto et al. 2010; Cartmill et al. 2001; Gumen et al. 2003; Moreira et al. 2001; Pursley et al. 2001). Although the noncyclic rate was reported to be very low (1.2%) in the cool season in a large-scale retrospective study, this rate increased approximately 10 fold (12.9%) and the pregnancy rate decreases approximately 2 fold (27% vs. 44%) in the warm season (López-Gatius 2003).

In addition to disruption of cyclic activity, the intensity of estrus sign and estrus detection failure increases due to higher milk yield in heat-stressed cows during warm season (De Rensis and Scaramuzzi 2003; Santos et al. 2009; Schüller et al. 2017). Besides, heat stress causes more frequent ovulation failure compared to the cool season in cows (García-Ispierto et al. 2019; Hansen 2019). Therefore, an increased incidence of anovular or noncyclicity leads to low fertility in cows that are exposed to heat stress (Santos et al. 2016). In ovulation synchronization methods such as

To Cite: Güner B, Bilen EK. 2022. Effect of Season on Ovulatory Response and Reproductive Performance in Noncyclic Lactating Dairy Cows Synchronized with Ovsynch, MJAVL Sciences. 12 (2) 98-105

Ovsynch synchronize follicular development, ovulation, and regression of the corpus luteum regardless of estrus signs (Pursley et al. 1997). Ovsynch effectively synchronized ovulation in both cyclic and noncyclic cows and is commonly used protocol to prevent ovulation failure (García-Ispierto et al. 2019; Gumen et al. 2003; Hansen 2019; Keskin et al. 2010). Thus, ovulatory response to the first GnRH (GnRH1) is a critical factor to synchronize the follicular development and obtain higher fertility in Ovsynch (Bello et al. 2006). The aim of this study was to compare the ovulatory response and pregnancy rates of noncyclic dairy cows that synchronized with Ovsynch during warm and cool seasons.

MATERIALS AND METHOD

Cows, Housing and Management

Lactating Holstein cows were enrolled the study (n=113) on a commercial dairy farm located in the South Marmara region, Bursa, Turkey. The study was conducted on 43 noncyclic cows (22 primiparous and 21 multiparous cows) during warm season and 70 noncyclic cows (37 primiparous and 33 multiparous cows) during cool season. Cows were housed in free stall barns with self-catching headlocks, and all barns had fans and sprinklers that were activated during the warm season (June-September) of the year. This cooling approach relied on sprinkles spraying water for 30 seconds with the application of 0.05 inches of water per cycle, followed by evaporation from the skin with 5 minutes of air from the fans. All cows were grouped according to their milk production and were milked three times daily at approximately 8 h intervals. Cows had free access to water and were fed complete mixed rations according to National Research Council recommendations (NRC 2001). Daily milk yield (DIM), reproductive health, and management records for each cow were monitored with a herd management system (Alpro 2000, DeLaval, Sweden).

Study Design

Body condition score (BCS) was determined using a 5-point (1= thin to 5 = fat) scoring system at the onset of Ovsynch in all cows (Ferguson et al. 1994). Average milk production for each cow was recorded from the 7 d before to the 7 d after AI. Seasons were defined as warm (June to August) and cool season (September to May). Daily temperature and humidity records were taken in order to evaluate heat stress (Whether online, 2019). Temperature Humidity Index (THI) was calculated by the formula according to NRC (1971). THI was also determined in the 74–80 range in warm season. Ultrasonographic examinations of ovaries were performed to determine the noncyclic cows at 7 d intervals (d -7 and d 7). The cow had no luteal tissue such as corpus luteum or luteal cyst and was described as non-cyclic. Following the description of noncyclic cows, Ovsynch was initiated with a GnRH treatment (buserelin acetate, 10 µg i.m., MSD, USA) followed by $PGF_{2\alpha}$ (cloprostenol, 500 µg i.m., Egevet, Turkey) 7 d later. A second GnRH was administered 56 h after the $PGF_{2\alpha}$, and all cows were inseminated at a fixed time (FTAI; 16 to 18 h) after the second GnRH. All cows were inseminated by technicians with frozen-thawed conventional semen.

Ultrasonographic examinations were also performed to determine the preovulatory follicle at the onset of Ovsynch. Preovulatory follicle sizes were classified as small (8-15 mm), medium (16-24 mm), and large (>24 mm) at the onset of Ovsynch (Gumen et al. 2003). Following the first GnRH injection (d 0), disappearance of preovulatory follicle and formation of corpus luteum at the $PGF_{2\alpha}$ (d 7) were defined as an ovulatory response to the GnRH1. Preovulator follicle size was determined at the time of FTAI. Cows were reexamined to determine ovulatory response to the GnRH2 7 d after FTAI (d 17). Pregnancy diagnosis was performed 31 d and 62 d after FTAI using transrectal ultrasonography.

Statistical Analysis

The SPSS 23.0 software (SPSS Inc, USA) was used for all statistical procedures. Days in milk, milk yields, body condition scores, and follicle size between groups (warm and cool seasons) were analyzed with independent samples t-test. The chi-square was used to compare all proportional data and the results were interpreted by taking Pearson chi-square or Fisher exact test. Statistical significance level was considered at $P \leq 0.05$ and statistical tendencies were defined at $0.05 < P < 0.10$.

RESULTS

There was no difference in average lactation number (1.77 ± 0.1 vs. 1.88 ± 0.1), milk yield (35.8 ± 1.1 kg/d vs. 36.1 ± 1.4 kg/d), BCS (2.65 ± 0.04 vs. 2.69 ± 0.05), and number of insemination (1.16 ± 0.2 vs. 1.23 ± 0.2) in cool and warm season, respectively ($P > 0.05$). However, days in milk (DIM) were different ($P < 0.05$) between warm (95.8 ± 8.9 d) and cool seasons (133.2 ± 9.1 d). In Table 1, follicle size at the onset of Ovsynch for warm season is 19.1 ± 1.1 mm, while cool season is 20.8 ± 0.9 mm. According to classification for follicle size, distributions of preovulatory follicle size groups (small, medium, large) were 37.2%, 44.2%, 18.6% in warm season and 18.6%, 54.3%, 27.1% in cool season. The percentage of small follicle size was statistically higher in warm season compared to that in cool season ($P < 0.05$). The results in Tables 2 and 3 are grouped as small, medium, and large for follicle size at the onset of Ovsynch.

However, there was no difference in preovulatory follicle size among small (13.5 ± 0.5 mm vs. 12.9 ± 0.5 mm), medium (18.5 ± 0.3 mm vs. 18.9 ± 0.5 mm), and large (31.4 ± 1.6 mm vs. 31.6 ± 2.0 mm) follicular size groups at the onset of Ovsynch according to warm (Table 2) and cool (Table 3) seasons, respectively (P >0.05). At the time of FTAI, preovulatory follicle size was not different (P >0.05) in warm (15.3 ± 0.3 mm) and cool (15.8 ± 0.3 mm) seasons (Table 1).

Table 1. *Ovulatory response, follicle size, and pregnancy rate in noncyclic cows that received Ovsynch during warm and cool season*

Parameters	**Warm season (n=43)**	**Cool season (n=70)**	**P**
Follicle size at the onset of Ovsynch (mm)	19.1 ± 1.1	20.8 ± 0.9	NS
Response to the GnRH1 (%, n/n)	90.7 (39/43)	81.4 (57/70)	NS
Response to the GnRH2 (%, n/n)	83.7 (36/43)	87.1 (61/70)	NS
Follicle size at FTAI (mm)	15.3 ± 0.3	15.8 ± 0.3	NS
Pregnancy rate on d 31(%, n/n)	34.9 (15/43)	35.7 (25/70)	NS
Pregnancy rate on d 62 (%, n/n)	25.6 (11/43)	32.9 (23/70)	NS
Pregnancy loss (%, n/n)	26.7 (4/15)	8.0 (2/25)	NS

Irrespective of seasonal effect, 84.9% of cows responded to GnRH1 of Ovsynch in this study. Besides, ovulatory response to the GnRH1 of Ovsynch was not different (P > 0.05) during warm (90.7%, 39/43) and cool (81.4%, 57/70) seasons. Besides, ovulatory response to the GnRH2 of Ovsynch was similar (P > 0.05) during warm (83.7%, 36/43) and cool (87.1%, 61/70) seasons, respectively (Table 1). Although there was a tendency (P = 0.05) for higher ovulatory response to the GnRH1 in primiparous cows during warm season (95.4%) compared to cool season (75.7%), there was no difference in ovulatory response to the GnRH2 (81.8% vs. 86.5%), respectively (P > 0.05). In multiparous cows, similar ovulatory response to the GnRH1 (85.7% vs. 87.9%) and GnRH2 (85.7% vs. 87.9%) was found during warm and cool seasons, respectively. Irrespective of seasonal effect and parity, ovulatory responses to the GnRH1 in small, medium and large follicular size groups were 93.1%, 84.2%, and 77.8%, respectively. Preovulatory follicular size (small, medium, large) at the onset of Ovsynch did not affect (P > 0.05) the ovulatory response to the GnRH1 during warm (100%, 89.5%, 75%, Table 2) and cool seasons (86.4%, 81.6%, 78.9%, Table 3), respectively. The second ovulatory response to the GnRH2 for small, medium, and large follicle size groups were 62.5%, 94.7%, 100% in warm season (Table 2), and 92.3%, 78.9%, %100 in cool season (Table 3). Although there was no difference in the second ovulatory response among different size follicles during cool season, the second ovulatory response was lower in cows that had small-size follicles than in cows that had medium and large-size follicles during warm season (P < 0.05).

Table 2. *Effect of follicle size at the onset of Ovsynch on ovulatory response and pregnancy rate in noncyclic cows during warm season*

Response to the Ovsynch	**Follicle size**		
	Small 8-15 mm	**Medium 16-24 mm**	**Large ≥24 mm**
Response to the GnRH1 (%)	100 (16/16)	89.5 (17/19)	75 (6/8)
Response to the GnRH2 (%)	62.5 (10/16)^{a}	94.7 (18/19)^{b}	100 (8/8)^{b}
Follicle size at FTAI (mm)	13.5 ± 1.2	15.7 ± 0.5	15.7 ± 1.4
Pregnancy rate on d 31 (%)	31.3 (5/16)	47.4 (9/19)	12.5 (1/8)
Pregnancy rate on d 62 (%)	31.3 (5/16)	26.3 (5/19)	12.5 (1/8)

$^{a, b}$: Values within a row with different superscripts differ (P < 0.05).

Table 3. *Effect of follicle size at the onset of Ovsynch on ovulatory response and pregnancy rate in noncyclic cows during cool season*

Response to the Ovsynch	**Follicle size**		
	Small 8-15 mm	**Medium 16-24 mm**	**Large ≥24 mm**
Response to the GnRH1 (%)	84.6 (11/13)	81.6 (31/38)	78.9 (15/19)
Response to the GnRH2 (%)	92.3 (12/13)^{ab}	78.9 (30/38)^{a}	100 (19/19)^{b}
Follicle size at FTAI (mm)	15.0 ± 0.6^{A}	15.6 ± 0.4^{A}	16.8 ± 0.6^{B}
Pregnancy rate on d 31 (%)	23.1 (3/13)	31.6 (12/38)	52.6 (10/19)
Pregnancy rate on d 62 (%)	15.4 (2/13)^{A}	28.9 (11/38)^{A}	52.6 (10/19)^{B}

$^{a, b}$: Values within a row with different superscripts differ (P < 0.05).
$^{A, B}$: Values within a row with different superscripts show tendency (0.05 < P < 0.10).

Pregnancy rate on d 31 was similar (P > 0.05) during the warm (34.9%, 15/43) and cool seasons (35.7%, 25/70; Table 1). Similarly, parity was not influenced by the season (P > 0.05). Pregnancy rates on day 31 were 36.4; (8/22) vs. 37.8%; (14/37) in primiparous cows and 33.3%; (7/21) vs. 33.3%; (11/33) in multiparous cows during warm and cool seasons, respectively. Cows that had an ovulatory response to the GnRH1 (36.5%; 35/96) had a 7.1% greater pregnancy rate (P >0.05) than cows that did not have an ovulatory response (29.4%; 5/17). The pregnancy rates and ovulatory response to the GnRH1 and GnRH2 in cows that had different follicle sizes at the onset of Ovsynch were shown in Table 2 and Table 3. There was no difference (P > 0.05) in pregnancy rate at d 62 in (25.6%, 11/43) warm and cool seasons (32.9%, 23/70). There was no difference in pregnancy loss (P > 0.05) between warm (26.7%, 4/15) and cool season (8%, 2/25; Table 1).

DISCUSSION

In the present study, it was aimed to evaluate the effect of season on hormonal response and pregnancy rates in noncyclic dairy cows that were synchronized with Ovsynch. Comfort zone was reported at 5 to 25 °C in dairy cows (McDowell et al. 1979) and a higher ambient temperature than 25 °C increases the adverse effect of heat stress on reproductive performance (Roth et al. 2001). Thorton et al. (2021) reported that Holstein cows were susceptible to heat stress due to higher core temperature, lower skin surface to mass ratio, and higher milk yield compared to other dairy breeds. It was reported that forecast data of meteorology are mostly estimated rectal temperature of individual cows and these values show the severity of heat stress and its adverse effect (Dikmen and Hansen 2009). Consistent with the previous study (Dikmen and Hansen 2009), environmental temperature and relative humidity were obtained from meteorological service and THI was higher than 72 during warm season in this study.

Heat stress impairs balance in the activity of the hypothalamo-hypophyseal-ovarian axis (De Rensis et al. 2017). Heat stress disrupts not only pre-antral follicles, but also steroidogenic capacity, oocyte quality, and development of the preovulatory follicle (De Rensis et al. 2021; Keskin et al. 2016; Schüller, et al. 2017). Roth et al. (2000) reported that plasma inhibin concentration was lower in cows exposed to heat stress. Lower plasma inhibin concentration caused a delayed regression of subordinate follicles and increased plasma follicle stimulating hormone (FSH) concentration (Wolfenson et al. 1995). Increased preovulatory follicle size (16.4 vs. 14.5 mm) and reduced subordinate follicle size (7.9 vs. 10.1 mm) were reported in heat-stressed cows that were assigned to shade (Badinga et al. 1993). Additionally, previous studies reported that heat stress seriously decreased size of dominant follicles and increased number of large follicular size (Badinga et al. 1993; Roth et al. 2000; Wolfenson et al. 1995). Schüller et al. (2017) found that increment of each unit of THI decreased follicle size by 0.1 mm at the time of insemination. Consistent with the previous studies, a statistically lower percentage of small-sized follicles (about %20) in warm season could be a significant factor compared to cool season in this study. Besides, the average preovulatory follicle size at the onset of synchronization protocol was 1.7 mm lower in warm season compared to that in cool season.

Ovulation synchronization protocols such as Ovsynch synchronize ovarian follicular dynamic and have been proposed as a plausible approach to improving reduced fertility in heat stress (Roth et al. 2001; Wolfenson and Roth 2019). Since ovulation failure is higher (12.4 vs. 3.4%) under heat stress (López-Gatius et al. 2005), the first GnRH of Ovsynch is required to induce ovulation (Dirandeh 2014; Sartori et al. 2001). The fertility success of Ovsynch is positively associated with ovulatory response to the GnRH1 and achievement of higher progesterone concentrations before PGF2α injection. Ovulation following GnRH1 results in the onset of a new follicular wave and ensures the synchronization with healthy preovulatory follicles (Bello et al. 2006; Bisinotto et al. 2010). Ovulatory response to the GnRH1 highly ranged from 56 to 97% at random stages at the onset of Ovsynch (Bello et al. 2006; Bisinotto et al. 2010; Carvalho et al. 2015; Chebel et al. 2006; Dirandeh 2014; Keskin et al. 2011; Vasconcelos et al. 1999). Besides, it was reported that noncyclic cows had higher ovulatory response to the GnRH1 than cyclic cows (Carvalho et al. 2015; Gumen et al. 2003; Moreira et al., 2001). Gumen et al. (2003) and Carvalho et al. (2015) reported that noncyclic cows (88% and 81.1%) had approximately 20% higher ovulatory response than cyclic cows (62% and 60.3%), respectively. Consistent with the previous studies (Carvalho et al. 2015; Gumen et al. 2003; Moreira et al. 2001), a large proportion of noncyclic cows (84.9%) responded to GnRH1 of Ovsynch in this study. In addition, there was no difference in ovulatory response to the GnRH1 regarding seasonal effect (90.7% for warm and 81.4% for cool season) in the present study. Besides, the difference in follicle sizes at the onset of protocol did not affect the ovulatory response to the GnRH1 of Ovsynch.

It was well known that hypothalamo-pituitary-ovarian axis activity and secretion of gonadotropins are negatively affected during heat stress (Hansen 2019; Schüller et al. 2017; Wolfenson and Roth 2019). The presence of more than 10 mm follicle size did not guarantee ovulation following administration of the first GnRH of Ovsynch (Bello et al. 2006),. The majority (75% in warm and 78.9% cool seasons) of cows had follicular cyst (large-size follicle group) responded to first GnRH of Ovsynch in this study. Consistent with our results, Gumen et al. (2003) reported that 72 to 90% of noncyclic cows with follicular cyst responded to GnRH administration.

It was reported that cows had a higher ovulatory response to the GnRH2 than the first GnRH in Ovsynch (Vasconcelos et al. 1999). Ovulatory response to the GnRH2 ranged from 75.9 to 100% in previous studies (Carvalho et al. 2015; Dirandeh 2014; Gumen et al. 2003; Moreira et al. 2001; Stevenson et al. 2006; Vasconcelos et al. 1999). In this study, our results for ovulatory response to GnRH2 in noncyclic cows were consistent (87.1%, for warm season and 83.7% for cool season) with the previous studies. However, the second ovulatory response was lower in cows that had small-size follicles at the onset of protocol compared to cows that had large and medium sized follicles during the warm season. It is well known that GnRH induces LH release and the lack of ovulation in response to GnRH may have been due to the stage of follicle development at the time of administration (Wijma et al. 2017). Heat stress and/or other factors that resulted in the small follicle diameter at the onset of the protocol may have continued to adversely affect the development of these follicles at the time of the second GnRH.

As in this study, the cooling approach based on evaporation from the skin from with fans following short-term spraying of water is the first strategy to prevent hyperthermia and disruptions of heat stress on fertility in dairy herds (Wolfenson and Roth 2019). Fertility was influenced by the efficiency of the cooling approach in cows that were exposed to heat stress (Roth 2008; Hansen 2019). While the pregnancy rate was 25-27% in Oklahoma and Kansas in heat-stressed cows (Voelz et al. 2016) that were housed with shade in a study, the pregnancy rate was 32% in Florida in heat-stressed cows that were housed with fans and sprinklers in another study (Zolini et al. 2019). Besides, Schüller et al. (2017) showed that exposure the higher (>72) THI one day before insemination caused a reduced pregnancy rate from 31 to 12% in cows. Although it was hypothesized that heat stress has a negative effect on reproductive performance (De Rensis et al. 2017; Schüller et al. 2017; Wolfenson and Roth 2019), there was no difference in pregnancy rate between warm and cool seasons in this study. Administration of GnRH at the onset of ovulation synchronization protocols is recommended to improve fertility due to low LH surge and ovulation failure during summer (Wolfenson and Roth 2019). A higher progesterone concentration before and after timed AI is critical for embryo establishment and a greater pregnancy rate at timed-AI protocols (Wiltbank et al. 2014). The use of timed AI protocol and higher response to the GnRH1 in warm season might have prevented the difference in fertility between seasons in this study. Higher ovulatory response to the GnRH1 is a potential indicator of a greater pregnancy rate in cows (Carvalho et al. 2015; Chebel et al. 2006). Carvalho et al. (2015) found a 16.4% greater pregnancy rate in cows that ovulatory response to the GnRH1 than in cows that did not ovulate. Similar to the previous report, although there was no statistical difference, the difference in pregnancy rate was 7.1% between cows that ovulated after GnRH1 and did not ovulate. However, similar body condition scores and follicle sizes at the time of FTAI might have contributed to explaining the similar pregnancy rate between warm and cool seasons (Keskin et al. 2016; Santos et al. 2001). Furthermore, delaying the time of insemination (>90 days) due to higher negative energy balance in early lactation might have contributed to obtaining higher fertility in cows subjected to Ovsynch during warm season (Wiltbank et al. 2008). Cows had peak milk yield in early lactation (approximately 45 to 70 days) and had greater metabolic heat stress than cows that had low milk yield (Wheelock et al. 2010). It was well known that continuation of the negative energy balance during peak milk yield and the adverse effect of heat stress could prevent the obtaining optimal pregnancy rate in noncyclic cows received Ovsynch protocol (Santos et al. 2009; Wiltbank et al. 2002). Wiltbank et al. (2008) stated that the pregnancy rate of noncyclic cows was similar to cyclic cows when Ovsynch protocol started after postpartum day 100. Delaying the onset of protocol (Wiltbank et al. 2008) and optimal precautions such as cooling approach (Roth 2008) and nutrition could tolerate the deleterious effect of heat stress on pregnancy rate in this study during warm season.

Although there was a great percentage of difference in pregnancy loss between warm season (26.7%) and cool season (8%), the lack of statistical difference might have resulted from the small number of animals in this study. Previous studies revealed that heat stress leading to elevated body temperature directly damages oocyte and also leads to early embryo death (Nanas et al. 2021). The results of the previous study clearly demonstrated that the warm season resulted in significantly more pregnancy loss when compared to cool season (Alnimer et al. 2009; Hansen 2002; Nanas et al. 2021; Wolfenson and Roth 2019). Although a low number of animals in groups might have masked the statistical difference, pregnancy rate on d 62 was relatively 22.2% (a difference of 7.3%) lower in this study.

CONCLUSIONS

The seasonal difference did not affect the first and second ovulatory response to the GnRH and pregnancy rate in noncyclic Holstein cows that received Ovsynch protocol. Pregnancy loss was relatively higher in warm season than cool season. It was concluded that delayed the onset of synchronization protocol, higher ovulatory response to GnRH of Ovsynch, and cooling strategies may offer similar fertility in noncyclic cows during warm season compared to that during cool season. Comprehensive future studies are needed to increase ovulation response and fertility under heat stress in noncyclic cows treated with the Ovsynch protocol.

ACKNOWLEDGEMENTS

The authors would like to thank Associate Professor Abdulkadir Orman for the statistical analysis. Our appreciation is extended to Prof. Dr. Ahmet Gümen and Prof. Dr. Abdulkadir Keskin for their contributions and editing of the manuscript.

ETHICAL STATEMENT

During the writing process of the study titled "Effect of Season on Ovulatory Response and Reproductive Performance in Noncyclic Lactating Dairy Cows Synchronized with Ovsynch", scientific rules, ethical and citation rules were followed; No falsification has been made on the collected data and this study has not been sent to any other academic media for evaluation. This study was approved by the ethics committee of the Lalahan Livestock Central Research Institute Animal Care Committee (approval number: 2009/27)

CONFLICT OF INTERESTS

The authors declared no conflict of interest.

AUTHORS CONTRIBUTION

All authors contributed equally.

REFERENCES

Alnimer MA, Tabbaa MJ, Ababneh MM and Lubbadeh WF 2009. Applying variations of the Ovsynch protocol at the middle ofthe estrus cycle on reproductive performance of lactating dairycows during summer and winter. Theriogenology. 72: 731-40.

Badinga L, Thatcher WW, Diaz T, Drost M, Wolfenson D 1993. Effect of environmental heat stress on follicular development and steroidogenesis in lactating Holstein cows. Theriogenology, 39(4): 797-810.

Bello NM, Steibel JP, Pursley JR 2006. Optimizing ovulation to first GnRH improved outcomes to each hormonal injection of Ovsynch in lactating dairy cows. J Dairy Sci. 89(9): 3413-24.

Bisinotto RS, Chebel RC, Santos JEP 2010. Follicular wave of the ovulatory follicle and not cyclic status influences fertility of dairy cows. J Dairy Sci. 93(8): 3578-87.

Cartmill JA, El-Zarkouny SZ, Hensley BA, Lamb GC, Stevenson JS 2001. Stage of cycle, incidence, and timing of ovulation, and pregnancy rates in dairy cattle after three timed breeding protocols. J Dairy Sci. 84(5): 1051-59.

Carvalho PD, Wiltbank MC, Fricke PM 2015. Manipulation of progesterone to increase ovulatory response to the first GnRH treatment of an Ovsynch protocol in lactating dairy cows receiving first timed artificial insemination. J Dairy Sci. 98(12): 8800-13.

Chebel RC, Santos JE, Cerri RL, Rutigliano HM, Bruno RG 2006. Reproduction in dairy cows following progesterone insert presynchronization and resynchronization protocols. J Dairy Sci. 89: 4205-19.

De Rensis F, Scaramuzzi RJ 2003. Heat stress and seasonal effects on reproduction in the dairy cow-a review. Theriogenology. 60: 1139-51.

De Rensis F, Garcia-Ispierto I, López-Gatius F 2015. Seasonal heat stress: Clinical implications and hormone treatments for the fertility of dairy cows. Theriogenology. 84(5): 659-66.

De Rensis F, López-Gatius F, Garcia-Ispierto I, Morini G, Scaramuzzi RJ 2017. Causes of declining fertility in dairy cows during the warm season. Theriogenology. 91: 145-53.

De Rensis F, Saleri R, Garcia-Ispierto I, Scaramuzzi R, López-Gatius F 2021. Effects of heat stress on follicular physiology in dairy cows. Animals. 11(12): 3406.

Dikmen S, Hansen PJ 2009. Is the temperature-humidity index the best indicator of heat stress in lactating dairy cows in a subtropical environment?. J Dairy Sci. 92(1): 109-16.

Dirandeh E 2014. Starting Ovsynch protocol on day 6 of first postpartum estrous cycle increased fertility in dairy cows by affecting ovarian response during heat stress. Anim Reprod Sci. 149(3-4): 135-140.

El-Tarabany MS, El-Tarabany AA 2015. Impact of maternal heat stress at insemination on the subsequent reproductive performance of Holstein, Brown Swiss, and their crosses. Theriogenology. 84(9): 1523-29.

Ferguson JD, Galligan DT, Thomsen N 1994. Principal descriptors of body condition score in Holstein cows. J Dairy Sci. 77: 2695- 2703.

Garcia-Ispierto I, De Rensis F, Pérez-Salas JA, Nunes JM, Pradés B, Serrano-Pérez B, López-Gatius F 2019. The GnRH analogue dephereline given in a fixed-time AI protocol improves ovulation and embryo survival in dairy cows. Res Vet Sci. 122: 170-4.

Gumen A, Guenther JN, Wiltbank MC 2003. Follicular size and response to ovsynch versus detection of estrus in anovular and ovular lactating dairy cows. J Dairy Sci. 86: 3184-94.

Hansen JE 2002. Embryonic stem cell production through therapeuticcloning has fewer ethical problems than stem cell harvest fromsurplus IVF embryos. J Med Ethics. 28: 86-8.

Hansen PJ 2019. Reproductive physiology of the heat-stressed dairy cow: implications for fertility and assisted reproduction. Ani Reprod. 16: 497-507.

Keskin A, Yilmazbas-Mecitoglu G, Gumen A, Karakaya E, Darici R, Okut H 2010. Effect of hCG vs. GnRH at the beginning of the Ovsynch on first ovulation and conception rates in cyclic lactating dairy cows. Theriogenology. 74(4): 602-07.

Keskin A, Yilmazbas-Mecitoglu G, Gumen A, Karakaya E, Celik Y, Okut H, Wiltbank, MC 2011. Comparison of responses to Ovsynch between Holstein-Friesian and Swedish Red cows. J Dairy Sci. 94(4): 1784-89.

Keskin A, Mecitoğlu G, Bilen E, Güner B, Orman A, Okut H, Gümen A 2016. The effect of ovulatory follicle size at the time of insemination on pregnancy rate in lactating dairy cows. Turk J Vet Ani Sci. 40(1): 68-74.

López-Gatius F 2003. Is fertility declining in dairy cattle? A retrospective study in northeastern Spain. Theriogenology. 60: 89-99.

López-Gatius F, López-Béjar M, Fenech M, Hunter RHF 2005. Ovulation failure and double ovulation in dairy cattle: risk factors and effects. Theriogenology. 63(5): 1298-1307.

McDowell D, Hooven N, Cameron K 1979. Effects of climate on performance of Holsteins in first lactation. J Dairy Sci. 68: 2418-35.

Moreira F, Orlandi C, Risco CA, Mattos R, Lopes FL, Thatcher WW 2001. Effects of presynchronization and bovine somatotropin on pregnancy rates to a timed artificial insemination protocol in lactating dairy cows. J Dairy Sci. 84: 1646-59.

Nanas I, Barbagianni M, Dadouli K, Dovolou E, Amiridis GS 2021. Ultrasonographic findings of the corpus luteum and the gravid uterus during heat stress in dairy cattle. Reprod Domes Anim. 56(10): 1329-41.

National Research Council 1971. A guide to environmental research on animals, National Academy of Sciences, Washington, DC.

National Research Council 2001. Nutrient requirements of dairy cattle, 7th revised ed., National Academy of Sciences, Washington, DC.

Pursley JR, Kosorok MR, Wiltbank MC 1997. Reproductive management of lactating dairy cows using synchronization of ovulation. J Dairy Sci. 80: 301-06.

Pursley JR, Fricke PM, Garverick HA, Kesler DJ, Ottobre JS, Stevenson JS, Wiltbank MC 2001. Improved fertility in noncycling lactating dairy cows treated with exogenous progesterone during Ovsynch. J Dairy Sci. 84: 1563.

Roth Z, Meidan R, Braw-Tal R, Wolfenson D 2000. Immediate and delayed effects of heat stress on follicular development and its association with plasma FSH and inhibin concentration in cows. J Reprod Fertil. 120(1): 83-90.

Roth Z, Arav A, Bor A, Zeron Z, Braw-Tal R, Wolfenson D 2001. Improvement of quality of oocytes collected in the autumn by enhanced removal of impaired follicles from preovulatory heatstressed cows. Reproduction. 122: 737-44.

Roth Z 2008. Heat stress, the follicle, and its enclosed oocyte: mechanisms and potential strategies to improve fertility in dairy cows. Reprod Domest Anim. 43: 238-44.

Santos JE, Thatcher WW, Pool L, Overton MW 2001. Effect of human chorionic gonadotropin on luteal function and reproductive performance of high-producing lactating Holstein dairy cows. J Anim Sci. 79(11): 2881-94.

Santos JEP, Rutigliano HM, SáFilho MF 2009. Risk factors for resumption of postpartum estrous cycles and embryonic survival in lactating dairy cows. Anim Reprod Sci. 110: 207-21.

Santos JEP, Bisinotto RS, Ribeiro ES 2016. Mechanisms underlying reduced fertility in anovular dairy cows. Theriogenology. 86: 254-62.

Sartori R, Fricke PM, Ferreira JC, Ginther OJ, Wiltbank MC 2001. Follicular deviation and acquisition of ovulatory capacity in bovine follicles. Biol Reprod. 65(5): 1403-09.

Schüller LK, Burfeind O, Heuwieser W 2014. Impact of heat stress on conception rate of dairy cows in the moderate climate considering different temperature humidity index thresholds, periods relative to breeding, and heat load indices. Theriogenology. 81: 1050-57.

Schüller LK, Michaelis I, Heuwiese, W 2017. Impact of heat stress on estrus expression and follicle size in estrus under field conditions in dairy cows. Theriogenology. 102: 48-53.

Stevenson JS, Pursley JR, Garverick HA, Fricke PM, Kesler DJ, Ottobre JS and Wiltbank MC 2006. Treatment of cycling or noncycling lactating dairy cows with progesterone during the Ovsynch. J Dairy Sci. 89: 2567-2578.

Thornton P, Nelson G, Mayberry D, Herrero M 2021. Increases in extreme heat stress in domesticated livestock species during the twenty-first century. Glob Chang Biol. 27(22): 5762-72.

Vasconcelos JLM, Silcox RW, Rosa GJM, Pursley JR, Wiltbank MC 1999. Synchronization rate, size of the ovulatory follicle, and pregnancy rate after synchronization of ovulation beginning on different days of the estrous cycle in lactating dairy cows. Theriogenology. 52(6): 1067-78.

Voelz BE, Rocha L, Scortegagna F, Stevenson JS, Mendonça LGD 2016. Treatment of lactating dairy cows with gonadotropin-releasing hormone before first insemination during summer heat stress. J Dairy Sci. 99(9): 7612-23.

Wheelock JB, Rhoads RP, VanBaale MJ, Sanders SR, Baumgard LH 2010. Effects of heat stress on energetic metabolism in lactating Holstein cows. J Dairy Sci. 93(2): 644-55.

Whether online 2019. https://www.weatheronline.co.uk/weather/maps/city.

Wijma R, Stangaferro ML, Masello M, Granados GE, Giordano JO 2017. Resynchronization of ovulation protocols for dairy cows including or not including gonadotropin-releasing hormone to induce a new follicular wave: Effects on re-insemination pattern, ovarian responses, and pregnancy outcomes. J Dairy Sci. 100(9): 7613-25.

Wiltbank MC, Gümen A, Sartori R 2002. Physiological classification of anovulatory conditions in cattle. Theriogenology. 57: 21-52.

Wiltbank MC, Gümen A, Lopez H, Sartori R 2008. Management and treatment of dairycows that are not cycling or follicular cysts. Cattle Pract. 16: 14-19.

Wiltbank MC, Souza AH, Carvalho PD, Cunha AP, Giordano JO, Fricke PM, Baez GM, Diskin MG 2014. Physiological and practical effects of progesterone on reproduction in dairy cattle. Animal. 8(s1): 70-81.

Wolfenson D, Thatcher WW, Badinga L, Savio JD, Meidan R, Lew BJ, Braw-Tal R, Berman A 1995. Effect of heat stress on follicular development during the estrous cycle in lactating dairy cattle. Biol Reprod. 52(5): 1106-13.

Wolfenson D, Roth Z 2019. Impact of heat stress on cow reproduction and fertility. Anim Front. 9(1): 32-38.

Zolini AM, Ortiz WG, Estrada-Cortes E, Ortega MS, Dikmen S, Sosa F, Giordano JO, Hansen PJ 2019. Interactions of human chorionic gonadotropin with genotype and parity on fertility responses of lactating dairy cows. J Dairy Sci. 102(1); 846-56.