



Effect of Parity and Milk Production on Conception Rate After Two Synchronization Protocols in Dairy Cows

Emad M. Abd El-Razek, Tamer M. Genedy, Ahmed H. Zaghloul

Department of Theriogenology, Faculty of Veterinary Medicine, University of Sadat City, Egypt.

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*Correspondence to:

tamer.genedy@vet.us
c.edu.eg

ABSTRACT

Our objective in this study the effect of parity and average milk production on the conception rate after resynchronization with G6G protocol or Ovsynch protocol. Lactating Holstein cows (n = 100) divided into two groups (Ovsynch and G6G) cows in this group I (n = 50) were received the following treatment sequence, 12 µg Buserelin on day zero, 500 µg IM of Cloprostenol on day seven, 12 µg Buserelin IM on day nine and received timed artificial insemination (TAI) 16 hours later. Group II (G6G): were received 500 µg IM Cloprostenol on day zero, 12 µg Buserelin on day two and six days later these cows received the Ovsynch as previously explained in the first group. Conception rate was (40 vs. 30 %) for G6G and Ovsynch protocol respectively primiparous cows, conception rate to G6G program was 46.15%, compared to 28.57% in cows synchronized with Ovsynch program. In Multiparous cows, the similar trend was observed but the difference was smaller (conception rate 37.8 vs. 30.55% in cows synchronized with G6G and with Ovsynch, respectively). In high producers, conception rate was numerically higher in the Ovsynch group (40%) compared to the G6G group (36%). While, in low producers, conception rate to G6G protocol (48%) tended ($P = 0.07$) to be higher than that to Ovsynch protocol (20%) in conclusion Administration of $\text{PGF}_2\alpha$ 8 d before initiation of Ovsynch increase the conception rate compared with the standard Ovsynch protocol in primiparous cows than in multiparous cows and in low producers than high producer cows

1. INTRODUCTION:

The reproductive efficiency in dairy cows has declined over the last several years and is considered lower than required (Lucy, 2001; de Varis and Risco, 2002; Washburn et al., 2002). This is thought to be due to many factors including inefficiency and inaccuracy of estrus detection, improper timing of insemination, delayed ovulation, anovulation, negative energy balance, selection for high milk production and inbreeding (Lucy, 2001). After the advent of AI, estrus detection became paramount for the success of reproductive management systems. However, detection of estrus is laborious and quite variable depending on the season, farm nutritional and health status and management skills (Moreira, 2001). The development of timed artificial insemination (TAI)

protocols based on the use of GnRH and $\text{PGF}_2\alpha$ precisely synchronizes the time of ovulation and eliminates the need for estrus detection (Pursley et al., 1995). This protocol is designated as the Ovsynch/TAI. It produces pregnancy rates greater or comparable to control groups that depend on estrus detection (Britt and Gaska, 1998). In the last decade, conception rates are at an all-time low of approximately 40% (Pursley et al., 1997a; Peter, 2000; Thatcher et al., 2002).

Controlled ovulation allows TAI without estrus detection. By synchronizing ovulation in cows selected for AI, 100% of the cows can be inseminated on the first day of the breeding period without waiting for the cow to display estrus. This is equivalent to 100% estrus detection rate for first AI and a 100% service rate. Thus TAI protocols were employed in the

dairy farm management program to improve conception and neglect the need for heat detection. However, the efficiency of these protocols must be evaluated in terms of fertility and farm economics (Peters, 2000). Later studies have demonstrated that initiation of the Ovsynch program at certain stages of the estrus cycle causes reduced pregnancy rates (Vasconcelos et al., 1999; Moreira et al., 2000). For example, initiation of the Ovsynch program between days 13 and 17 of the cycle (the time during which spontaneous regression of the CL occurs prior to the time that Prostaglandin F₂ alpha (PGF₂ α) is administered at 7 days after the administration of GnRH) causes the cows to ovulate prematurely relative to insemination and are not likely to conceive (Thatcher et al., 2001). A second problematic stage occurs early in the estrus cycle. At this stage, spontaneous ovulation has already occurred, and the new dominant follicle is too small to ovulate in response to GnRH administration (Moreira, et al., 2000). As a consequence, at the second administration of GnRH the dominant follicle is considered aged (Thatcher et al., 2002) and cows that are included in the Ovsynch program in the early estrus cycle are less fertile (Vasconcelos et al., 1999b and Moreira et al., 2000). Moreover, conception rate is low if anestrus cows are subjected to the ordinary Ovsynch protocol (Thatcher et al., 2002). It is well known that primiparous cows constitute about 30-50% of the dairy herd. These cows are usually anestrus post-partum due to the fact that first lactation cows not only have to ensure milk production for their off-springs but also their own growth demands are still found (Thatcher et al., 2001 and Sanz et al., 2004). Consequently, poor conception results are expected following synchronization of these anestrus cows using Ovsynch protocol. Pursley et al. (1997b) reported that, Ovsynch protocol allowed effective management of TAI in lactating dairy cows without need for estrus detection with pregnancy rate of 37% and 53% vs. 5% and 35% for treated and control groups at 60 and 100 days postpartum. A program commonly used to improve responses to TAI protocols is to administer 2 injections of PGF₂α 14 d apart, with the second injection 10 to 12 days before the first GnRH of the TAI protocol to pre-synchronize the estrus cycle (Moreira et al., 2001). GnRH induces ovulation in 76% of the cows without a CL (Galvao et al., 2007a) and 88% in anovular cows (Gumen and Seguin, 2003). Therefore, combining GnRH and PGF₂α for pre-synchronization might

benefit anovular cows by inducing estrus cyclicity before the initiation of the timed AI program. Yilmaz et al. (2011) compared the efficiency of the G6G and the ordinary Ovsynch as ovulation synchronization programs in dairy cows and heifers. The authors noted higher conception figures which were significant in heifers. Moreover, it was reported that in this protocol (G6G) the rate of the cows synchronized for Ovsynch was 92% and the pregnancy rate was 50% as compared to 27% for cows synchronized with Ovsynch (Bello et al., 2006). Our aim is Implementation of an efficient resynchronization protocol to decrease the inter-insemination intervals of cows diagnosed non-pregnant.

2. MATERIALS and METHODS

2.1 Experimental location :

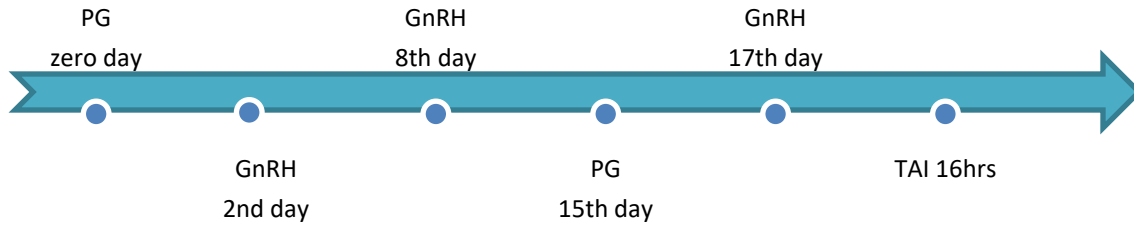
This study was carried out on 100 cows. Animals were reared in a well-managed private Holstein herd located in Northwestern Egypt.

2.2 Animals:

Primiparous and pleuriparous cows were used in this study in order to improve their reproductive and productive efficiency under Egyptian conditions. Cows were fed a totally mixed ration (TMR) three times daily on eight occasions per day aiming at increasing their dry matter intake (DMI). All rations were formulated in line with the national research council (NRC, 2001) recommendations. Close up prepartum transition ration was manipulated to have a negative dietary cationic - anionic difference (DCAD) value of 40 mEq/Kg DM. Water troughs were freely accessible at one corner of the yard and provided clean fresh water all the time. Water troughs were also shaded to protect against sunlight.

2.3 Experimental Design:

All cows in this study were allocated into two equal groups. Group I (Ovsynch): cows in this group (n = 50) were received the following treatment sequence, 12 µg Buserelin (Receptal®, Intervet International Boxmeer B.V., Holland) IM on day zero, 500 µg IM of Cloprostenol (Estrumate®, a synthetic PGF₂α, Schering – Plough Animal Health) on day seven, 12 µg Buserelin IM on day nine and received timed artificial insemination (TAI) 16 hours later. Group II (G6G): cows in this group (n = 50) were received 500 µg IM Cloprostenol on day zero, 12 µg Buserelin on day two and six days later these cows received the Ovsynch as previously explained in the first group.



Schedule for injection of G6G/Ovsynch in lactating dairy cows

2.4 Statistical analyses:

Data were tested for normal distribution using Kolmogorov-Smirnov test. Data were presented as mean \pm SE or displayed as percentages. Significance was declared at $P < 0.05$.

For normally distributed data, differences between means were tested using independent samples T- test with leaven's interpretations (SPSS 2007, version 15), while Mann-Whitney test was used to analyze statistical differences between data not following a normal distribution. Moreover, Chi- square test was used to analyze the differences between proportions such as incidence rates of infertility problems and conception rate

3. RESULT

Application of G6G synchronization protocol was associated with a 10 point increase in conception rate when compared to the ordinary Ovsynch protocol (40 vs. 30%, respectively), however this difference did not reach a statistical significance as illustrated in figure 1. Although the differences in conception rate were not significant between cows synchronized with Ovsynch and those with G6G, similar trends were observed in Primiparous and Multiparous cows. In primiparous cows, conception rate to G6G program

was 46.15%, compared to 28.57% in cows synchronized with Ovsynch program. In Multiparous cows, a similar trend was observed but the difference was smaller (conception rate 37.8 vs. 30.55% in cows synchronized with G6G and with Ovsynch, respectively). Moreover, conception rate in G6G group was numerically higher in primiparous cows (46.15%), compared to multiparous cows (37.8%) a trend which was not observed for the Ovsynch protocol (Table 1 and figure 2). Although the differences in conception rate were not significant between cows synchronized with Ovsynch and those with G6G, similar trends were observed in Primiparous and Multiparous cows. In primiparous cows, conception rate to G6G program was 46.15%, compared to 28.57% in cows synchronized with Ovsynch program. In Multiparous cows, a similar trend was observed but the difference was smaller (conception rate 37.8 vs. 30.55% in cows synchronized with G6G and with Ovsynch, respectively). Moreover, conception rate in G6G group was numerically higher in primiparous cows (46.15%), compared to multiparous cows (37.8%) a trend which was not observed for the Ovsynch protocol (Table 1 and figure 2).

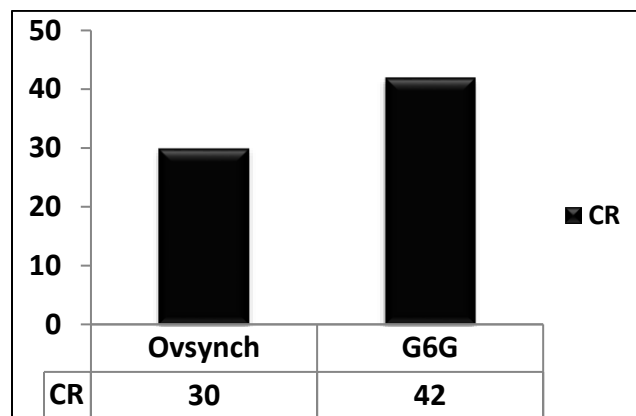


Figure 1): Effect of synchronization program (G6G vs. Ovsynch) on conception rate (CR) in Holstein cows (Chi-square analysis, $P > 0.05$).

Table 1): Effects of parity on conception rate to Ovsynch and G6G synchronization protocols in dairy cows:

criteria	Primiparous cows		Multiparous cows		P value	
	Ovsynch	G6G	Ovsynch	G6G	Primi	Multi
Total	14	13	36	37	NA	NA
CR (n, %)	4 (28.57)	6 (46.15)	11 (30.55)	14 (37.80)	0.44	0.46

CR= Conception rate

Means within the same column carrying different letters are significantly different (P<0.05)

Producers, conception rate was numerically higher in the Ovsynch group (40%) compared to the G6G group (36%). While, in low producers, conception rate to G6G protocol (48%) tended (P = 0.07) to be higher than that to Ovsynch protocol (20%).

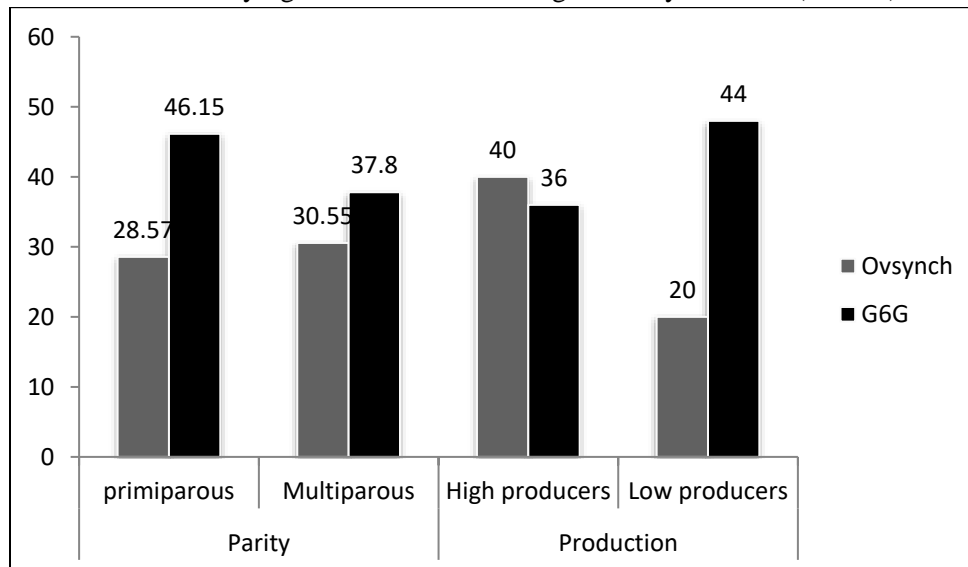
In spite of lacking as statistical power, conception rate to Ovsynch in high producers was twice (40%) that in low producers (20%), while conception rate to G6G was 8 points higher in low producers (44%), compared to high producers (36%).

Table 2): Effects of average daily milk on conception rate to Ovsynch and G6G synchronization protocols in dairy cows:

criteria	High producers		low producers		P value	
	Ovsynch	G6G	Ovsynch	G6G	High	Low
Number	25	25	25	25	NA	NA
CR (n, %)	10 (40.00)	9 (36.00)	5 (20.00)	11 (44.00)	0.91	0.07

CR= Conception rate

Means within the same column carrying different letters are significantly different (P<0.05)

**Figure 2): Effects of parity and average daily milk production on conception rate to Ovsynch and G6G synchronization programs in dairy cows:**

4. DISCUSSION

G6G presynchronization protocol depends on the idea portion of the estrus cycle (d 10 to 21). In the current study, that the first PG injection will induce luteolysis of all mature administration of PGF $_{2\alpha}$ 8 d before the first GnRH injection corpora lutea. Then, a GnRH injection 2 days late will of Ovsynch shifted some cows into the early luteal phase of induce an ovulation. So, the traditional Ovsynch protocol the estrus cycle at the time of the first GnRH injection that will be initiated at the 6th day of the cycle which is the would have been in the later stages of the estrus cycle had optimal time for initiation of the Ovsynch protocol as they not received PGF $_{2\alpha}$. Despite this shifting of cows to reported by Vasconcelos et al. (1999) and Moreira et al. earlier stages of the estrus cycle at initiation of the first (2000). Many protocols based on GnRH and PGF $_{2\alpha}$ has GnRH injection in the G6G group, reproductive been used for TAI in dairy cows. In the current study, both performance did not improve compared with the standard GnRH and PGF $_{2\alpha}$ (Ovsynch) were used for estrus Ovsynch protocol.

synchronization in dairy cows for TAI and obtained an In the present study, conception rate for high producing overall conception rate (CR) of 30% which appeared similar dairy cows was 40% and 36% while conception rate for low to that previously reported by Pursley et al. (1997a, 1997b); producing dairy cows was 20% and 44% for Ovsynch and Fricke et al. (1998) and Cordoba and Fricke (2001) and G6G synchronization protocol respectively which means slightly higher than 27% that reported by Bello et al. (2006) that Ovsynch synchronization protocol was more effective and lower than 37% and 38% that reported by Moreira et al. for high producing dairy cows which appeared in complete (2001) and El-Zarkouny et al. (2004) respectively.

In the current study, the using of PG and GnRH (G6G) followed after 6 days with the traditional Ovsynch protocol obtained an overall CR of 40% which appeared lower than 50% that reported by Bello et al. (2006) and Ribeiro et al. (2011). The rationale behind this “presynchronization” due to the effect of the first PG on the mature corpora lutea, strategy was based on a previous study (Vasconcelos et al., 1999) in which day of the estrus cycle when Ovsynch was initiated affected synchronization and conception rate. Lactating dairy cows in early (d 1 to 4) and late (d 17 to 21) stages of the estrus cycle exhibited larger follicles at ovulation and lower conception rates after TAI compared with cows initiating Ovsynch on d 5 to 9 of the estrus cycle (Vasconcelos et al., 1999). Furthermore, cows in the first half of the estrus cycle (d 1 to 12) exhibited a greater synchronization rate in response to Ovsynch than cows in the second half (d 13 to 21) of the estrus cycle (Vasconcelos et al., 1999). In a similar study conducted with dairy heifers, G6G (46.15%) might be due to the increased probability of initiation of Ovsynch during the early to midluteal phase of the estrus cycle (d 5 to 10) resulted in optimal pregnancy GnRH, and the elevated circulating progesterone rates compared with other stages of the cycle (Moriera et al., 2000). Thus, strategies to presynchronize cows so that initiation of Ovsynch occurs during the early luteal phase of the estrus cycle may improve reproductive performance of cows receiving Ovsynch and TAI (Moriera et al., 2000).

We hypothesized that administration of a luteolytic dose of PGF $_{2\alpha}$ at a random stage of the estrus cycle beginning 8 d before initiation of Ovsynch would induce luteal regression in those cows with a responsive CL, thereby increasing the proportion of cows that initiate Ovsynch on d 5 to 6 of the estrus cycle and decreasing the proportion of cows early in the estrus cycle (d 1 to 4) or during the later

agreement with (45.8 and 33.8% for high and low producing dairy cows, respectively) that reported by (Peters and Pursley 2002). While G6G synchronization protocol was slightly effective for low producing dairy cows (48%) than high producing cows (36%). The elevated conception rate in low producing dairy cows after using of G6G might be due to the effect of the first PG on the mature corpora lutea, which were more in the low producing cows than the high producing cows, and initiation of a new cycle before the second injection of GnRH. In the present study, conception rate for multiparous dairy cows was 30.55% and 37.8% for Ovsynch and G6G synchronization protocol respectively which appeared similar to 38% that was reported by (Souza et al., 2008) while conception rate for primiparous dairy cows was 28.57% and 46.15% for Ovsynch and G6G synchronization protocol respectively which appeared lower than 65% that reported by (Souza et al., 2008). The improved conception rate in the dairy cows treated with G6G (46.15%) might be due to the increased probability of a dominant follicle that would ovulate following the third GnRH, and the elevated circulating progesterone concentrations prior to the administration of the last PG.

CONCLUSION

Administration of PGF $_{2\alpha}$ 8 d before initiation of Ovsynch did not improve conception rate compared with the standard Ovsynch protocol although it is numerically increased. Milk production doesn't affect in the response to both protocols. Further research is needed to fully assess the efficacy of hormonal protocols for synchronization of ovulation and TAI for reproductive management of lactating dairy cows.

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