

Characterization of follicular dynamics and luteal function during short estrous cycle in suckled postpartum Mehsana buffaloes*

M. K. AWASTHI¹, F. S. KAVANI², G. M. SIDDIQUEE³, A.J. DHAMI⁴ AND M.T. PANCHAL¹

Department of Animal Reproduction, Gynaecology & Obstetrics
College of Veterinary Science & Animal Husbandry,
Anand Agricultural University, Anand, 388 001 (Gujarat)
E-mail: mkawasthidurg@indiatimes.com

ABSTRACT

The present study was aimed to characterize the pattern of follicular growth and progesterone concentration during short estrous cycle in postpartum suckled water buffaloes. Twenty freshly calved Mehsana buffaloes were selected and animals (n=3) showing short estrous cycle were scanned daily for ovarian follicular development from day of first postpartum estrus to next using a real-time B-mode ultrasound scanner equipped with a 6.5 MHz convex linear array transducer. The blood samples were collected from each animal on Days 3, 7 and 10 after first postpartum estrus for estimation of plasma levels of progesterone. These animals showed one-wave follicular growth during short estrous cycle and short luteal phase with mean plasma progesterone concentration of 0.49, 0.80 and 0.26 ng/ml on Days 3, 7 and 10 post-estrus, respectively, suggesting subnormal functioning of CL. It is concluded that short estrous cycle is characterized by one-wave of follicular development observed in few animals during early postpartum period in suckled water buffaloes.

Key words: Follicular dynamics, Postpartum buffaloes, Short estrous cycle, Short luteal phase, Ultrasonography

INTRODUCTION

Short estrous cycle has been reported in postpartum cattle (Williams and Ray, 1980; Perry *et al.*, 1991) and buffaloes (Baruselli *et al.*, 1997) with short luteal phase showing subnormal blood progesterone concentrations (Manns *et al.* 1983). Analyses of various ultrasound studies on follicular dynamics show that water buffaloes have one-wave (Taneja *et al.*, 1996; Awasthi *et al.*, 2006) or two- and three-wave patterns of follicular growth (Baruselli *et al.*, 1997; Manik *et al.*, 1998; Awasthi *et al.*, 2006) during estrous cycle of normal length. However, the follicular dynamics associated with first postpartum spontaneous short estrous cycle

has not been reported in buffaloes. In perspective, the aim of present experiment was to characterize the follicular development and luteal function during short estrous cycle in water buffaloes at early postpartum period.

MATERIALS AND METHODS

Experimental animals:

Freshly calved Mehsana buffaloes (n=3) with normal parturition and displaying short estrous cycle length were selected at Livestock Research Station, S D Agricultural University, Sardar Krushinagar, Gujarat for present study. The experiment was conducted during breeding season from September to February. The animals were observed twice daily for behavioural signs of estrus soon after parturition with teaser buffalo bull. The buffaloes were considered in estrus only when they accepted bull mounting.

*Part of Ph.D. Thesis ¹Associate Professor, Dept. of ARGO, CVSc&AH, Anjora, Durg, P.B. No. 6 (C.G.) ²Professor and Head, ³Associate Professor, ⁴Professor, Department of ARGO, CVSc&AH, Sardar Krushinagar, Gujarat ⁵Research Scientist, Livestock Research Station

Ultrasound

The performed scanner (Medical, S convex linear array transducer placed in the rectum to visualize the ovaries. Several planes were scanned, more than 4 ml of the sample was frozen on dry ice and used for analysis using a buffalo. The postpartum ovulation after a short estrous cycle was observed in at least one other follicle in the cohort) was observed in the dominant follicle (Kn) to grow and was recorded.

Plasma p

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Ultrasound scanning:

The ultrasound examinations were performed using a real-time B-mode ultrasound scanner (Sigma-110 Master-Vetson, Kontron Medical, SAS, France) equipped with a 6.5 MHz convex linear array transducer designed for intra-rectal placement. The transducer was placed over the ovaries and scanning was accomplished in several planes to identify all the follicles greater than 4 mm in diameter. Desired images were frozen on the screen and measurements were taken using a built in caliper system. The ovaries of each buffalo were scanned daily from Day 14 postpartum onwards until first postpartum ovulation and soon after detection of first postpartum behavioural estrus to next ovulation after a short estrous cycle length. The dominant follicle was characterized as the one, which grew to at least 10 mm and exceeded the diameter of other follicles. Subordinate follicles (follicular cohort) were defined as those that appeared to originate from the same follicular pool as the dominant follicle (Baruselli *et al.*, 1997). A dominant follicle and its cohort were defined as a wave (Knopf *et al.*, 1989). The day of emergence of a follicular wave was defined by first follicle to grow above 4 mm (Ginther, 1995). Ovulation was recorded as described by Nasser *et al.* (1993).

Plasma progesterone assay:

About 10-15 ml of jugular blood sample was collected in heparinized vials from each animal on Days 3, 7 and 10 after first postpartum estrus for estimation of plasma levels of progesterone. Plasma was separated out and stored frozen at -20°C till analyzed. One or two drops of 0.01% merthiolate (thiomersal) was added as preservative. Progesterone concentration was estimated in plasma samples using a radioimmunoassay (RIA) kit employing standard technique.

Statistical Analysis:

The retrospective data of animals showing short estrous cycle (n=3) were utilized for present study. The growth rate of the dominant follicle

was calculated by linear regression analysis using a Microsoft Excel computer program. The data for wave emergence, number of follicles in a wave and follicular diameter were analyzed following statistical methods described by Snedecor and Cochran (1986). These data were compared with corresponding data of one-wave follicular growth during normal estrous cycle in another study (Awasthi *et al.*, 2006).

RESULTS AND DISCUSSION

Short estrous cycle was observed in three animals soon after parturition in the present study. First postpartum ovulation was detected on Days 16, 22 and 25 postpartum, respectively, in these animals without overt signs of estrus and subsequently they showed first postpartum ovulatory estrus on Days 24, 31 and 33 postpartum with short luteal phase of 8, 9 and 8 days, respectively. The successive estrus was observed on Days 34, 42 and 43 postpartum with estrous cycle length of 10, 11 and 10 days, respectively.

The follicular turnover studied in three cyclic buffaloes with short estrous cycle was characterized by the emergence of one wave of follicular growth beginning at different times after preceding ovulation. The solitary follicular wave emerged on day 1.33 ± 0.27 after preceding ovulation (ovulation = day 0, Table 1 and Fig. 1). The mean number of follicles (size ≥ 4.00 mm) recruited during one-wave cycle was recorded as 3.66 ± 0.27 . The solitary dominant follicle showed a steady linear growth rate of 0.73 ± 0.15 mm/day before ovulation. The mean maximum diameter of the ovulatory follicle was recorded as 12.56 ± 0.23 mm. The mean diameter of the ovulatory follicle was significantly greater ($P < 0.05$) than the diameter of the largest subordinate follicle on day 4 of wave emergence and remained significantly greater ($P < 0.01$) until ovulation on Day 10 or 11. The mean plasma progesterone concentration was recorded as 0.49 ± 0.01 , 0.80 ± 0.02 and 0.26 ± 0.02 ng/ml on Days 3, 7 and 10 post-estrus, respectively.

The short estrous cycles recorded during

Table 1. Characteristics (mean ± SE) of short estrous cycle in Mehsana buffaloes having one-wave follicular growth (n=3)

Characteristics	Observation
Emergence of follicular wave (days)	1.33 ± 0.27 (1-2)
Mean number of follicles (size > 4.00 mm) per follicular wave	3.66 ± 0.27 (3-4)
Linear growth rate of ovulatory follicle (mm/day)	0.73 ± 0.15
Mean maximum diameter (mm) of ovulatory follicle	12.56 ± 0.23 (12.0-12.9)
Duration of short estrous cycle (days)	10.33 ± 0.27 (10-11)

Data in parentheses indicates range

early postpartum period (day 24-43) corroborated well with the report of Short *et. al.* (1990), who concluded that short estrous cycles commonly occur during the first 30 to 40 days after parturition. The incidence of short estrous cycle has been reported previously also in buffaloes (Usmani *et. al.*, 1985; Baruselli *et. al.* 1997).

The animals exhibiting short estrous cycle had one-wave follicular growth in present study, which corroborates with early report in buffaloes (Baruselli *et. al.*, 1997). The days of follicular wave onset were nearly same as reported for one-, two-, or three-wave follicular growth during normal estrous cycle in buffaloes (Taneja, *et. al.*, 1996; Baruselli *et. al.*, 1997, Awasthi *et. al.*, 2006). Similarly, daily growth rate of ovulatory follicle of solitary wave during short estrous cycle did not differ significantly with that of ovulatory follicle during growth phase of one-wave cycle of normal estrous cycle length (0.73 ± 0.15 vs 0.65 ± 0.25 mm/day, Awasthi *et. al.* 2006). Likewise, non-significant difference was observed between mean maximum diameter of ovulatory follicle during short estrous cycle in present study and that of one-wave cycle of normal estrous cycle length in another study (12.56 ± 0.23 vs 12.9 ± 0.59 mm, Awasthi *et. al.* 2006). However, the pattern of one-wave follicular growth of short estrous cycle differed to that of one-wave cycle of normal estrous cycle length. The ovulatory follicle of solitary wave of short estrous cycle exhibited only growth phase after its emergence before ovulation, whereas three distinct phases of follicular development were observed in animals with one-

wave cycle of normal estrous cycle length, viz. growth phase, regression phase and atypical "regrowth phase" (Awasthi *et. al.* 2006). With early demise of corpus luteum the first dominant follicle ovulated during short estrous cycle as the plasma progesterone concentration declined with regression of corpus luteum. Under normal circumstances with one- or two-wave follicular growth of normal estrous cycle length the first dominant follicle does not ovulate with concurrent greater plasma progesterone concentration during mid cycle (Awasthi *et. al.* 2006).

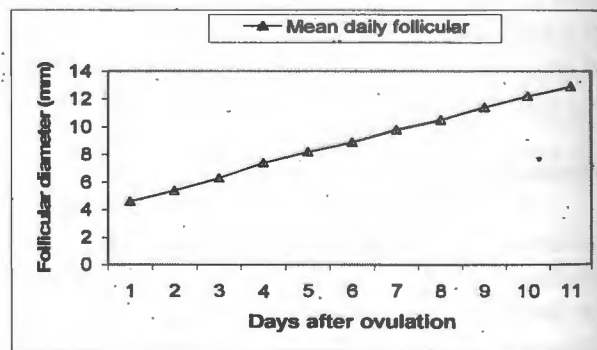


Fig.1 Mean growth profile of dominant follicle in Mehsana buffaloes during short estrous cycle

There was no significant difference in size between the future dominant follicle and the largest subordinate follicle at the time of wave emergence in one-wave follicular growth during short estrous cycle in present study. However, dominant follicle grew at a faster rate and its diameter was significantly greater by day 4 after wave emergence. This is in accordance with the observations reported in cattle (Kulick *et. al.*,

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1999; Ginther *et al.*, 2000) and buffaloes (Awasthi *et al.*, 2006) in one-, two- and three-wave follicular growth of normal estrous cycle length. The present observation further suggests that the pattern of growth profile of dominant and/or ovulatory follicle and their largest subordinate follicle is similar for both short as well as normal estrous cycle.

The animals with short estrous cycle length showed a short luteal phase and had lower plasma progesterone concentration (< 1.0 ng/ml) on Days 3, 7 and 10 post-estrus indicating failure of proper formation of corpora lutea. Short luteal phase has been reported at the onset of estrous cycle after a postpartum anestrus in both cattle (Savio *et al.*, 1990; Burke *et al.*, 1994) and water buffaloes (Usmani *et al.*, 1985; Baruselli *et al.*, 1994). Following first postpartum ovulation, plasma progesterone levels are correlated with morphologic and functional activity of CL in buffaloes (Jainudeen and Hafez, 2000). The early decline of plasma progesterone level in such cycles is attributed to short life span of CL (Hafez and Hafez, 2000). It would appear that the corpora lutea associated with short estrous cycles have short life span as a result of (a) lack of luteotrophic support, (b) failure of luteal tissue to recognize a luteotropin and/or (c) enhanced secretion of luteolytic agent, mostly PGF_{2α}, by involuting uterus during early postpartum period (Lishman and Inskeep 1991, Garverick *et al.* 1992).

In conclusion the short estrous cycle was observed in 3 out of 20 postpartum buffaloes with short luteal phase. These animals showed one-wave follicular growth during short estrous cycle. The short estrous cycle is a usual phenomenon observed in few animals during early postpartum period in suckled water buffaloes.

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