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Serum mineral changes in repeat breeder cows treated with dexamethasone and insulin

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Abstract
Seventy-four Postpartum crossbred cows aged between 2nd and 6th parity were selected and randomly divided into 3 experimental groups *viz*., control group, group II and III. In control group, the cows (n=50) were not subjected for any treatment protocol. In group II (Dexamethasone) cows were received, Inj Dexamethasone - 40 mg I.M, for three days from Day 8 to Day 10 of the cycle (Day 0 – oestrus). Inj. Prostaglandin - 500 mcg, I.M, on Day 11. On the day of induced oestrus (Day 0i), all the 12 cows were randomly divided into 2 subgroups *viz*., sub group 1(Dex-GnRH): On the day of induced oestrus (Day 0i), inj. GnRH- 10 mcg I.M, was administered along with insemination and subgroup 2 (Dex-GnRH-Insulin) GnRH -10 mcg I.M and Insulin - 0.25 units /kg b. wt S.C., was administered along with the insemination. In group III (insulin) were administered with Biphasic Insulin -0.25units /kg BW S.C., for three days from Day 8 to Day 10 of the cycle (Day 0 – oestrus)' inj. Prostaglandin - 500 mcg, I.M. was administered on Day 11. On the day of induced oestrus (Day 0i), all the 12 cows were randomly divided into 2 subgroups *viz*., Ins-GnRH: On the day of induced oestrus (Day 0i), inj. GnRH - 10 mcg i.m. was administered along with insemination; Insulin-GnRH-Insulin: inj. GnRH -10 mcg i.m. and inj. Insulin - 0.25 units /kg BW s.c. was administered along with the insemination. In both the treatment subgroups, animals were re-inseminated on the second day and if the DF persisted, they inseminated on third day also. Serum Ca concentrations were lower on the day of mid cycle (Day 10) in both the treatment groups when compared to control group. Statistical analysis revealed a significant decrease (p<0.05) in DEX group when compared with control group. On the day of induced oestrum (Day 0i), the Ca and P levels were significantly (p<0.01) lower in INS group than DEX group.

Keywords: Serum minerals, repeat breeder cows, dexamethasone and insulin therapy

Introduction
Repeat breeding syndrome (RBS) is one of the frustrating gynaecological maladies of crossbred cows, leading to infertility (Rangnekar *et al.*, 2002) [3]. The incidence of this RBS problem has been reported to be 10 - 25% in exotic as well as Indian breeds (Sharma *et al*., 1988) [4]. Bhat *et al*. (2012) [5] reported that the highest prevalence of repeat breeding was recorded in crossbred Jersey (28.39%) followed by crossbred Holstein Friesian cows (HF) (27.62%) and local cows (27.27%). However, Talukdar *et al*. (2016) [15] reported that the higher incidence of repeat breeding was recorded in Holstein crossbred (57.72%) than Jersey crossbred (35.77%) and lowest incidence recorded in local cattle (6.61%). El-Shahata and Maatyb (2010) [8] emphasized the crucial role of calcium in enhancing the number and size of ovarian preovulatory follicles, as well as influencing the ovulation rate. They suggested that low calcium levels in acyclic animals might be due to a failure to maintain normal calcium levels resulting from metabolic disturbances or increased calcium excretion. Elevated levels of dietary phosphorus can lead to reduced serum calcium levels. Hyperphosphatemia, characterized by high phosphorus levels, lowers calcemia. This occurs through the inhibition of the renal alpha-hydroxylase enzyme, responsible for converting 25-OH-vitamin D to 1.25-diOH-vitamin D (DHCC), ultimately resulting in decreased intestinal calcium absorption (Lean *et al*., 2006; Goff *et al*., 2014) [7, 9]. Lager & Jordan (2012) [10] recommend maintaining phosphorus levels within a range of 4.8-7.4 mg/dL.

Materials and Methods

The study was conducted in the crossbred cows brought with the history of infertility to the Large Animal Gynaecology Unit of Veterinary Clinical Complex, Veterinary College and Research Institute, Orathanadu, Thanjavur (DT) from in and around Thanjavur district of Tamil Nadu. Pluriparous (2^{nd} – 6^{th} parity) crossbred cows which were reported to be in oestrus and failed to conceive after three or more consecutive inseminations were selected for the study. Selected and randomly divided into 3 experimental group's *viz*., control group, group II and III. In control group, the cows (n=50) were not subjected for any treatment protocol. In group II (Dexamethasone) cows were received, Inj Dexamethasone - 40 mg I.M, for three days from Day 8 to Day 10 of the cycle (Day 0 – oestrus). Inj. Prostaglandin - 500 mcg, I.M, on Day 11. On the day of induced oestrus (Day 0i), all the 12 cows were randomly divided into 2 subgroups *viz*., sub group 1(Dex-GnRH): On the day of induced oestrus (Day 0i), inj. GnRH- 10 mcg I.M, was administered along with insemination and subgroup 2 (Dex-GnRH-Insulin) GnRH -10 mcg I.M and Insulin - 0.25 units /kg b. wt S.C., was administered along with the insemination. In group III (insulin) were administered with Biphasic Insulin -0.25units /kg BW S.C., for three days from Day 8 to Day 10 of the cycle (Day 0 – oestrus)' inj. Prostaglandin - 500 mcg, I.M. was administered on Day 11. On the day of induced oestrus (Day 0i), all the 12 cows were randomly divided into 2 subgroups *viz*., Ins-GnRH: On the day of induced oestrus (Day 0i), inj. GnRH - 10 mcg I.M. was administered along with insemination; Insulin-GnRH-Insulin: inj. GnRH -10 mcg I.M. and inj. Insulin -0.25 units /kg BW S.C. was administered along with the insemination. Blood collection carried on Day 0 (oestrum) and Day 10 in control group. Whereas in Group II and III (treatment) Blood collection carried out on Day 0 (oestrum), Day 10 and day of induced oestrum (DOi).

Results and Discussion

Minerals play a crucial role in facilitating the actions of hormones and enzymes at the subcellular level, operating in a coordinated manner to regulate the reproductive and productive functions of domestic animals. Notably, minerals such as calcium, phosphorus, and magnesium affect the animal's capacity to utilize other essential trace minerals (Amle *et al*., 2014) [1].

The mean (±SE) serum calcium concentrations (mg/dl) in experimental and control groups cows during different days of blood collection are depicted in Table 1and the mean (±SE) serum phosphorus concentrations (mg/dl) in experimental and control groups cows during different days of blood collection are depicted in Table 2. The mean serum calcium level was found to be significantly higher in Day 10 and Day 0i group repeat breeder crossbred cows with highest value of 7.32±0.23 b mg/dL in control groups and the lowest value of 5.72±0.98 a mg/dL in DEX group. This is in contrast with earlier reports where significantly lower levels (Ashturkar *et al*., 1995 and Burle *et al*., 1995) [2, 11] or non-significant difference between two groups were observed (Sharma *et al*., 1984 and Singh and Pant, 1998) [14, 13]. Calcium dependent mechanisms are involved in steroid biosynthesis in ovaries and LH release from pituitary (Hurley and Doane, 1989) [12]. The higher serum calcium level may probably alter the biochemical environment of the uterus which may not be conducive for fertilization and embryo survival and this aspect needs further studies. The mean serum calcium level was found to be non-significantly between control and treatment groups in all days except significant difference (p<0.01) noticed between treatment group II and III on day of induced oestrum. Phosphorus is crucial for the transfer of biological energy, especially through adenosine triphosphate. A deficiency in phosphorus can impede the fertilization process, potentially leading to early embryonic death and resulting in conditions like repeat breeding and anoestrus in bovines.

Table 1: Comparison of serum Calcium values between control and treatment groups of repeat breeding crossbred cows

Sl. No.	Days of cycle	Calcium (mg/dL)			Significance	F value
		Control	DEX	INS		
1.	Day 0	7.28±0.30	6.67±0.46	6.85±0.93	NS	153.653
2.	Day 10	$7.32±0.23^{b}$	$5.72±0.98^{a}$	$6.60±0.46^{ab}$	*	178.076
3.	Day 0i	-	$8.22±0.80^{b}$	$6.42±0.46^{a}$	**	183.539

Values within the row with different superscripts differ significantly ** (p<0.01) * (p<0.05) NS – Not significant (p>0.05)

Table 2: Comparison of serum Phosphorus values between control and treatment groups of repeat breeding crossbred cows

Sl. No.	Days of cycle	Phosphorus (mg/dL)			Significance	F value
		Control	DEX	INS		
1.	Day 0	3.28±0.20	4.06±0.19	3.07±0.52	NS	12.244
2.	Day 10	$3.13±0.13^{a}$	$3.18±0.25^{b}$	$2.61±0.24^{a}$	NS	13.051
3.	Day 0i	-	$3.81±0.20^{b}$	$2.60±0.17^{a}$	**	25.994

Values within the row with different superscripts differ significantly ** (p<0.01)

Conclusion

In this study findings suggest that the mean serum calcium level was found to be significantly higher in Day 10 and Day 0i group repeat breeder crossbred cows with highest value of 7.32±0.23 b mg/dL in control groups and the lowest value of 5.72±0.98 a mg/dL in DEX group. The mean serum phosphorus level was found to be non-significantly between control and treatment groups in all days except significant difference (p<0.01) noticed between treatment group II and III on day of induced oestrum in repeat breeder crossbred cows.

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