SEASONAL VARIATIONS IN THE HYPOPHYSIAL RESPONSIVENESS TO GnRH IN CYCLING BUFFALO (*Bubalus bubalis*)

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Summary

The present study investigated the hypophysial responsiveness in terms of GnRH induced LH and FSH release in cycling buffalo during the tropical summer and winter climatic conditions (seasons). Peripheral plasma LH and FSH levels were measured at 1 hour before and 6 hours subsequent to the administration of GnRH (1 ug/kg body weight) or saline on Day 14 of oestrous cycle in 2 groups of buffalo (n = 6 each) during summer and winter seasons. Although GnRH induced LH peak concentrations did not differ during the two seasons, time to attain LH peak concentration was shorter ($p < 0.05$) and the area under LH peak was 39% higher ($p < 0.05$) during winter season in comparison to summer season. However, season had no effect on GnRH induced peak FSH concentration, time to attain peak FSH concentration and the area under FSH peak. Pretreatment basal LH and FSH levels did not differ during the two seasons. The present study suggests that the summer season adversely affects the GnRH stimulated release of LH in buffalo.
(**Key Words** : GnRH, LH, FSH, Seasons, Buffalo)

Introduction

Buffalo occupies a prominent place in the economy of Asian countries where about 96% of the world population of buffaloes is found. Reproductive efficiency of buffalo is hampered by seasonality in cyclicity (Roy et al., 1972). High environmental temperatures during the tropical summer season have been reported to suppress reproductive performance in this species (Gangwar, 1980). This lowered reproductive performance during summer season is marked by diminished hypophysial activity in terms of low basal FSH levels (Janakiraman et al., 1980). Heat stress under controlled climatic conditions has been known to reduce basal as well as preovulatory surge peak LH levels in cow also (Madan and Johnson, 1973). Although, in buffalo (Bubalus bubalis) basal LH levels during the oestrous cycle do not show seasonal variation (Kaker et al., 1980; Rao and Pandey, 1983), LH pulse frequency during the luteal phase and LH pulse frequency and amplitude during the follicular phase are lower during hot season in comparison to those during the cooler breeding season (Aboul-Ela and Barkawi, 1988). Although literature is available on the seasonal variations in basal hormonal levels for hypophysial (Kaker et al., 1980; Rao and Pandey, 1983; Razdan et al., 1982) as well as ovarian hormones (Rao and Pandey, 1982; 1983), information available on the effects of climatic variations on hypophysial responsiveness is very scanty in buffalo (Aboul-Ela et al., 1983). It is not clear whether the capability of the pituitary to release gonadotropins in response to GnRH is altered by climatic variations. The present study was, therefore, undertaken to compare the hypophysial responsiveness to exogenous GnRH treatment during the hot (summer) and cold (winter) tropical climates in buffalo.

Materials and Methods

Animals and treatments

Pubertal, cycling Murrah buffaloes, weighing between 450 and 550 kg, which had exhibited at least one normal oestrous cycle (21-25 days) previously were monitored for behavioral signs of oestrus by parading a vasectomized teaser bull every 8 hours. Twelve animals which showed signs of oestrus were selected and randomly divided into two groups. These animals were not inseminated at oestrus. Cyclicity was also confirmed subsequently by

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checking progesterone profiles in these animals. Group I (experimental) animals were administered synthetic GnRH decapeptide intravenously at a dose of 1 ug/kg body weight on Day 14 of the cycle while Group II (control) animals received sterile saline during the summer season (May to July, mean maximum temp. 39.5℃, mean minimum temp. 25.2℃, relative humidity 51-97%). GnRH/saline treatment was repeated in the same animals in an identical manner during the winter season (December to February, mean maximum temp. 20.3℃, mean minimum temp. 7.4℃, relative humidity 50-96%).

Blood sampling and hormone analysis

Blood samples were collected in heparinized tubes from left jugular vein of animals 1 hour before treatment, immediately before treatment and at 15 min intervals for 1 hour followed by 30 min intervals for 5 hours following GnRH/saline treatment. Plasma was separated within 1 hour of blood collection and stored at −20℃ till subsequent analysis for LH and FSH by RIA. Blood samples were also collected once daily from Day 1 (day 0 = day of oestrus) to Day 13 for estimation of progesterone levels for confirmation of cyclicity. LH and FSH were estimated by double antibody RIAs as described earlier (Palta and Madan, 1995). The sensitivity of RIAs for LH and FSH was 0.4 ng/ml and the intra- and interassay coefficients of variations were < 10%. Progesterone was estimated by a method detailed earlier (Prakash and Madan, 1986).

Statistical analysis

Time to peak gonadotropin concentration was the length of time from GnRH administration to the time of highest concentration of gonadotropin. The duration of the GnRH induced response curve was defined as the length of time for which the gonadotropin concentration remained 2 × SD above the mean pretreatment gonadotropin concentration. For determination of the area under the response curve, the GnRH induced response curve was plotted on arithmetic coordinate paper and the number of sq mm was counted. The correlations between peak gonadotropin concentrations and the areas under response curves were obtained as described earlier (Snedecor and Cochran, 1967). Since the two were highly correlated ($R = 0.889$, $p < 0.01$ for LH and $R = 0.946$, $p < 0.01$ for FSH), the area under response curve was employed for comparison of the differences in GnRH induced gonadotropin release between different seasons by paired 't' test as described by Snedecor and Cochran (1967).

Results and Discussion

GnRH administration on Day 14 of oestrous cycle produced a significant elevation ($p < 0.01$) in the plasma LH and FSH levels within 15 min in all the animals in both seasons (figure 1). The peak LH concentrations following GnRH administration were not different during the summer and winter seasons. However, the total LH released in response to GnRH was 39% higher ($p < 0.05$) during winter season (figure 2). Time to attain peak LH concentration was shorter ($p < 0.05$) during winter season (1.75 ± 0.17 h) in comparison to that during the summer season (2.08 ± 0.14 h). Peak FSH concentration and total FSH released in response to GnRH (figure 2) as well as time to attain peak FSH concentrations (2.75 ± 0.10 and

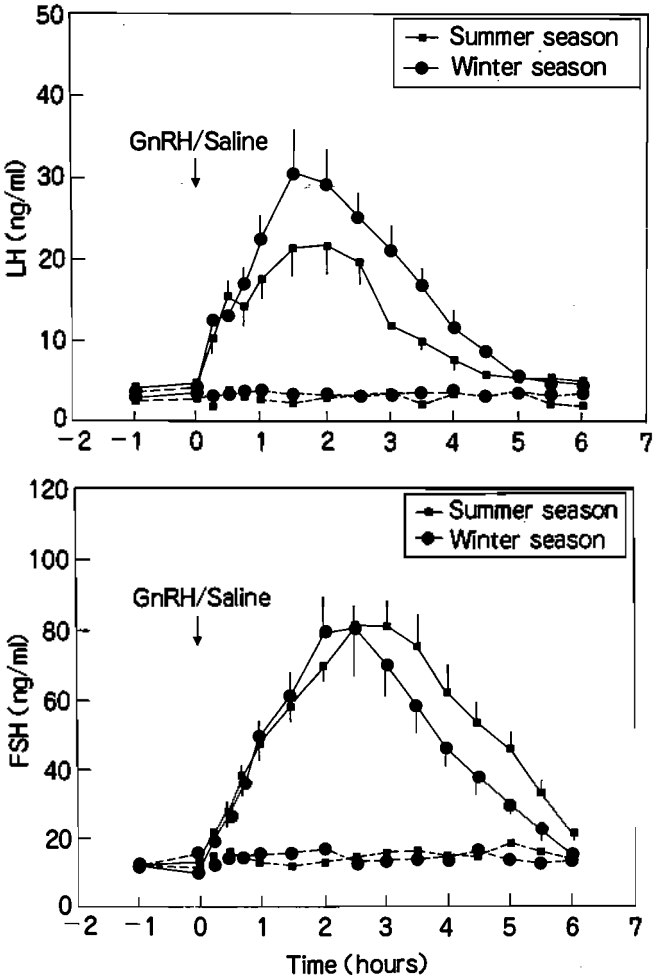

Figure 1. Release pattern of LH and FSH in response to administration of GnRH (1 ug/kg body weight) or saline on Day 14 of oestrous cycle during summer and winter seasons (Day 0 = day of oestrus). Values are Mean ± SEM (n = 6).

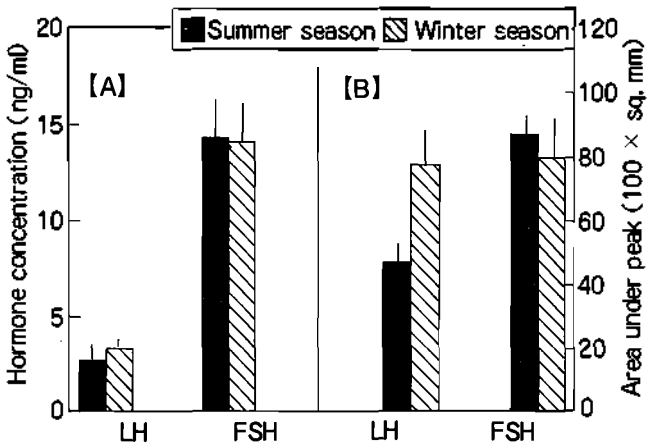

Figure 2. [A] Basal LH and FSH levels and [B] Area under the response curves following the administration of GnRH (1 ug/kg body weight) on Day 14 of oestrous cycle during summer and winter seasons (Day 0 = day of oestrus). Values are Mean ± SEM (n = 6).

2.50 ± 0.11 h during the summer and winter seasons, respectively) were not significantly different during the two seasons.

Administration of saline did not produce any elevation in the peripheral plasma LH and FSH levels. A comparison of the pooled pretreatment and post saline treatment LH and FSH levels during the two seasons (figure 2) did not give any significant difference. The plasma progesterone profile of the animals from Day 1 to Day 13 (data not shown) presented normal progesterone levels indicating normal cyclicity in all the animals. The mean plasma progesterone levels ranged from 4.31 ± 0.48 to 5.92 ± 0.96 and from 3.72 ± 0.49 to 5.08 ± 0.50 ng/ml between Days 10 and 13 during the summer and winter seasons, respectively.

Pretreatment basal levels of LH and FSH obtained in the present study are similar to those reported earlier (Aboul-Ela et al., 1983; Batra and Pandey, 1983). Time to attain peak LH levels following GnRH administration is also similar to earlier reports in cow (Kaltenbach et al., 1974) and buffalo (Aboul-Ela et al., 1983). Day 14 of oestrous cycle was chosen for GnRH administration since the ovarian steroid hormones, which affect the hypophysial responsiveness through feedback do not exhibit major fluctuations around this time of oestrous cycle in buffalo (Bachlaus et al., 1979). Moreover, the pituitary LH content which is highly correlated with the magnitude of GnRH induced LH release (Moss et al., 1980; Crowder et al., 1982) has been reported to undergo a drastic reduction following the preovulatory LH surge and is restored only by Day 14 cows (Schoenemann et al., 1985). Administration of GnRH at a dose of 1 ug/kg body weight was able to induce elevations in LH and FSH levels with peak concentrations comparable to those found during natural preovulatory surge in buffalo (Batra and Pandey, 1983).

Although the GnRH induced LH peak concentrations were not different during the two season, the LH peak was attained in a shorter time ($p < 0.05$) following GnRH treatment during winter season indicating higher pituitary responsiveness during the colder breeding season. This is in agreement with the results reported in an earlier study in which 150 ug GnRH was administered 8 days after ovulation in buffaloes (Aboul-Ela et al., 1983). The results of the present study, however, differ from this report since a significantly higher area under GnRH induced LH peak was obtained during the winter season in the present study, indicating a higher secretory capacity of the pituitary in response to GnRH during the winter season. To our knowledge this is the first report in which GnRH induced FSH release was compared during the summer and winter seasons in buffalo. No seasonal variation was observed in the hypophysial responsiveness in terms of GnRH-induced FSH release.

High environmental temperatures have been reported to result in a reduction in the basal LH levels and the magnitude of preovulatory LH surge in cow (Madan and Johnson, 1973; Miller and Alliston, 1974). LH levels have been reported to be higher on the day of oestrus in cooler season than during the hotter months, with no effect on other days of cycle (Rao and Pandey, 1983). Since the occurrence of preovulatory surge like rise in gonadotropin levels is dependent upon a prooestrus rise in estradiol (Kesner et al., 1981), lower estradiol levels on the day before oestrus in buffalo in hotter months, in comparison to cooler months (Rao and Pandey, 1983) may be contributing towards lack of occurrence of preovulatory LH surge during the hot season in buffalo (Kaker et al., 1980). The present study, however, suggests that the pituitary responsiveness per se may be lower, at least in terms of GnRH induced LH release, during the summer season. This is in agreement with earlier reports in which LH pulse frequency was found to be higher during luteal phase in winter as compared to summer months in Egyptian buffalo (Aboul-Ela and Barkawi, 1988).

In conclusion the results of the present study show that the hypophysial responsiveness is adversely affected by tropical hot season. This may be a factor contributing to lower reproductive performance of buffalo during the summer season.

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