## HORMONAL AND CLINICAL MONITORING OF POSTPARTUM OVARIAN FUNCTION IN SURTI BUFFALOES

R.G. SHAH, A.J. DHAMI AND F.S. KAVANI

Department of Animal Reproduction, Gynaecology and Obstetrics College of Veterinary Science and Animal Husbandry Anand Agricultural University, Anand - 388 001 Received 15-6-2012 Accepted 28-9-2012

### ABSTRACT

The hormonal (P4 and E2) profile and ovarian changes of 39 postpartum Surti buffaloes were studied at weekly interval from calving to at least 91 days postpartum with a view to characterise the ovarian function. The profile of both P4 and E2 hormones showed sequential biweekly trend. As against calving, the mean plasma progesterone concentration was significantly higher (4.39  $\pm$  0.67 vs 1.19  $\pm$  0.33 ng/ml) and that of estradiol was lower (6.77  $\pm$  1.39 vs 153.05  $\pm$  21.74 pg/ml) by 13-16 weeks postpartum. A total of 70 ovulations were monitored through steroids profile, which showed fertile oestrus as 55.72 %, silent ovulation 17.14%, anovulation 5.14%, unserved oestrus 5.71%, and early embryonic death 15.71%, which also included the long luteal phase. The non-conceived animals had a combined pattern of delayed rise and low level of progesterone during luteal phase indicative of insufficient luteal function than that of conceived animals, which had normal progesterone profile. The fluctuations observed in 17ß estradiol levels during early postpartum period were non-specific and erratic. The rising level of 17ß estradiol was associated with folliculogenesis as evident by rectal palpation prior to first heat postpartum.

**KEY WORDS:** Buffalo, Plasma progesterone, Estradiol, Postpartum Ovarian function, Silent ovulation, Embryonic mortality

# INTRODUCTION

The economics of dairy cattle and buffaloes lie in ensuring proper and optimal reproductive rhythm of each individual female in the herd. Any deviation or prolongation in the breeding rhythm results in a progressive economic loss due to widening of dry period, and reduced numbers of calving and lactation during lifetime of the animal. Infertile cow/buffalo means a loss in milk production and economic profit. Efforts should therefore be made to enhance fertility in dairy animals by narrowing down their dry periods to the barest minimum range of 60 to 90 days. Progesterone profile has been used to diagnose ovarian dysfunctions in Egyptian buffaloes by Barkawi et al. (1986) and in Indian cows and buffaloes by Mahapatra et al. (1988). The fertility of buffaloes was closely related to the normal profile of progesterone (Sarvaiya and Pathak, 1991). The present communication is on monitoring of postpartum ovarian activity of buffaloes through clinical and hormonal assays.

### MATERIALS AND METHODS

Thirty-nine postpartum Surti buffaloes of University Farm, Navsari (Gujarat) were studied for the profile of hormone progesterone (P4) and 17ß estradiol (E2) as well as ovarian changes per rectum at weekly interval from calving to at least 91 days postpartum. The objective was to characterise ovarian function and associated peridata on reproductive performance. All buffaloes under study had normal calving and were Gynaeco-clinically screened to rule out genital abnormalities. The postpartum reproductive performance, ovarian events and progesterone profiles were defined objectively as per Barkawi et al. (1995) with little modification as under.

Silent oestrus was considered to have occurred when behavioural signs of standing oestrus were not observed, eventhough there were other signs of oestrus. In case of silent oestrus progesterone

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level reached >1.0 ng/ml and continued at that level for two consecutive samples at weekly interval. In case of ovulatory oestrus, the day of oestrus was considered as the day of ovulation provided oestrus was followed by an increase in progesterone level for at least 2 consecutive samples. Animal was considered in anoestrum when the progesterone level was < 1.0 ng/ml and continued at that level for at least 3 consecutive weeks. If progesterone concentration decreased after having previously been high for more than 20 days after a natural service or correctly-timed insemination, it was assumed that the embryo had been lost (Mc Leod and Williams, 1991). All the postpartum buffaloes were checked for oestrus daily with the help of bull parading in morning and evening hours. The animals detected in oestrus were bred by natural service or by artificial insemination and they were palpated per rectum for pregnancy diagnosis at 45 days post-breeding. The buffaloes were classified as sub-fertile. Blood samples collected at weekly interval from day of calving till 91 day postpartum were used to monitor steroid hormone P4 and E2 profiles through RIA. A total of 70 ovulations were monitored among 18 buffaloes showing ovarian activity over the 90-day postpartum period, and the data were interpreted.

### **RESULTS AND DISCUSSION**

The pooled mean biweekly plasma P4 and E2 concentrations found from calving to at least 91 days postpartum in 39 Surti buffaloes are shown in Table 1.

| Sr.<br>No. | Weeks<br>postpartum | No. of<br>Observations | Progesterone<br>(ng/ml) | Estradiol-17β<br>(pg/ml)   |
|------------|---------------------|------------------------|-------------------------|----------------------------|
| 1          | At Calving          | 28                     | $1.19\pm0.33~^{bcd}$    | $153.05 \pm 21.74^{\rm a}$ |
| 2          | 1-2 Weeks           | 99                     | $0.36\pm0.12^{d}$       | $41.10\pm 6.82^b$          |
| 3          | 3-4 Weeks           | 78                     | $0.49\pm0.15^{d}$       | $24.93 \pm 4.49^{c}$       |
| 4          | 5-6 weeks           | 104                    | $0.56\pm0.16^{cd}$      | $15.96 \pm 2.44^{cd}$      |
| 5          | 7-8 Weeks           | 87                     | $1.09\pm0.33^{bcd}$     | $13.72\pm2.29^{cd}$        |
| 6          | 9-10 Weeks          | 74                     | $2.13\pm0.49^{bc}$      | $9.00\pm2.15^{d}$          |
| 7          | 11-12 Weeks         | 57                     | $2.40 \pm 0.66^{b}$     | $11.57 \pm 2.60^{d}$       |
| 8          | 13-16 weeks         | 52                     | $4.39\pm0.67^a$         | $6.77 \pm 1.39^{\rm d}$    |

Table 1. Mean ( $\pm$  SE) biweekly hormonal concentrations from calving to at least 91 days postpartum in 39 Surti buffaloes

Means within the column with different superscripts differ significantly (P < 0.05).

The mean plasma progesterone and 17ß estradiol concentrations recorded on the day of calving (Table 1) declined significantly (P <0.01) by 1-2 wks postpartum and showed lowest level by 1-2 wks and 13-16 wks postpartum, respectively. Thereafter P4 profile again showed an increasing trend from 3-4 wks onwards reaching a significantly higher level by 13-16 wks postpartum. But the E2 levels continued to decline till the end of experimental period. The trend of abrupt decline of postpartum oestrogen and progesterone profiles compared well with the reports of Tegegne et al. (1993) and Tiwari et al. (1995) in cows and buffaloes. Jainudeen et al. (1981), however, recorded very low or negligible level of progesterone up to 60 to 90 days postpartum, particularly in anoestrus Swamp buffaloes. Moreover, suckling resulted into lower level of 17ß estradiol during early postpartum period (Bellin et al., 1984). The 17ß estradiol profile of individual animals in our study

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fluctuated erratically during early postpartum period.

Of the total 70 ovulations monitored through progesterone profile during the first 91 days postpartum, the frequency of fertile oestrus was 55.72%, silent ovulation 17.14%, anovulation 5.71%, unserved oestrus 5.71% and early embryonic death 15.71%.

Of the 17.14 per cent (n=12) postpartum buffaloes showing silent ovulation, two conceived by first insemination. Overall oestrous cycle length of these buffaloes was  $22.2 \pm 2.28$  days (n=10). Of them 40 per cent (n=4) buffaloes had expressed normal cycle length ( $21.25 \pm 0.25$  days), 30 per cent (n=3) had shorter cycle length of 14.66  $\pm$  1.20 days and 30 per cent (n=3) had longer cycle length of 31.0  $\pm$  3.0 days. Four buffaloes (5.71%) expressed first anovulatory oestrus after an average interval of 24.25  $\pm$  2.86 days and they all exhibited next ovulatory oestrus. The present finding of overall 38.56 per cent ovarian dysfunctions observed through plasma progesterone profile, was in close agreement with the reports of Barkawi et al. (1986) and Butterfield and Lishman (1990). Mahapatra et al. (1988) through P4 profile found the percentage of HF cows in anoestrus, silent oestrus and oestrus as 31.1, 17.8 and 22.9, respectively and the incidence of long luteal phase (which included embryonic mortality) was 13.4 per cent. Mc Leod and Williams (1991) through progesterone profile had identified anoestrus (6.7%), follicular cysts (6.7%), luteal cysts (8.0%) and pregnancy (1.6%) at the time of clinical examination of postpartum cows. Honparkhe et al. (2008) used plasma profile to differentiate true anestrous and subestrus cows and buffaloes and thereby to determine hormone therapy such as hydroxyl-progesterone or eCG.

Nearly 16 per cent (n=9) of embryonic death was presumed in the present study based on higher progesterone levels on day 21-post breeding and delayed return to oestrus after an interval of 31.09 ± 2.83 days. Our results indicated that the luteal activity of the first postpartum oestrous cycle was lower than the subsequent cycles. Smolders et al. (1996) also revealed that at the first oestrous cycle postpartum, only 28 per cent of cows had normal luteal phase of 12-17 days, and the remainders had shorter luteal phase. At the second cycle, 56% of cows had a normal luteal phase. Conception rate after first insemination was higher in cows with a short dioestrus period (10-25 days) than those with dioestrus lasting for 26-50 days. Progesterone is essential for establishment and maintenance of pregnancy in cattle and buffaloes. Early embryonic mortality has been stated to be the most responsible cause of making the animal to repeat for multiple cycles (Jainudeen et al., 1981). Most of the early embryonic mortality occurs during days 8 to 18 after AI and as many as 40 per cent of fertilized ova die (Sreenan and Diskin, 1985). The failure of the signal system between the embryo and the mother around day 16 after oestrus "the maternal recognition of pregnancy" has been an important cause of repeat breeding syndrome (Heap et al., 1988). In the present study, anoestrus buffaloes were diagnosed through progesterone profile. AN. 202 revealed basal progesterone profile throughout 91 days of sampling, and exhibited first heat on 120 day postpartum. Thereafter she remained as dead anoestrum till day 370. Similarly, AN. 501 also showed basal to non-detectable P4 profile between day 21 and 63 postpartum and conceived at oestrus expressed on 113 day postpartum. Whereas, AN 204 revealed abnormal hormonal profile as indicated by elevation of progesterone and 17ß estradiol without palpable CL and follicle or oestrus. Conception in this buffalo was delayed and it occurred on 148 day postpartum. Similarly, AN. 306 had also abnormal P4 profile between day 13 and 42 without ovulation and/or CL, and first estrus in this buffalo was prolonged (373 days postpartum). Basal progesterone profile was found in buffaloes having smooth and inactive ovaries upto 40 days postpartum. Yadav et al. (1995) also reported basal progesterone level in buffaloes with smooth and inactive ovaries.

In present study determination of P4 profile of Surti buffaloes could characterise their normal and abnormal ovarian activity. Thus, there is need for devising models for the early detection and treatment of abnormal ovarian cycles and to stimulate ovarian pattern conducive to high conception rates, using hormonal preparations to improve reproductive efficiency. Embryonic mortality cases

observed in the study had longer luteal phase and consequently the higher progesterone on day 21 of infertile oestrous cycle. Erb et al. (1976) found subnormal progesterone after ovulation in repeat breeder cows (43.75%) than those of fertile cows. From our findings, it appeared that the infertile group had delayed corpus luteum formation than that of fertile buffaloes. Further, the fertility of buffaloes under study was closely related with the normal profile of progesterone and their postbreeding levels were in close agreement with the previous report of many workers in cows (Kimura et al., 1987; Shelton et al., 1990) and in buffaloes (Sarvaiya and Pathak, 1991; Panchal et al., 1992).

Our findings indicate that overall reproductive efficiency is impaired due to deficiency of progesterone, which is essential for embryo survival. Further, non-conceived animals had a combined pattern of a delayed rise and low level of progesterone during luteal phase indicative of insufficient luteal function than those of the conceived animals, possibility due to diminished response to LH, which may contribute to embryo mortality in subfertile animals (Shelton et al., 1990). The erratic and non-specific changes observed in plasma estradiol concentrations may be related to the release of FSH and it's effect on follicular growth and atresia. Peters and Lamming (1984) noted that gonadotropin secretion stimulates follicular growth and the production of estradiol and perhaps inhibin. Such endocrine changes initiate ovarian cycles about 2 weeks after calving. Thus, the rising level of 17ß estradiol was associated with folliculogenesis as it was evident by per rectal palpation prior to first heat postpartum. In 7 fertile buffaloes, occurrence of follicular activity resulted into oestrus within couple of days (AN. 603, 605, 606, 610, 611, 612 and 404), whereas in 3 infertile buffaloes though the follicular activity was evident oestrus did not follow (AN. 502, 503 and 205).

From the endocrine profile, follicular development (activity) and manifestation of oestrus in fertile and infertile buffaloes, it is clear that in fertile buffaloes appearance of first and second follicular activity resulted into formation of mature Graafian follicle and ovulation resulting in early postpartum oestrus compared to infertile buffaloes, in which development and maturation of follicle was interrupted (AN. 502, 503 and 205) or else follicular activity was not evident (AN. 501, 202 and 204) either due to basal 17ß estradiol (E2) profile or its sharp fluctuations. The variation between fertile and infertile buffaloes could be due to the difference in their endocrine status. Suckling or the presence of a calf is believed to exercise its inhibitory effect by influencing oestrogen production by follicular cells and reduced the positive feedback effect on the hypothalamic-pituitary axis (Hanzen, 1986). Moreover, the anterior pituitary content of LH is replenished in suckled cows within about 2-3 weeks of calving. Therefore, resumption of ovarian activity is delayed until the frequency of LH pulses increases to the levels found during pro-oestrus.

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