Short Communication

FOLLICULAR DYNAMICS IN ANOESTRUS CROSSBRED COWS

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ABSTRACT

In-vivo follicular developmental pattern was characterized in postpartum anoestrus (PPA) crossbred cows using the ultrasound imaging technology. A total of 12 PPA and six normally cycling cows were selected and investigated. In PPA cows, the ultrasonographic examination of ovaries was initiated randomly on any day and examined every other day continuously for a period of 25 days. In cyclic animals, the ovaries were ultrasonographically monitored every other day from the day of observed oestrus (Day 0) to Day 10 targeting the dominant follicle (DF) of first follicular wave. Ultrasonographic monitoring of the follicular turnover in PPA cows revealed that both the ovaries exhibited dynamic follicular activity. A maximum of two waves could be recorded during the observation period of 25 days, with each wave extending for a period of 8 – 10 days. The follicular wave of PPA exhibited coordinated growth, static and regression phases, with significantly ($P < 0.05$) prolonged static phase than their counterpart in normally cycling cows. Eventhough the DFs attained >10mm diameter, the animal did not evince any symptom of oestrum which might be due to deficient steroidogenic capacity of the follicular cells as a result of aberrant follicular microenvironment.

Keywords: Follicular dynamics, Postpartum anoestrus, Crossbred cows

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Ovarian rebound in postpartum period is a crucial event which determines the inter-calving interval in the farm animals especially cattle and buffaloes. Unfortunately, postpartum anoestrus (PPA) is an ovarian dysfunction which delays the resumption of cyclicity after parturition and is attributed to many factors ranging from negative energy balance to stress of any origin (Opsomer *et al*., 1996). Ovaries of acyclic cattle were often referred to be inactive (Roberts, 1971). However, with the advent of imaging diagnostics, it was found that acyclic animals also displayed variable degree of ovarian activity, characterized by follicular turnover in one or both ovaries but without expression of oestrus signs, which

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was documented in buffaloes (Ghuman *et al*., 2010; Das *et al*., 2013). However, report on follicular turnover in acyclic Indian crossbred cows is scanty. Hence the present research was undertaken to characterize *in-vivo* follicular developmental pattern in anoestrus crossbred cows using ultrasound imaging technology.

Healthy lactating crossbred Jersey cows (5-6 yrs; second and third lactation) maintained at the Post Graduate Research Institute of Animal Sciences, Kattupakkam and University Research Farm, Madhavaram of Tamilnadu Veterinary and Animal Sciences University were monitored regularly. The animals which have not expressed oestrus signs even after three months of parturition were subjected to gynaeco-clinical and ultrasonographic examination. The animals which had smooth ovaries with no luteal structure in both the ovaries were determined as true anoestrus animals (Roberts, 1971). A total of 12 PPA crossbred cows were selected for further investigation. The ultrasonographic examination (Real time B-mode ultrasound scanner -SONOVET 600- equipped with 7.5 MHz transrectal transducer) of ovaries was initiated randomly on any day and examined every other day continuously for a period of 25 days. As a positive control, normal follicular turnover was studied in six lactating crossbred cows maintained at Centralized Embryo Biotechnology Unit, Department of Animal Biotechnology, Madras Veterinary College, Chennai. A total 12 cycles (two cycles / animal) were studied. The ovaries were ultrasonographically monitored every other day from the day of observed oestrus (Day 0) to Day 10 targeting the emergence and regression of first follicular wave (Wave I) dominant follicle (DF). The Wave I was studied because it is more consistent in all the cycles and DF is anovulatory in nature (Satheshkumar, 2015). The day-to-day identity of follicles was profiled in all the animals, based on which the parameters of follicular development viz., maximum diameter, growth phase, growth rate, static phase, regression phase and regression rate were arrived (Satheshkumar, 2015) and statistically analysed for significance as described by Snedecor and Cochran (1994).

The developmental pattern of DF in the PPA crossbred cows was represented in Fig. Ultrasonographic monitoring of the follicular turnover in PPA crossbred cows revealed that both the ovaries exhibited extensive follicular activity and the DFs developed in a wave-like fashion similar to that of normally cycling animals. The findings were in agreement with the observations of Ghuman *et al*. (2010) and Das *et al*. (2013) in buffaloes. A maximum of two waves could be recorded during the observation period of 25 days, with each wave extending for a period of 10 - 14 days. Our study has confirmed that the phenomenon of follicular growth and atresia continues even in PPA condition among the crossbred cows. To the best of our knowledge this is the first ultrasonographic documentation of follicular dynamics in acyclic Indian crossbred cows.

The characteristics of DF during a wave in the PPA and normally cycling crossbred cows were presented in Table. The follicular wave of PPA exhibited coordinated growth, static and regression phases as that of normally cycling cows. Perusal of the data revealed that, there are no significant differences in the parameters between the

cyclic and acyclic animals except for the static phase of the DF. The static phase was significantly ($P < 0.05$) prolonged in anoestrus animals than their counterpart in normally cycling cows. Absence of suppressive effect from the corpus luteum (Satheshkumar *et al*., 2012) would have led to the sustenance of DF for an extended period in acyclic cows.

It was very obvious that the DF could reach a diameter of >10mm, but the animal did not evince any symptom of oestrum. The DF proceeded regressed after reaching the maximum diameter. Inadequate plane of nutrition or stress during the postpartum period leads to reduced concentrations of serum LH, fewer pulses of LH, reduced plasma concentrations of insulin-like growth factor I (IGF-I) and insulin which have direct effects on the ovarian follicular function (Braw-Tal *et al*., 2009). Recently, an *in-vitro* proteomic analysis of follicular microenvironment revealed that failure of the intra-follicular IGF / IGFBP system, led to ovulatory disturbances in acyclic buffaloes (Satheshkumar *et al*., 2019).

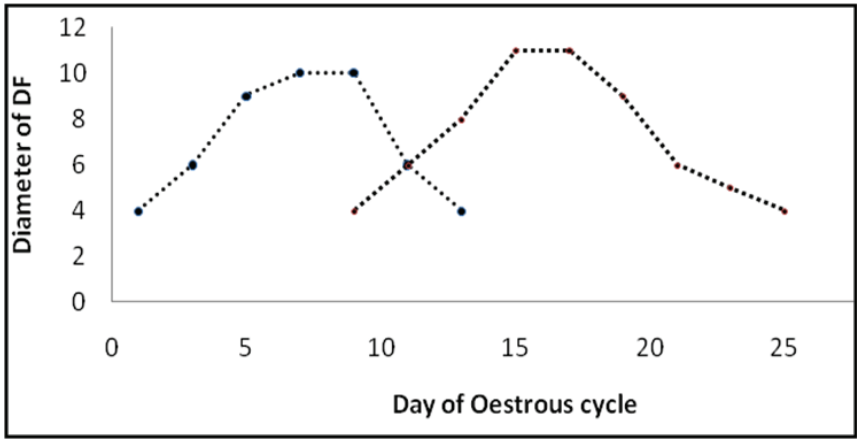

Fig. Follicular wave pattern in anoestrus crossbred cows

Table. Follicular turnover in anoestrus crossbred cows

S. No	Characteristics of Dominant follicle during a wave	Anoestrus	Cyclic (Wave I)	Significance
1	Growth phase (days)	5.6 ± 0.2	6.0 ± 0.0	NS
2	Growth rate (mm / day)	1.1 ± 0.3	1.7 ± 0.1	NS
3	Maximum diameter (mm)	10.2 ± 0.3	10.6 ± 0.6	NS
4	Static phase (days)	2.6 ± 0.2 [b]	1.7 ± 0.7 [a]	*
5	Regression phase (days)	4.8 ± 0.3	6.3 ± 1.8	NS
6	Regression rate (mm / day)	1.3 ± 0.3	1.1 ± 0.3	NS

a,b - Values with different superscripts within a row vary significantly ($P<0.05$)

*Significant ($P<0.05$); NS - Not significant

The deficient metabolic and endocrine status of the follicular microenvironment would have affected the steroidogenic capacity of the follicular cells and the oestrogen production thereon. Development of oestrogenically inactive large follicles and lack of ovulation were found to be the major factors for the incidence of PPA in crossbred cows

It is concluded that the follicles develop in wave-like fashion in PPA cows, similar to that of normally cycling animals, but the DFs did not destine to ovulation resulting in acyclicity. The finding warranted further molecular analyses of follicular microenvironment to arrive at possible causes of aberrant steroidogenesis and thus aiding in designing protocols to alleviate the condition accordingly.

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