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Follicular Dynamics and Oestrous Detection in Thai Postpartum Swamp Buffaloes (*Bubalus bubalis*)

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Contents

This study characterized follicular activity and oestrous behaviour from 5 to 9 days post-calving up to the 4th ovulation postpartum (pp) in 16 multiparous (range 2–7 parities) Thai swamp buffalo cows (*Bubalus bubalis*), aged 4–12 years and weighing from 432 to 676 kg. Ovarian follicular activity was examined by transrectal ultrasonography (TUS) every morning. Oestrous detection was performed twice daily by direct personal observation of behaviour and for presence of clear cervical mucus discharge and indirectly by video camera recording during 21 h/day. A follicular wave-like pattern was present before the 1st ovulation leading to short oestrous cycles. Growth rates and maximum diameters of the ovulatory follicles did not differ between the 1st and 4th ovulations. However, growth rate for non-ovulatory dominant follicles (DF) before the 1st ovulation was lower than for the ovulatory follicle ($p < 0.05$). In addition, the diameter of all ovulatory follicles (14.3 ± 0.46 mm, n = 39) was significantly larger ($p < 0.01$) than those of the preceding last but one non-ovulatory DF (10.8 ± 0.20 mm, n = 5), but similar to the last preceding non-ovulatory DF diameter (12.92 ± 0.96 mm, n = 14). Short oestrous cycles were most common between the 1st and 2nd ovulations (93.75%, 15/16 cows, 10.2 ± 0.38 days) decreasing in prevalence thereafter (50%, 3/6 buffaloes, 12.0 ± 1.53 days). Oestrous signs were relatively vague around the 1st ovulation pp to become more easily detectable thereafter. This study suggests that properly fed swamp buffaloes could be mated successfully within 2 months pp, at their 2nd spontaneous ovulation, provided oestrous detection is at least performed daily at 06:00–08:00 hour.

Introduction

Postpartum anestrus and poor oestrous detection are causes of subfertility among swamp buffalo cows reared under tropical conditions, as illustrated by the presence of > 18 months days open (Chantarakhana et al. 1981). This suboptimal reproductive performance is because of postpartum anestrus (Zicarelli 2001) or reduced ovarian activity (Jainudeen et al. 1983) in suckled swamp buffaloes. For amelioration of fertility, knowledge of ovarian cyclicity with respect to follicular development and dynamics is essential (Ali et al. 2003). Although much data on follicular waves in riverine-type buffaloes have been published, the physiological control of recruitment, selection, growth, dominance and atresia of ovarian follicles in postpartum swamp buffaloes remains to be elucidated. Only few studies on follicular development in swamp buffaloes after parturition have been documented (Jainudeen et al. 1983; Ali et al. 2003; Presicce et al. 2003) and indicated impaired or lack of follicular development in the postpartum period. This could be because of lack of adequate stimulation by gonadotrophic hormones as significant variations in LH levels between Days 3 and 90 after calving were not observed in Murrah buffaloes (Arya and Madan 2001). In addition, induction of oestrus including ovulation through hormonal treatment in the buffalo has been relatively unsuccessful (Manik et al. 1998; Rastegarnia et al. 2004; Presicce et al. 2005). A better understanding of follicular wave dynamics could facilitate the development of a methodology for influencing ovarian function and oestrus in both cyclic and non-cyclic animals. The use of real-time ultrasound imaging has greatly improved our knowledge of follicular dynamics in cattle and buffaloes (Sirosis and Fortune 1988; Taneja et al. 1996; Henao et al. 2000; Presicce et al. 2003; Vassena et al. 2003). Moreover such an approach could shed light upon the conflicting data published by Jainudeen et al. (1983) who observed that postpartum follicular development was absent, as diagnosed by rectal palpation but where a parallel endocrine evaluation indicated functional luteal tissue.

Oestrous detection is one of the most important components for any effective breeding programme (Pelissier 1982). Many methods for oestrous detection have been developed in cattle management such as chalking the tail-head, chin ball markers and heat mount detectors (Dinsmore and Cattell 1993; Xu et al. 1998). Unfortunately, these methods are not useful in the swamp buffalo owing to its behaviour and habitat. In Thailand, most buffaloes are free-ranging during daytime and kept in a pen at night. As a result, oestrous detection methods suitable for swamp buffalo behaviour and adapted for these specific ways of husbandry are needed.

The objective of our study was to characterize postpartum follicular dynamics and oestrous symptoms in swamp buffaloes.

Materials and Methods

Animals

Sixteen multiparous postpartum swamp buffaloes (*Bubalus bubalis*), aged 4–12 years, parity range 2–7, weighing 432–676 kg and being between 5 and 9 days postpartum (pp), were included in this study. The animals belonged to the Surin Research and Breeding Buffalo Center (Surin Province, Thailand). After calving, all buffaloes and their calves were grouped with one adult, sexually active and proven fertile buffalo bull for the detection of oestrus and natural breeding. They were fed daily with 3–5 kg/day/head of commercial concentrate, and dry grass and water were offered *ad libitum*. The animals were allowed to roam freely in grass fields during daytime. Body weight was measured weekly. The experiment was carried out during the cool season, from November 2004 to February 2005.

Transrectal ultrasonographic examination

Ovarian follicular dynamics were transrectally examined daily during the morning (09:00–12:00 hour) with a real-time B-mode linear array ultrasound scanner (Aloka SSD-500, Tokyo, Japan), equipped with a 7.5 MHz transrectal transducer. Examination started on day 5–9 pp until the fourth ovulation, pregnancy or until day 100 pp. Follicles were measured using the in-built scale provided with the ultrasound unit, and their growth and regression individually monitored as described by Sakaguchi et al. (2004). When follicles reached a diameter of ≥5 mm (day of wave emergence), they were individually followed until a dominant follicle (DF, the largest follicle that grew to a diameter of ≥10 mm and was at least 2 mm larger than other follicles) could be identified. Ovulation (i.e. the disappearance of a DF – the ovulatory follicle (OF) – followed by the development of a corpus luteum in the same position) was also recorded to define the inter-oestrous interval (i.e. the interval between two consecutive ovulations). An oestrous cycle was considered normal when ranging between 19 and 22 days in duration. Deviations below or above this interval were defined as short and long cycles, respectively. The growth rate of the follicle, the length of the growth phase, the maximum diameter, the length of the stationary phase, the rate of regression and the length of the regression phase were determined from TUS recordings.

Oestrous detection

Oestrous symptoms, including attempts to mount, being mounted, being smelled by other cows or the bull, were scored individually on a daily basis using a mature teaser. Oestrous detection was performed between 07:00 and 08:00 hour and between 17:00 and 18:00 hour. A video camera continuous recording was made 21 h per day (00:01–09:00 hour and 12:01–24:00 hour), and presence of clear cervical mucus discharge was noted.

Progesterone assay

Blood was collected by jugular venipuncture three times a week from a week postpartum until the occurrence of the 4th spontaneous ovulation. Blood serum was harvested and stored at −20°C until progesterone concentrations were determined using a solid-phase enzymeimmunoassay (EIA) (Pregnane EIA: CL425, Brown et al. 2005).

Meteorological data

Data (ambient temperature recording every three hours, from November 2004 to February 2005) were obtained from a nearby Meteorological station in the Surin province.

Statistics

The SPSS statistical software (version 11.5, IBM Company, Chicago, IL, USA) was used for the analyses of data, which were subjected to ANOVA Duncan's multiple comparison test (a $p < 0.05$ was considered threshold for significance), a K-W multiple comparison *Z*-value test (a $p < 0.01$ was considered significant) or paired-samples test (a $p < 0.01$ was considered significant).

Results

In this study, oestrous symptoms and mating were observed in almost all animals. Ten buffaloes were pregnant after the 2nd ovulation, while five buffaloes were pregnant after the 3rd ovulation. One buffalo got pregnant on day 110 postpartum following the 4th ovulation.

During the experiment, the mean body weight of the buffaloes significantly decreased (532.5 ± 17.87 vs 509.1 ± 17.81 kg, n = 16, $p < 0.001$). The number of non-ovulatory DF observed before ovulation varied between zero and three. Follicular dynamics in most of the buffaloes was characterized by the emergence of waves of follicles (≥5 mm) with only one follicle (the 1st DF) developing up to a larger size than the others. After regression, a new one (the 2nd DF) could become the dominant follicle before an ovulation would occur [from the 3rd DF in fact the ovulatory follicle (OF)]. Thus, follicular development occurred in a wave-like pattern of which typical examples are illustrated in Fig. 1.

The mean time intervals from parturition to the first, second and third ovulation were 39.8 ± 3.38 day (n = 16), 51.1 ± 3.73 day (n = 16) and 66.0 ± 6.25 day (n = 6), respectively. The mean diameters of ovulatory follicles slightly increased from the first, second to third ovulation (13.50 ± 0.52 mm, n = 16; 14.31 ± 0.38, n = 16; 14.17 ± 1.08 mm, n = 6). The growth rate and maximum diameter of ovulatory follicles did not differ between the first, second, third and fourth ovulations ($p > 0.05$) (Table 1).

After the first ovulation, most buffaloes (15/16 buffaloes, 93.75%) showed a short oestrous cycle (10.2 ± 0.38 day) in accordance with the first DF being the ovulatory follicle during the cycle. Short cycles decreased in prevalence thereafter (50%, 3/6 buffaloes, 12.0 ± 1.53 day, between the 2nd and 3rd ovulations). Only buffalo no.4 showed a long oestrous cycle (1/16, 6.25%, 27 day) after the 1st ovulation (Table 2 and Fig. 1).

The mean diameter of an ovulatory follicle that was followed by pregnancy and that of the resulting corpus luteum was significantly larger than those in the non-pregnant animal (Table 3).

Plasma progesterone (P_4) remained below 0.7 ng/ml from the first week pp until the 1st ovulation occurred. After the 1st ovulation, the subsequent P_4 levels increased (Fig. 1). Plasma P_4 levels ranged between 1.2 and 3.2 ng/ml in pregnant buffaloes.

Only a few buffalo cows showed more or less obvious oestrous signs (attempt to mount, being mounted, being

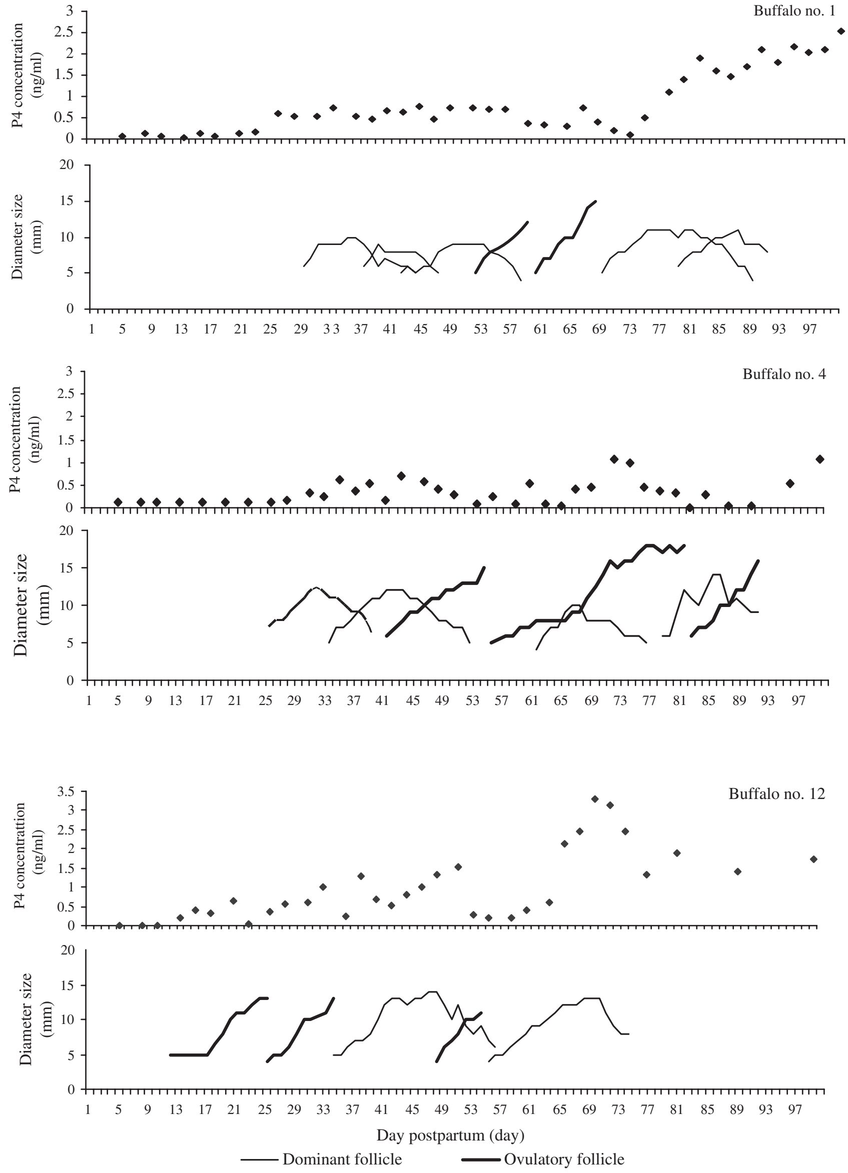

Fig. 1. Representative examples (buffalo cows 1, 4 and 12) of diameter changes for the dominant (DF) and ovulatory (OF) follicles during postpartum follicle waves. P4: progesterone concentrations (ng/ml) in peripheral blood

Table 1. Growth rate, static phase, regression rate and maximum diameter of preceding non-ovulatory dominant follicles (DF) and ovulatory follicles (OF), in 16 postpartum swamp buffalo cows (Mean ± SEM)

Follicle >10 mm before/ and at ovulation	Follicle (n)	Growth rate (mm/day)	Static phase (day)	Regression rate (mm/day)	Maximum diameter (mm)
Last but one DF	5	0.86 ± 0.04	3.00 ± 0.63	0.98 ± 0.06	10.80 ± 0.20[a]
Last DF	14	0.83 ± 0.05	3.11 ± 0.51	0.88 ± 0.07	12.92 ± 0.96
1st OF	16	1.02 ± 0.07	–	–	13.50 ± 0.52
2nd OF	16	1.09 ± 0.04	–	–	14.31 ± 0.38
3rd OF	6	1.15 ± 0.08	–	–	14.17 ± 1.08
4th OF	1	0.9	–	–	14
All OF	39				14.3 ± 0.46[b]

[ab]Values with a different superscript (within column) are significantly different at the 0.05 level using a one-sample *T*-test.

smelled by other cows and being smelled by a bull) or the presence of cervical mucus during the 1st ovulatory postpartum oestrus (75.0% by camera observation, 31.3% by personal observation and 31.3% by mucus presence, n = 16). Oestrous signs were more obvious and easier to detect during the 2nd ovulatory postpartum oestrus increasing to 93.8% through camera observation, 75.0% by personal observation and 68.8% by mucus presence (n = 16). This level was maintained thereafter at the 3rd ovulatory postpartum oestrus: 100.0% by camera, 50.0% by personal and 33.3% by mucus presence (n = 6) up to the 4th ovulatory postpartum oestrus (100% by three methods, n = 1) as summarized in Table 4.

Table 2. Length of oestrous cycle postpartum, diameter of ovulation follicles, of corpora lutea and duration of inter-ovulation intervals between of the 1st–2nd and the 2nd–3rd ovulations in 16 postpartum swamp buffaloes

Inter-ovulation interval	Estrus cycle after parturition	No. of animal	Mean (±SEM) of ovulating follicle diameter(mm)	Mean (±SEM) of CL diameter(mm)	Mean (±SEM) inter-ovulation interval (days)
1st–2nd ovulation	Short	15	13.20 ± 0.49	11.91 ± 0.33	10.20 ± 0.38
	Long	1	15.0	14.13	27.0
2nd–3rd ovulation	Short	3	16.33 ± 0.33	13.48 ± 1.49	12.00 ± 1.53
	Normal	3	12.00 ± 1.00	11.88 ± 1.79	20.33 ± 0.33

Table 3. Mean (±SEM) diameter of ovulating follicles followed by a pregnant or a non-pregnant state and the diameter of the respective corpus luteum in 16 postpartum swamp buffaloes

Status	Ovulating follicles/ corpora lutea (n)	Mean (±SEM) diameter (mm) of ovulating follicle	Mean (±SEM) diameter (mm) of corpus luteum
Pregnant	14	14.50 ± 0.39[a] (range: 11.00–17.00)	14.76 ± 0.34[a] (range: 12.67–16.92)
Non-pregnant	23	13.70 ± 0.44[b] (range: 10.00–17.00)	12.21 ± 0.37[b] (range: 8.88–15.64)

[ab]Values with a different superscript (within column) are significantly different at the 0.05 level using a one-sample *T*-test.

Table 4. Signs of oestrus recorded either by personal observation, video camera observation and cervical mucus discharge from 3 days before ovulation (1–4 ovulations) in 16 postpartum swamp buffaloes

Estrus detection method	Number of buffaloes showing estrus signs 3 days before ovulation (%)			
	1st ovulation (n = 16)	2nd ovulation (n = 16)	3rd ovulation (n = 6)	4th ovulation (n = 1)
Personal observation	5 (31.25)	12 (75.00)	3 (50.00)	1 (100.00)
Camera observation	12 (75.00)	15 (93.75)	6 (100.00)	1 (100.00)
Mucus presence during rectal palpation	5 (31.25)	11 (68.75)	2 (33.33)	1 (100.00)

The results indicate that there were oestrous signs from 3 days before ovulation onwards, but in varying quantity and intensity. In this experiment, oestrous signs were observed by personal observation between 07:00–08:00 hour and 17:00–18:00 hour, differing slightly from the video camera recordings (06:00–07:00 hour and 18:00–19:00 hour) (Fig. 2). The lowest ambient temperature was recorded during the early morning oestrous recordings (Fig. 2).

Discussion

After parturition, follicular growth in Thai swamp buffaloes showed a wave-like pattern with one or two waves in each oestrous cycle. The growth rate and the largest size of the first postpartum ovulating follicle were similar to those in a study in the Italian buffalo (Presicce et al. 2005). However, the mean (±SEM) interval from parturition to the first postpartum ovulation (39.81 ± 3.38 day pp, n = 16) in this study was remarkably shorter than that found in limited suckled Nili-Ravi buffaloes (60.7 ± 3.3 day, Usmani et al. 2000) and suckled buffaloes (88.0 ± 4.7 day, Jainudeen et al. 1983) but quite longer than in river buffaloes studied by Presicce et al. (2005). In the last study, the climate conditions were Mediterranean and not tropical. Short oestrous cycles were common between the 1st and 2nd ovulation in our study similar to a report in milked buffaloes (Usmani et al. 1990). It is likely that follicular dynamics in most suckling swamp buffaloes becomes to be normal after the second ovulation (13/16, 81.3%, this study).

Silent heat is a major problem for heat detection in buffaloes (Jainudeen 1986). In our study, the oestrous symptoms that were evaluated by three methods prior to ovulation were rather vague or absent at the first ovulation but evident in the second, third and fourth postpartum ovulatory periods. The close-circuit camera probably is the best tool for oestrous detection owing to its higher sensitivity and efficiency than other methods (Cavalieri et al. 2003). For the local farmer who cannot afford a camera, the best time for daily personal evaluation of oestrous behaviour is between 06:00 and 08:00 hour because of the less stressful and relatively cool weather conditions. This result is supported by Srivastava and Sahni (2003), who reported that buffaloes showed more oestrous activity in the morning (06:00–07:30 hour) than in the afternoon (14:00–15:30 hour) or during the night (22:00–23:30 hour).

The season of calving influences ovarian activity in buffaloes. Singh and Nanda (1993) reported that the ovarian activity after calving was significantly delayed in buffaloes that calved between February and May (116–148 day), compared to the rest of the year (36–64 day). Furthermore, Tailor et al. (1990) reported that 60.7% of the buffaloes showed oestrous activity in winter, 17.6% in the rainy season, 17.3% in spring and 4.2% in summer. The temperature conditions in Thailand do not

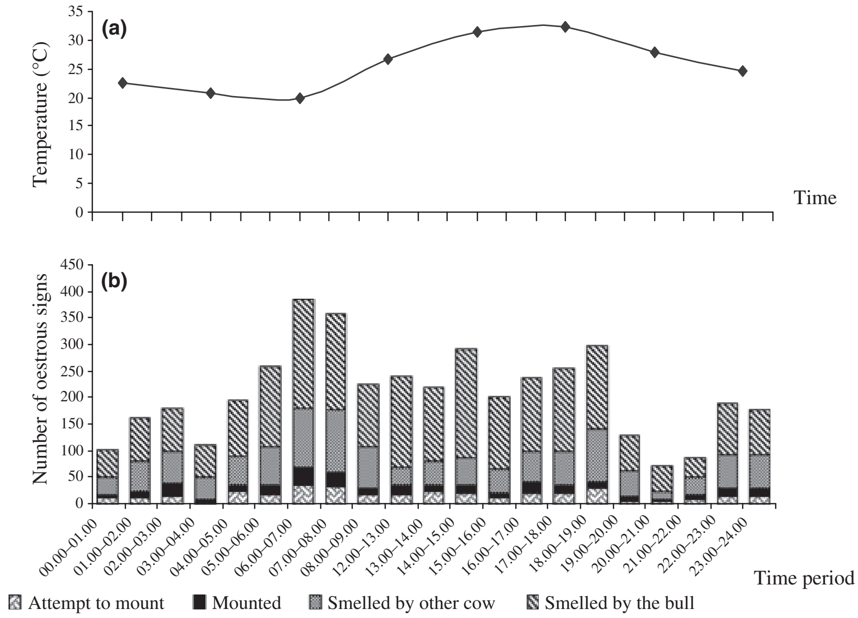

Fig. 2. (a) Means of ambient temperature (recorded every 3 h) for period November 2004–February 2005. (b) Summary oestrous behaviour events (total number of signs) observed in 16 postpartum swamp buffaloes from 3 days before to spontaneous ovulation using video camera recordings for 21 h per days (except during 09:00–12:00 hour)

significantly differ between seasons, but adequate nutrition is more difficult to provide during the summer period. In this study, the experiment was performed during the relatively cool season of the year when high numbers of animals calved (non-published data). To identify a possible annual pattern of ovarian activity in the swamp buffalo, the oestrous activity of Thai swamp buffaloes should also be studied in the other seasons.

In general, the main feed of buffaloes is rice straw and stubble especially during the dry season, and 60–70% of all households keep one to less than five buffaloes. Moreover, the villager generally selects a well-developed, big bull to be castrated for work, because the castrated animals will become tamer and easier to handle (Chantaraprateep et al. 1987) keeping a physically somewhat retarded animal for breeding. This strategy might result in a low reproductive performance in the swamp buffalo. To improve postpartum buffalo reproductive efficiency, proper husbandry management such as good quality nutrition, optimal oestrous detection and a fertile buffalo bull is crucial. In this study, the postpartum buffalo could be successfully mated within 2 months pp at the second spontaneous ovulation, even without hormonal treatment. Adequate oestrous detection methods are essential for the evaluation of cyclicity. Lack of oestrous behaviour can be a major problem as reported by Jainudeen et al. (1983) who diagnosed anoestrus in 26 out of 38 suckling swamp buffaloes during the first 150 days postpartum although functional corpora lutea did develop.

In conclusion, our study demonstrates that follicles of the postpartum swamp buffalo cow developed in a wave-like pattern that appeared to be irregular at the start. Poor oestrous signs and a short subsequent cycle were observed to be related to the first ovulation after calving, but normalizing at the 2nd and 3rd ovulations. Observation using video camera recordings clearly proved to be an excellent method to detect oestrus in the swamp buffalo, but this method is time-consuming. However, personal observation is a useful and cheap method for oestrous detection. This study demonstrated that the most oestrous signs were exhibited at 06:00–00:08 hour, when ambient temperatures were slightly lowest. In conclusion, properly fed Thai swamp buffaloes can be mated successfully within 2 months pp, at their 2nd spontaneous ovulation, provided adequate oestrous detection is at least performed daily.

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Conflict of interest

None of the authors have any conflict of interest to declare.

Author contributions

M Yindee, M Techakumphu, C Lohachit, S Sirivaidyapong, A Na-Chiangmai, H Rodriguez-Martinez, GC van der Weyden and B Colenbrander designed the study.

References

Ali A, Abdel-Razek AK, Abdel-Ghaffar S, Glatzel PS, 2003: Ovarian follicular dynamics in buffalo cows (*Bubalus bubalis*). Reprod Dom Anim **38,** 214–218.

Arya JS, Madan ML, 2001: Postpartum gonadotrophins in suckled and weaned buffaloes. Indian Vet J **78,** 406–409.

Brown J, Sue W, Karen S, 2005: Endocrine manual for reproductive assessment of domestic and non-domestic species. In: Brown J (ed.), Endocrine Workbook. Smithso-

nian's National Zoological Park, Conservation & Research Center, Washington, DC, USA, pp. 57–58.
Cavalieri J, Flinker LR, Anderson GA, Macmillan KL, 2003: Characteristics of oestrus measured using visual observation and radiotelemetry. Anim Reprod Sci **76,** 1–12.
Chantarakhana C, Usankornkul S, Kamnerdpecth V, Phuket NSR, Veerasit P, Pookesorn W, 1981: Age of First Calving and Calving Development Project. Chulalongkorn University, Bangkok, Thailand, pp. 50–55.
Chantaraprateep P, Virakul P, Lohachit C, Kunavongkrit A 1987. Buffalo production in the village and problems. In: Chantaaprateep P (ed.), Swamp Buffalo Reproduction, ISBN 974-567-005-7. Dep Obst Gyn and Reprod., Bangkok, pp. 271–280.
Dinsmore RP, Cattell MB, 1993: Field trial of a radiotelemetry estrus detection system. J Dairy Sci **76**(Suppl. 1)**,** 227.
Henao G, Oliver-Anger M, Maldoado-Estrada JG, 2000: Follicular dynamics during postpartum anestrous and the first estrous cycle in suckled or non-suckled Brahman (*Bos indicus*) cows. Anim Reprod Sci **63,** 127–136.
Jainudeen MR, 1986: Reproduction of the water buffalo. In: Morrow DA (ed.), Current Therapy in Theriogenology. WB Saunders, Philadelphia, pp. 443–449.
Jainudeen MR, Bongso TA, Tan HS, 1983: Postpartum ovarian activity and uterine involution in the suckled swamp buffalo (*Bubalus bubalis*). Anim Reprod Sci **5,** 181–190.
Manik RS, Singa SK, Palta P, Madan ML, 1998: Effect of presence of a dominant follicle on the superovulatory response in buffalo (*Bubalus bubalis*). Theriogenology **50,** 841–852.
Pelissier CL. 1982: Identification of reproductive problems and their economic consequences. In: Pelissier CL (ed.), Proc Natl Invitational Dairy Cattle Reprod Workshop, Ext Comm Policy Sci Educ Admin., US Dep Agric.
Presicce GA, Parmeggiani A, Senatore EM, Stecco R, Barile VL, De Mauro GJ, De Santis G, Terzano GM, 2003: Hormonal dynamics and follicular turnover in preputeal Mediterranean Italian buffaloes (*Bubalus bubalis*). Theriogenology **60,** 485–493.
Presicce GA, Bella A, Terzano GM, Santis GD, Senatore EM, 2005: Postpartum ovarian follicular dynamics in primiparous and pluriparous Mediterranean Italian Buffaloes (*Bubalus bubalis*). Theriogenology **63,** 1430–1439.
Rastegarnia A, Niasari-Naslaji A, Hovareshti P, Sarhaddi F, Safaei M, 2004: The effect of different doses of Gonadorelin on ovarian follicle dynamics in river buffalo (*Bubalus bubalis*). Theriogenology **62,** 1283–1291.
Sakaguchi M, Sasamoto Y, Uzuki T, Takahashi Y, Yamada Y, 2004: Postpartum ovarian follicular dynamics and estrous activity in lactating dairy cows. J Dairy Sci **87,** 2114–2121.
Singh R, Nanda AS 1993. Environmental variables governing seasonality in buffalo breeding. J Anim Sci. **71**: 119 (abstract).
Sirosis J, Fortune JE, 1988: Ovarian follicular dynamics during the estrous cycle in heifers monitored by real-time ultrasonography. Biol Reprod **39,** 308–317.
Srivastava SK, Sahni KL, 2003: Fertility following twice and thrice daily estrus detection in Murrah buffaloes. Buffalo Bulletin **22,** 59–61.
Tailor SP, Jain LS, Gupta HK, Bhatia JS, 1990: Estrus and conception rates in buffaloes under village conditions. India J Anim Sci **60,** 1020.
Taneja M, Ali A, Signh G, 1996: Ovarian follicular dynamics in water buffalo. Theriogenology **46,** 121–130.
Usmani RM, Dailey RA, Inskeep EK, 1990: Effect of limited sucking and varying prepartum nutrition on postpartum reproductive traits of milked buffaloes. J Dairy Sci **73,** 1564–1570.
Usmani RH, Ahmad N, Shafiq P, Mirza MA, 2000: Effect of subclinical uterine infection on cervical and uterine involution, estrus activity and fertility in postpartum buffaloes. Theriogenology **55,** 563–571.
Vassena R, Adams GP, Mapletoft RJ, Pierson RA, Singh J, 2003: Ultrasound image characteristics of ovarian follicles in relation to oocyte competence and follicular status in cattle. Anim Reprod Sci **76,** 25–41.
Xu ZZ, McKnight DJ, Vishwanath R, Pitt CJ, Burton LJ, 1998: Estrus detection using radiotelemetry or visual observation and tail painting for dairy cows on pasture. J Dairy Sci **81,** 2890–2896.
Zicarelli L. 2001: Buffalo milk production world-wide. In: Dixon WJ (ed.), VI World Buffalo Congress, May 20–23, **1**: 202–230.

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