



Effect of GnRH on the day of Insemination on Ovulatory Response in Crossbred Cows

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ABSTRACT

The effect of gonadotropin releasing hormone (GnRH) analogue on the ovulatory response of pre-ovulatory follicle (POF) during winter and summer months of the year was studied. A total of 120 crossbred cows brought for insemination with oestrus signs during winter months (December 2022 – January 2023; n=52) and summer months (April – May 2023; n=68) were studied. Oestrus (Day 0) was identified by gynaeco-clinical and ultrasound examination and animals were inseminated with (n =33) or without (n = 87) GnRH analogue (Buserelin acetate 10 mcg; im). The biometry of POF and regressing corpus luteum (RCL) were measured ultrasonographically on Day 0 and the POF was observed every 24 hours until ovulation. The diameters of the POF and RCL on Day 0 were similar between the periods of study. Ovulations occurred on the mean days of 1.00 and 1.37 ±0.18 in GnRH group during summer (S-Gn) and winter months (W-Gn) respectively. The respective mean days in non-GnRH group were 1.14 ±0.05 and 1.33±0.08 during summer (S-O) and winter months (W-O). In S-Gn group all the animals (100%) ovulated on Day 1, when compared to W-Gn animals (62.50%). On exploring the possible effect of RCL on ovulation, significantly increased percentages of ovulations occurred in the contralateral ovary to the ovary having the RCL in GnRH groups and vice-versa in non-GnRH groups. Due to increased percentage of induced ovulations, it could be assumed that GnRH administration on the day of insemination may be beneficial during the summer months of the year.

Keywords: Crossbred cattle, Insemination, GnRH, Ovulation, Season.

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INTRODUCTION

Ovulation of a competent oocyte at an appropriate time is a pre requisite for successful conception. However, in recent years, we could experience increased incidence of delayed ovulation along with prolonged oestrus in crossbred cows (unpublished data). It was reported that an injection of gonadotropin releasing hormone (GnRH) at the time of insemination at detected oestrus might reduce the delayed ovulation (Bloch *et al.*, 2006) and increase the proportion of cows ovulating with subsequent increase in progesterone concentration post-insemination (Mee *et al.*, 1993). Effect of GnRH administration at the time of insemination on pregnancy has been reported with varied results. A meta-analysis study concluded that GnRH at the time of insemination increased the pregnancy by 12.5 per cent (Morgan and Lean, 1993), while another study reported no benefits of GnRH in cows with relatively low pregnancy (Valenza *et al.*, 2012). The varied responses were attributed to the GnRH dose, the number of services and fertility status of cows (Hubner *et al.*, 2022). However, there were no reports on effects of season and corpus luteum (CL) proximity to follicle on the ovulatory response to GnRH at the time of insemination. Previous study by Satheshkumar *et al.* (2015) suggested alterations in steroidogenesis within the follicular microenvironment for the altered follicular biometry and lowered fertility during the hot season than the cold season. Based on which, it was hypothesized that seasonal changes in follicular micro-environment could alter the ovulatory response to GnRH. Hence, the present research was conducted to study the effect of GnRH analogue on the ovulatory response of oestral follicle during cold and hotter months of the year.

MATERIALS AND METHODS

A retrospective analysis of 120 pluriparous and lactating crossbred cows which were brought for insemination with oestrus signs during winter months (December 2022 – January 2023; n=52) and summer months (April – May 2023; n=67) to the Gynaecology Unit of Veterinary Clinical

Complex, Veterinary College and Research Institute, Orathanadu were carried out based on the records.

Oestrus (Day 0) was diagnosed based on the clinical signs and gynaeco-clinical examinations. The stage was confirmed by ultrasound examination based on the presence of dominant follicle and regressing CL (Satheshkumar, 2018). The cases were segregated based on insemination on Day 0 with (n =33) or without (n = 87) administration of GnRH analogue (Buserelin acetate 10 mcg; im). During summer (S-Gn) and winter(W-Gn) months, 17 and 16 animals were inseminated with GnRH, and in summer (S-O) and winter (W-O) months, 51 and 36 cows were inseminated without GnRH.

The biometry of pre-ovulatory follicle (POF) and regressing corpus luteum (RCL) were measured ultrasonographically on Day 0. The POFs were observed ultrasonographically in all the animals every 24 hours until ovulation. The ovulation was detected by the absence of POF in the subsequent examination (Satheshkumar *et al.*, 2012).

The data on POF and RCL biometry were subjected for one-way ANOVA statistical analysis. The data on the day of ovulation in relation to GnRH administration and proximity of RCL in relation to POF was analysed by Chi-Square (2 x 2 contingency) test (Snedecor and Cochran, 1994).

RESULTS AND DISCUSSION

The follicular and RCL biometries during oestrus in summer and winter months were presented in Table 1. Perusal of the data indicated that the diameter of the OF on Day 0 was non-significantly ($P > 0.05$) larger during summer months (12.19 ± 0.21 mm) than winter months (11.58 ± 0.32 mm), which was in concurrence with the previous reports of Satheshkumar *et al.* (2015). There were no significant ($P > 0.05$) differences in the diameter of RCL between the two periods (9.63 ± 0.21 and 9.56 ± 0.22 mm respectively). OFs were observed majorly in the right ovary during the hotter (60.76%) and colder (54.17%)

Table 1: Mean \pm SE of follicular and RCL diameter on the day of oestrus (Day 0)

	Diameter of RCL (mm)	Diameter of POF (mm)	Side of POF	
			Right ovary (%)	Left ovary(%)
Winter (n=52)	9.56 \pm 0.22	11.58 \pm 0.32	54.17	45.83
Summer (n=67)	9.63 \pm 0.21	12.19 \pm 0.21	60.76	39.24
Significance	NS	NS		

NS: Not significant ($P > 0.05$)

months. Similarly, Stevenson (2019) reported increased incidence of dominant follicles in the right ovary than in the left ovary. Explanations for right-dominated ovulations in ruminants often included the proximity of the left ovary to the rumen and other extrinsic factors such as temperature or pressure fluctuations and mechanical contractions of the rumen (Pineda, 1989).

The ovulatory response of the POF in animals inseminated on Day 0 with or without GnRH was presented in Table 2. Irrespective of the season, 78.16 and 81.81 per cent of cows had ovulated on Day 1 in non-GnRH and GnRH groups respectively, while rest of the animals ovulated on Day 2. The present values on Day 1 ovulation were much higher than Burnett *et al.* (2022), who reported only 22.4 – 28.4 percent ovulations by 24 h post insemination without GnRH and 15.4-36.1 per cent ovulations in GnRH group. They correlated the findings with the intensity of oestrus expression.

In the current study, ovulations occurred on the mean days of 1.00 ± 0.00 and 1.37 ± 0.18 in GnRH group during hotter and colder months, while ovulations occurred on the mean days of 1.14 ± 0.05 and 1.33 ± 0.08 during the respective periods in non-GnRH groups respectively. There were no significant ($P > 0.05$) differences in the mean day of ovulation between the groups. The finding corroborated with the reports of Ryan *et al.* (1994) who recorded that administration of busserelin acetate along with insemination has not affected the interval from the onset of oestrus to ovulation. The administration of GnRH was expected to have an effect on LH secretion and pulse frequency (Bloch *et al.*, 2006). Previous reports stated that, GnRH injection at the onset of oestrus was found to increase the intensity of the preovulatory LH surge and shorten the interval from the onset of oestrus to the LH surge and subsequently ovulation (Kaim *et al.*, 2003). However, in our study GnRH was administered only when the animal was presented for insemination, which was much later from the onset of

oestrus. Hence the timing of ovulations in cows between GnRH and non-GnRH groups were not much affected in the present study. In summer months all the animals in GnRH group (100%) ovulated on Day 1, when compared to colder months (62.50%).

Table 3: Ovulations in relation to proximity of RCL in crossbred cows inseminated during winter and summer months.

Groups	Side of ovulation in relation to RCL proximity	
	Ipsilateral	Contralateral
Winter (n=52)	35 (67.30%)	17 (32.70%)
Summer (n=68)	53 (77.94%)	15 (22.06%)

Percentage in parenthesis; X^2 : $P < 0.05$.

The relation between RCL proximity and ovulation was presented in Table 3. On perusing the possible effect of RCL on ovulation, significantly ($P < 0.05$) increased percentages of ovulations were observed in POFs on the ipsilateral ovary to the ovary having the RCL, in both the seasons. The findings corroborated with Stevenson (2019) who also reported that ovulatory follicles in cows tended to be ipsilateral (54.8%) to the CL more often than contralateral (45.2%). Further research has to be conducted to explore the possible effect of follicle-RCL relationship on ovulation.

CONCLUSIONS

From the study, it was observed that majority of ovulations occurred in right ovaries and there were no significant differences in the mean day of ovulations in animals inseminated with or without GnRH. However, based on the findings it could be assumed that GnRH administration on the day of insemination may be beneficial in terms of earlier ovulation during the hotter months of the year.

Table 2: Effect of season on ovulatory response and mean days elapsed to ovulation.

Groups	No. of animals ovulated on Day 1		No. of animals ovulated on Day 2		Mean Day of ovulation
W-O (n=36)	24 (66.67%)	68 (78.16%)	12 (33.33%)	19 (21.84%)	1.33±0.08
S-O (n=51)	44 (86.2%)		7 (13.80%)		1.14 ± 0.05
W-Gn (n=16)	10 (62.5%)	27 (81.81%)	6 (37.50%)	6 (18.19%)	1.37±0.18
S-Gn (n=17)	17 (100%)		0		1.00 ± 0.00
Significance					NS

Percentage in parenthesis; X^2 : $P < 0.05$; NS: Not significant ($P > 0.05$);

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CONFLICT OF INTEREST

None.

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