The reproductive pattern and efficiency of female buffaloes

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Abstract

Buffaloes play a prominent role in rural livestock production, particularly in Asia. Reproductive efficiency is the primary factor affecting productivity and is hampered in female buffalo by (i) inherent late maturity, (ii) poor estrus expression in summer, (iii) distinct seasonal reproductive patterns, and (iv) prolonged intercalving intervals. Ovarian function is central to these issues; hence, the focal point of this review is ovarian function in Bubalus bubalis, particularly, in relation to seasonal changes. Ovarian anatomy, follicular and luteal development development, and hormonal profiles during the estrous cycle are discussed. Review of the literature revealed a paucity of critically derived information on follicular and ovulatory patterns in buffalo, particularly, in relation to seasonal estrus/birthing. Efforts may be directed at understanding the process (recruitment, development, atresia) and temporal pattern (follicle selection, dominance, subordinate follicle suppression, follicle numbers, and, preovulatory changes) of follicular dynamics using techniques which permit serial assessment of changes occurring over time. Emphasis may be directed towards investigating follicular “waves” as a functional unit, rather than the estrous cycle, in the context of whole animal endocrinology. The data obtained from such basic studies may then be used to develop and test models for enhancing reproductive efficiency. © 2000 Elsevier Science B.V. All rights reserved.

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1. Introduction

Among the African (*Syncerus caffer*) and Indian (*Bubalus* spp.) groups of buffaloes, only the Indian wild buffalo (*Bubalus arni*) has been domesticated and has been named *Bubalus bubalis*. Subspecies of *B. bubalis* are referred to as river buffalo and swamp buffalo. While river buffaloes are commonly found in the Indian subcontinent, Mediterranean region of Europe, South America and the Caribbean, swamp buffaloes are commonly seen in Southeast Asia and China. The river buffalo is a triple purpose species contributing milk, draft-work and meat, while the swamp buffalo is considered a dual-purpose species — draft and meat. Despite river and swamp buffalo possessing a different number of chromosomes (2n = 50 and 48, respectively), they are able to interbreed and produce fertile hybrid progeny (Xiao, 1988).

More than 50% of the estimated world population of 148 million buffaloes is in India (Agarwal and Tomar, 1998). Murrah, Nili-Ravi, Surti, Mehsana, Nagpuri and Bhadawari are the most common breeds. Buffaloes contribute 10% of the world’s total milk production, virtually all of which (> 99%) is produced in developing countries (Shah, 1988). Regarding meat production, an estimated 1.6 million metric tons of buffalo meat is produced annually (Agarwal and Tomar, 1998). As a beast of burden, the buffalo is considered the tractor of most of Southeast Asia, providing 20% to 30% of farm power in China, Thailand, Indonesia, Malaysia and the Philippines.

It is clear that buffaloes play a prominent role in rural livestock production, particularly in Asia, and factors affecting productivity are of paramount importance to agricultural economics in this region of the world. Reproductive efficiency is the primary factor affecting productivity and is hampered in female buffalo by (i) inherent late maturity, (ii) poor estrus expression in summer, (iii) distinct seasonal reproductive patterns, and (iv) prolonged intercalving intervals (Madan, 1988; Madan and Raina, 1984). Hence, the focal point of this review is the form and function of the ovaries of *B. bubalis*, especially as they are affected by reproductive status and season.

2. Ovarian anatomy

The buffalo ovary is elongated and considerably smaller than that of cattle. The average dimensions of the buffalo ovary vary between $22-26 \times 11-18 \times 11-14$ mm$^3$ (Fadde et al., 1974) with a maximum and minimum weight of 6.1 and 2.9g, respectively (El-Wishy et al., 1971). The corpus luteum (CL) of the buffalo is often deeply embedded in ovarian stroma and is generally smaller than that of cattle (maximum weight 2.3 g, maximum diameter 15 mm; Roy and Mullick, 1964). Examination of excised ovaries (Danell, 1987) revealed that the left–right distribution of ovulation (as indicated by the presence of a CL) was similar; the CL was detected in left and right ovaries in 51.2% and 48.8% of observations, respectively.
3. Estrous cycle, estrus and ovulation

Although not compared critically, there appear to be considerable differences in reproductive traits among different breeds of buffaloes. The average length of the estrous cycle has been reported to be 20–22 days for river buffaloes; however, great variations have been observed under rural conditions (Agarwal and Purbey, 1983; Madan, 1988). The average duration of estrus appears to be slightly longer in river buffalo (23.8 ± 6.2 h; Danell, 1987) than in swamp buffalo (19.9 ± 4.4 h; Kanai and Shimizu, 1983; Shimizu, 1987). A seasonal variation was reported in one study (Janakiraman, 1978) wherein the duration of estrus was estimated to be 14, 18, and 8–10 h in the monsoon, winter, and summer seasons, respectively, in river buffaloes.

Salient signs of estrus in river buffalo (Surti and Nili-Ravi breeds) are decreased milk yield, bellowing, vulvar swelling and mucus discharge (Kamizi, 1983; Rao and Kodagali, 1983; Danell et al., 1984). Swamp buffaloes exhibit a higher frequency of, and submission to, vulvar sniffing during estrus (McCool et al., 1989). The interval from the end of estrus to ovulation has been estimated to be 11 h in Indian buffaloes (nondescript breeds, Luktuke and Ahuja, 1961), 12–24 h in Surti buffaloes (Danell, 1987), 14.8 ± 0.4 h in Nagpuri buffaloes (Raut and Kadga, 1988) and 13.9 h in swamp buffaloes (Kanai and Shimizu, 1986).

4. Follicle development

Follicular development has not been studied in detail in buffalo. Left (49.3%) and right (50.7%) ovaries have a similar number of primordial follicles (Danell, 1987). The population of primordial follicles was estimated to be 19,000 (Samad and Nasseri, 1979). In a more critical study (Danell, 1987), Surti buffaloes possessed 10,132 (anestrus animals) to 12,636 (cycling animals) primordial follicles. In cattle, the estimated mean number of primordial follicles decreased from 133,000 at birth to less than 3000 at 20 years of age (Erickson, 1966). Although the number of primordial follicles appears to be considerably lower in buffaloes than in cattle, the effect of age on the population has apparently not be evaluated in buffaloes. In addition, buffalo ovaries contain only about 20% of the number of antral follicles found in cattle ovaries (48 versus 233; Ty et al., 1989). Cycling versus noncycling Surti buffalo heifer ovaries possessed 46.3 versus 67.8 follicles > 1 mm, and 16.8 versus 23.6 follicles > 2 mm, respectively (Danell, 1987). The average number of follicles visible on surface of each ovary was 5.2 ± 1.0 (Kumar et al., 1997). In Surti buffaloes, the average proportion of antral follicles that appeared to be atretic, histologically, was 71% (Danell, 1987), compared to 82% (71.4% during follicular phase and 94.4% during luteal phase) in swamp buffaloes (Ocampo et al., 1994). Evaluation of the number of follicles > 2 mm in ovaries excised at three different time periods of the estrous cycle revealed a greater number between Days 1–8 and 9–11 than during Days 12–21 (Danell, 1987). Large (7–8 mm) follicles could be palpated during Days 9 to 13 of the estrous cycle in 65% of post-pubertal buffalo heifers (Singh et al., 1984). Phase-related differences in the number and health of antral follicles reported in these studies provide rationale for the
hypothesis that follicular development occurs in a wave-like pattern in buffaloes, similar to cattle. This hypothesis remains to be tested.

5. Hormonal profiles during estrous cycle

Reproductive endocrinology of the buffalo has received considerable attention during the last two decades. Although steroid hormone concentrations have been measured with reasonable accuracy, measurement of gonadatropins has been hindered by a lack of purified hormone preparations for antibody production and radio-iodination for development of homologous LH and FSH immunoassays. Moreover, lack of uniform standards among different laboratories poses another difficulty. Hence, the results of LH and FSH profiles should be interpreted with caution at this stage.

The basic pattern of changes in hormone profiles of buffaloes during the estrous cycle closely resembles that of cattle. The concentration of blood progesterone is at its nadir (0.1–0.3 ng/ml) during estrus and remains close to 1 ng/ml for the next 3–4 days (Arora and Pandey, 1982). The first significant increase in progesterone concentration occurs about 7 days after estrus (Ahmed et al., 1977). Peak progesterone values of 4.0–5.1 ng/ml (Bachlaus et al., 1979; Arora and Pandey, 1982; Takkar et al., 1983) have been recorded about 15 days after estrus. Circulating estradiol concentrations remain low during the luteal phase with minor fluctuations (10–20 pg/ml) around Days 4 and 10 of the estrous cycle in river buffalo (Batra and Pandey, 1982; Samad et al., 1988), but not in swamp buffalo (Kanai and Shimizu, 1984). Peak concentrations of estradiol (30–35 pg/ml) were detected on the day of estrus or one day before (Batra and Pandey, 1982), followed by a decline to 5–10 pg/ml within two days. This pattern is indicative of enhanced estradiol production by the preovulatory follicle during proestrus. Circulating concentrations of LH reached a peak (20–35 ng/ml) at the onset of estrus followed by a sharp decrease within a day; LH remained low (1–3 ng/ml) during the luteal phase (Batra and Pandey, 1982; Kanai and Shimizu, 1984). The duration of the LH surge has been estimated to be 7–12 h (Batra and Pandey, 1982; Kanai and Shimizu, 1986). Peak LH concentrations were estimated to occur about 14.8 h after the peak in estradiol concentration (Batra and Pandey, 1982). Higher FSH levels (57–65 ng/ml) have been observed during the beginning of estrous cycle than during the luteal phase (10–17 ng/ml; Razdan et al., 1982) but such a difference was not observed by others (Janakiraman et al., 1980). For changes in hormone profiles during different seasons, the reader is referred to the section under endocrine alterations during summer.

6. Seasonality

6.1. Seasonal behavior

There is a complex dependency of bovine reproduction on soil, plant and climatic factors (Predojevic et al., 1988), particularly in tropical and subtropical parts of the world. Although buffaloes are polyestrous, they exhibit a distinct seasonal variation in
display of estrus, conception rate, and calving rate (Ahmad et al., 1980; Madan, 1988; Singh and Nanda, 1993; Singh and Singh, 1985; Singh et al., 1989; Parsha et al., 1986; Shah, 1988; Tailor et al., 1990). Buffaloes and Zebu cows located at the same farms in Pakistan (i.e., managed under similar fodder and climatic conditions) showed a different pattern of breeding. The breeding frequency in buffaloes was highest during the winter, decreased in autumn and spring, and was lowest in the summer (Shah, 1988). The Zebu cattle in this study exhibited the opposite seasonal variation; breeding frequency was highest during the summer. Results of other studies done in India are consistent with the Pakistan study; 64% to 75% of buffaloes exhibited estrus during September to January (Singh and Nanda, 1993; Singh and Singh, 1984; Tailor et al., 1990).

The interval to first postpartum estrus is also significantly affected by the season of calving (Basu, 1962; Reddy et al., 1986; Madan, 1988; Singh, 1993; Singh and Nanda, 1993). Buffaloes calving in late winter and early summer have lower reproductive efficiency compared to those calving during other periods (Siddappa and Patil, 1979; Madan and Raina, 1984; Singh and Nanda, 1993). The resumption of ovarian activity after calving was significantly delayed in buffaloes that calved from February to May (116–148 days) compared to the rest of the year (38–64 days; Singh and Nanda, 1993). Furthermore, conception rates were lower between February and August (Madan, 1988), and number of services per conception was higher in summer calvers than those calving in other seasons.

In rural Punjab (lat 29° to 32°N, long 75° to 76°E; temperature extremes: 0°–46°C; day length: 10–14 h) most buffaloes (58%) are pregnant during the heat of the summer. However, among the remaining nonpregnant buffaloes, the majority (77%) become anestrus during the hot summer months (Singh et al., 1989). Seasonal suppression (summer versus winter) of reproductive function has been documented by a shorter duration of estrus (8–10 versus 18 h; Janakiraman, 1978), apparent prolongation of the interval from estrus to ovulation (15.8 ± 0.4 versus 14.9 ± 0.4 h in Nagpuri buffaloes; Raut and Kadu, 1988), and fewer ovulatory cycles (71% versus 92%; Janakiraman, 1978) during the summer months. Unobserved (silent) estrus and abbreviated duration of estrus are common in swamp buffalo during the summer (Kanai and Shimizu, 1983). In one study (Singh et al., 1985), the incidence of true anestrus (defined as the absence of large follicles and CL in the ovaries) was 78% in July and 14% in November. Poor libido and semen quality of bulls may also contribute to lower fertility during the summer.

6.2. Endocrine alterations during summer

There is no consensus among studies on the effects of season on the function of the corpus luteum. Some authors suggest that luteal function is adversely affected during the summer season; maximum circulating concentrations of progesterone were 2.0 ± 1.2 ng/ml during the summer versus 3.1 ± 0.2 ng/ml during the winter (Rao and Pandey, 1982). Others report that higher concentrations of progesterone were recorded during summer than during fall and winter (Shafie et al., 1982; Takkar et al., 1983; Singh and Chaudhary, 1992). Still others detected similar progesterone concentrations during the summer and winter months (1.3 ± 0.4 and 1.3 ± 0.2 ng/ml, respectively), but found that concentrations were lower in the fall (0.7 ± 0.2 ng/ml; Kumar and Rattan, 1992).
Lower circulating concentrations of FSH (measured during estrus and during the luteal phase) were detected during summer than winter months (Janakiraman et al., 1980; Razdan et al., 1982). Lower LH values during summer than winter have also been reported by some (Rao and Pandey, 1983), but not by others (Sheth et al., 1978). No optimum LH peak was detected in anestrus versus cycling buffaloes during summer (Razdan and Kaker, 1980; Razdan et al., 1984). The amplitude and frequency of LH secretion during the follicular phase were lower during the summer than during the winter (Aboul-Ela and Barkawi, 1988). Furthermore, a lower LH pulse frequency during the luteal phase was observed in the summer (2.1 h) than winter (3.2 h).

6.3. Factors affecting seasonal behavior

It is not clear to what extent the seasonal breeding pattern is a genetic characteristic of buffalo or a result of environmental factors. However, evidence suggests a strong influence of biometeorological factors on the endocrine system of buffaloes (i.e., day length, ambient temperature, relative humidity and rain fall; Shah, 1988). The seasonal pattern may also be attributed to be a consequence of, or adjustment to, meager availability of green fodder during the summer months. However, that this is not the sole factor affecting seasonality is borne out by the results of the Pakistan study in which buffalo displayed a different breeding pattern than cattle kept under similar environmental conditions (Shah, 1988). Greater breeding activity was detected during autumn in buffaloes than in cattle. On the hot, humid plains of Papua New Guinea, most swamp buffaloes continued to come into heat despite losing weight due to poor nutrition, while cows kept under the same conditions became anestrus (Report, 1981).

Further, reduced sexual activity in the buffalo coincides with both an increase in ambient temperature (Dessoukey and Juma, 1973; Singh et al., 1989) and increasing day length (Razdan and Kaker, 1980), but studies done to date have not been designed to distinguish between the effects of photoperiod and other environmental factors. The incidence of true anestrus was significantly correlated with mean maximum and minimum air temperature ($r = 0.8$ and $0.9$, respectively) and with mean relative humidity ($r = -0.7$; Singh et al., 1985). The proportion of buffaloes exhibiting estrus during the period of short day length was significantly greater than during the period of long day length (74% versus 26%, respectively; Tailor et al., 1990).

In an elaborate study examining the interval from parturition to first estrus (Singh, 1993; Singh and Nanda, 1993), data collected during two successive years were compared to distinguish between the effects of photoperiod and other environmental factors (i.e., successive years had the same photoperiod changes, but different rainfall and ambient temperature patterns). Only 23% of all detected estrus were recorded in the months with increasing temperature, suggesting some suppressive effects of higher temperatures (Fig. 1). However, the highest temperature was recorded in June whereas the lowest incidence of estrus was in July. Moreover, 15% of estrus observations occurred in September versus only 5% in March despite a similar temperature during these 2 months. Relative humidity was maximum in January (74%) and minimum in May (46%), and had no distinct relationship with ovarian activity. From February
through June, the relative humidity was higher and the temperature was lower ($P < 0.05$) in 1986 than in 1985 due to early rainfall; however, no difference in the incidence of estrus was detected between years. Day length was also negatively correlated ($r = -0.7, P < 0.01$) with the interval from parturition to first estrus. These patterns indicate that decreasing day length may be a stronger determinant of the onset of postpartum ovarian activity, whereas ambient temperature and relative humidity may have relatively lesser influence.

7. Estrus synchronization

Covert or silent estrus constitutes the single largest factor responsible for poor reproductive efficiency in buffalo. Aside from seasonal factors affecting the display of estrus, the most important contributing factor is an inability to detect estrus. The traditional farmer possesses only one to four buffaloes and usually no bull; hence, there is little opportunity for behavioral interaction among estrous animals. Artificial control of the estrous cycle has provided an efficient means of increasing the reproductive capacity of buffalo by obviating the need for frequent visual inspections. Progesterone- or norgestomet-containing devices (injections, PRID, pessaries, CIDR, ear implants) along with PMSG, estradiol and/or prostaglandin ($\text{PGF}_2\alpha$) have been used successfully to improve synchrony of estrus and conception in buffalo (Rao and Rao, 1979, 1983; Rao et al., 1985; Singh et al., 1983; Saini et al., 1986; Chohan et al., 1995). Results of single- or double-injection regimes of $\text{PGF}_2\alpha$ in buffalo are comparable to those
obtained in cattle (Kamonpatana et al., 1979; Pathiraja et al., 1979; Pant and Singh, 1980; Kamonpatana et al., 1987; Rao and Venkatramaiah, 1989; Dhaliwal and Sharma, 1990; Dhaliwal et al., 1987; Sahasrabudhe and Pandit, 1997). The endocrine changes following PGF$_2$$_{\alpha}$ – induced luteolysis appear to be similar to those occurring at natural estrus (Kamonpatana et al., 1979). In addition, use of one fifth the conventional dose of PGF$_2$$_{\alpha}$ by intravaginal submucosal injection was equally as effective in inducing estrus as conventional treatment (Rao and Rao, 1988; Rao and Venkatramaiah, 1989; Subramaniam et al., 1989). However, results of others (Dhaliwal et al., 1987) suggested that the intravaginal route was not as effective for inducing synchrony as conventional treatment for fixed-time insemination. Better synchrony and conception rates have been documented after synchronization during the breeding season (winter) than the nonbreeding season (summer; Rao and Rao, 1983), but a remarkably high percentage (88%) of subestrous buffaloes expressed standing estrus after PGF$_2$$_{\alpha}$ treatment during the hot summer months (Sahasrabudhe and Pandit, 1997).

8. Multiple ovulations in response to exogenous gonadotropins

The ovarian response of buffaloes to superstimulatory treatment has been less than one third of that reported in cattle. By using Folltropin®, SuperOV®, FSH-P® or PMSG, only 50% to 55% of buffaloes respond (i.e., > 2 ovulations), and of those that respond, only two to four ovulations are induced, producing only one to two transferable embryos (Parnpai et al., 1985; Karaivanov, 1986; Vlakhov et al., 1986; Alexiev et al., 1988; Sharifuddin and Jainudeen, 1988; Singh et al., 1988; Singla and Madan, 1990; Misra et al., 1991; Kasiraj et al., 1992; Agarwal et al., 1996). Although experiments on multiple ovulation in buffalo were initiated over three decades ago, systematic examination of the ovarian response to gonadotropin treatment relative to intrinsic follicular development has not been done. The world’s first buffalo calf through embryo transfer was born in the USA in 1983 (Drost et al., 1983).

Various schedules of gonadotropin treatment, such as PMSG (Parnpai et al., 1985; Vlakhov et al., 1986; Karaivanov, 1986; Alexiev et al., 1988; Nanda and Bhat, 1988; Misra, 1993), tapering doses of FSH (Singh et al., 1988; Singla and Madan, 1990; Misra et al., 1991), and single injection of FSH (Kasiraj et al., 1992) have been adopted from superovulatory protocols in cattle. Higher ovulation rates have been reported when gonadotropin treatment was initiated during the mid-luteal phase (Day 12) than the early (Day 3–6) or late luteal (Day 13–15) phase (Mehmood et al., 1988; Karaivanov et al., 1990). Higher ovulations were achieved when treatments were initiated in the absence of a dominant follicle than the presence (Taneja et al., 1995).

9. Conclusion

In conclusion, buffaloes display a distinct seasonal variation in reproductive pattern. More than 50% of animals bear the summer stress during their pregnancy, however, in the remaining nonpregnant animals, summer season leads to silent heat and/or anestrus.
There is a strong need to identify the factors responsible so as to devise effective breeding strategies. In addition, current knowledge of basic and seasonal patterns of follicle development in the buffalo is insufficient. Efforts may be directed at understanding the process (recruitment, development, atresia) and temporal pattern (follicle selection, dominance, subordinate follicle suppression, follicle numbers, and, preovulatory changes) of follicular dynamics using techniques which permit serial assessment of changes occurring over time. Emphasis may be directed towards investigating follicular “waves” as a functional unit, rather than the estrous cycle, in the context of whole animal endocrinology. The data obtained from such basic studies may then be used to develop and test models for enhancing reproductive efficiency. It would be particularly interesting to determine responses to treatment in relation to the intrinsic pattern of follicle development. The existence of a wave-pattern of follicular dynamics in buffaloes and temporal relationships among follicles may be expected to impact directly on the design of, and response to, synchronization and superstimulation protocols. In addition there is strong need for a concerted and organized effort to produce purified native and recombinant gonadotropins for developing buffalo-specific immunoassays and for therapeutic/superovulatory treatments.

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