

## PROGESTERONE CONCENTRATIONS OF PREGNANT REPEAT BREEDER COWS FOLLOWING POST INSEMINATION PRID and GnRH TREATMENTS

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### Abstract

*The objective of this study is to determine the effects of application of progesterone (PRID) and GnRH injection after artificial insemination on serum progesterone concentrations in the pregnant repeat breeder cows. In this experiment, 45 repeat breeder Holstein cows were divided into 3 treatment groups. In group I, repeat breeder cows received an injection of GnRH on day 12 after artificial insemination. In group II, repeat breeder cows received a PRID from day 4 to day 11 and in group III, materials received a PRID from day 11 to day 18. Before and 7 days after the treatments, and on 21 days after artificial insemination blood samples were obtained from all treated cows to assay serum progesterone concentrations. In control group 15 repeat breeder cows did not receive any treatment. After artificial insemination, blood samples were periodically obtained from all control repeat breeder cows to assay serum progesterone concentrations.. Pregnancy rates were %20 in group I, %26,6 in group II, %40 in group III and %20 in group IV. There were no significant differences between four groups ( $P>0.05$ ). In all treatment groups progesterone concentration of pregnant repeat breeder cows are numerically greater than the progesterone concentration of pregnant repeat breeder cows in control group. In conclusion, serum progesterone concentrations of pregnant repeat breeder cows were numerically higher in treatment groups. But the treatments did not improve pregnancy rates in repeat breeder cows.*

**Key words:** Cow, Repeat breeder, PRID, GnRH

### INTRODUCTION

In dairy cows, luteal insufficiency and lower progesterone concentrations are known as a cause of embryonic mortality and reduce the pregnancy rates during early embryonic development (1-3).

During the pre-implantation phase of embryonic development, direct progesterone supplementations and GnRH / hcG injections are the approaches to improve embryonic survival in repeat breeder cows (4,5).

It has been hypothesized that increasing peripheral progesterone concentrations during the diestrus after insemination may improve embryo development and may suppress luteolysis, resulting in reduced embryonic loss (2,3). In controlled studies where relationship between early embryo development and maternal progesterone concentrations was examined, higher progesterone concentrations were associated with improved embryo development (6).

Delay in normal rise in progesterone concentrations between days 4 and 5 post

ovulation and low systemic progesterone concentrations during the subsequent diestrus reduce pregnancy rates and lower conception rates (2).

One approach to improve embryonic survival in repeat breeder cows is direct application of progesterone. Progesterone Releasing Intravaginal Devices (PRID), Controlled Internal Drug Release (CIDR) and Synchro-Mate ear implants are some of the preparations which are available (2,7).

Gonadotropin-releasing hormone (GnRH) injection is another approach to increase progesterone concentrations and enhance embryonic survival in repeat breeder cows after artificial insemination (3).

### MATERIAL AND METHOD

This study was conducted on 60 Holstein cows in three commercial dairy herds, located in the area of Nicosia and Famagusta in North Cyprus from May to September 2008. Cows included in the study were in their first to fifth lactations, producing an average of 9.000 kg milk

per a lactation period and had three to six unsuccessful inseminations within the current lactation (repeat-breeders). Cows were inseminated at estrus by veterinarians prior to group allocation. After artificial insemination 45 repeat breeder Holstein cows were divided into 3 treatment groups. In group I, repeat breeder cows received an injection of GnRH on day 12 after artificial insemination. In group II, repeat breeder cows received an intravaginal progesterone device (PRID) from day 4 to day 11 and in group III, animals received an intravaginal progesterone device (PRID) from day 11 to day 18. In group I, blood samples were obtained from the day of GnRH injection, after 7 day from the GnRH injection and after 21 day from the artificial insemination to assay serum progesterone concentrations. In group II and III, blood samples are obtained at the application day of PRID and also the removal day of PRID and after day 21 from the artificial insemination to assay the serum progesterone concentrations. In the control group 15 repeat breeder cows did not receive any treatment. After artificial insemination, blood samples were periodically obtained from all control repeat breeder cows to assay serum progesterone concentrations. Cows that were detected in estrus after day 18 were re-inseminated and recorded as non-pregnant to the prior artificial insemination. Cows not observed in estrus was palpated per rectum 45-50 days after insemination to determine pregnancy status. The differences in progesterone concentrations and pregnancy rates between treatment cows and control cows were analyzed by using Mann Whitney Test and program of SPSS 14.01.

## RESULTS

Of the 60 cows used in the study, 30 were allocated in the PRID-treated groups, 15 in the GnRH-treated group and 15 in the control group. In the GnRH-treated group, 3 cows were found pregnant and 12 non-pregnant at the rectal palpation on day 45 after the first inseminations. In the PRID-treated group between days 4-11, 4 cows and in PRID-treated group between days 11-18, 6 cows were found as a pregnant at rectal palpation. In the control group, 3 cows palpated as pregnant (Table 1).

Progesterone concentrations of pregnant repeat breeder cows in treated groups compared with progesterone concentrations

of pregnant repeat breeder cows in control group on application days of treatments, 7 days after applications and 21 days after artificial insemination (Table 2-4).

## DISCUSSIONS

Progesterone is an essential hormone in the maintenance of pregnancy in cows. The higher levels of progesterone concentration in the early pregnancy are related to the embryonic development and increase in interferon- $\tau$  production and pregnancy rates (8). Low systemic progesterone concentrations on day 5 post-ovulation or delay in normal rise in progesterone between days 4 and 5 post-ovulation have been related with reduced pregnancy rates (2, 6, 9, 10).

Several researchers believe that seven days between days 5-19 after insemination is sufficient for the application of progesterone (1, 2, 6, 10-14). In the above studies, intravaginal PRID or CIDR applications were mostly used and in this study as well PRID has been applied on two different groups of repeat breeder cows between days 4-11 and 11-18 for seven days.

In this study, repeat breeder cows (n=15) received progesterone application between days 4-11 after insemination. With this application on these days, it has been aimed to keep higher blood progesterone level during blastogenesis and allow time for the corpus luteum's progesterone secretion after being formed. In this study, another group of repeat breeder cows (n=15) received progesterone application between days 11-18 after insemination. On these days, it was aimed to stop corpus luteum's regression, support interferon- $\tau$  production and recognizing of pregnancy by cow. In both PRID-treated groups, progesterone concentrations of repeat breeder cows were found higher than the control group.

Gonadotrophic hormones are known alternatives of progestogen application and they also increase endogen progesterone level (2, 3).

In this study, GnRH hormone injection after insemination is used to increase the percentage of pregnancy in repeat breeder cows (15-17). GnRH injections also use for stimulating ovulation during insemination and it also increase the level of progesterone by providing luteal support in early and

medium luteal phases (18, 19). However there are different thoughts about the effect of increasing endogenous progesterone by using luteotropic hormon (17, 20, 21). Different results from GnRH applications on 11-14 days after insemination is related to follicular wave. In three-follicular wave cycles, GnRH applications on 11-14 days of cycle coincide with the period which secondary follicular wave is at peak and there is high estradiol concentration during this period. The effect of applied GnRH is detected as an atresiation, luteinization and ovulation of developed follicle in the 2 and 3-wave cycles (22).

In this study, treated group of repeat breeder cows (n=15) received GnRH injection on day 12 after insemination. The objective of GnRH injection on the 12<sup>th</sup> day

is to decrease the estradiol concentration which is high during this period by luteinizing dominant follicle and suppress the rise of oxytocin receptor and secretion of PGF<sub>2α</sub> in uterus. Many researchers also suggest days of 11 -13 for GnRH applications in luteal phase (18, 23-25).

In this study, blood progesterone concentrations of pregnant animals on the 19<sup>th</sup> day after insemination were numerically higher than the pregnant animals in control group but they were not statically significant. It was explained that the GnRH injections increased blood progesterone concentration by forming accessory corpus luteum. Several researchers support forming accessory corpus luteum after GnRH injections (3, 22, 26).

Table 1 Pregnant and non-pregnant cows in treatment groups and control group

Groups	No. of pregnant cows (%)	No. of non-pregnant cows(%)
Group I (GnRH-treated)	3/15 (20%)	12/15 (80%)
Group II (PRID-treated, 4-11 days)	4/15 (26.6%)	11/15 (73.3%)
Group III (PRID-treated,11-18 days)	6/15 (40%)	9/15 (60%)
Group IV (Control group)	3/15 (20%)	12/15 (80%)

Table 2. Progesterone concentrations on injection day, 7 days after injection and 21 days after artificial insemination in group I and control group

Groups	Pregnant repeat breeder cows		
	Progesterone (ng/ml)		
	Day 12	Day 19	Day 21
Group I (GnRH-treated)	13.12 ± 2.47	17.31 ± 1.10	15.93 ± 5.63
Group IV (Control group)	8.41 ± 2.58	5.61 ± 0.65	7.44 ± 1.15

Table 3. Progesterone concentrations on application day, removal day and 21 days after artificial insemination in group II and control group

Groups	Pregnant repeat breeder cows		
	Progesterone (ng/ml)		
	Day 4	Day 11	Day 21
Group II (PRID-treated, 4-11 days)	1.49 ± 0.44	9.82 ± 2.47	10.84 ± 3.31
Group IV (Control group)	0.92 ± 0.28	5.98 ± 2.30	7.44 ± 1.15

Table 4. Progesterone concentrations on application day, removal day and 21 days after artificial insemination in group III and control group

Groups	Pregnant repeat breeder cows		
	Progesterone (ng/ml)		
	Day 11	Day 18	Day 21
Group III (PRID-treated,11-18 days)	5.97 ± 1.02	6.88 ± 0.56	10.11 ± 1.44
Group IV (Control group)	5.98 ± 2.30	5.61 ± 0.65	7.44 ± 1.15

## CONCLUSIONS

In conclusion, serum progesterone concentrations of pregnant repeat breeder

cows were numerically higher in treatment groups. But the treatments did not improve pregnancy rates in repeat breeder cows.

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