

Influence of rectal massage of ovaries in anestrus buffaloes

M HONPARKHE¹, S P S GHUMAN², D DADARWAL³, JAGIR SINGH⁴, G S DHALIWAL⁵ and N SINGH⁶

Guru Angad Dev Veterinary and Animal Sciences University, Ludhiana, Punjab 141 004 India

Received: 6 February 2008; Accepted: 5 July 2008

Key words: Anestrus, Buffalo, Ovarian follicle, Progesterone, Rectal massage

Economic losses to dairy farmers rearing buffaloes could be immense due to huge incidence (up to 80%) of anestrus (Nanda *et al.* 2003). Repeated massage of ovaries has been claimed to be responsible for the resumption of cyclicity in ovarian afuction affected dairy cattle (Mwaanga *et al.* 2004). However, whether buffalo ovaries also resume activity following per-rectal ovarian massage is not yet determined. Ovarian massage is a simple treatment method that should be considered especially while dealing with economically disadvantaged buffalo farmers. Hence, the objective of present study is to stimulate the ovaries through repeated rectal massage in anestrus buffaloes.

At the university dairy farm, 15 buffaloes with the history of anestrus were selected for study. Animals were fed green fodder, wheat straw, concentrates including mineral mixture throughout the year according to the recommended feeding schedule. Animals were divided into group 1 (3–4-year-old heifers, n=9, 401.6±10.9 kg b. wt.) and group 2 (pluriparous buffaloes, n=6, 482.5±25.0 kg b. wt.). On every alternate day for 25 days, all the animals were subjected to rectal ovarian (bilateral) massage for 5 min followed by ovarian ultrasound examination and jugular vein blood sampling. Throughout the period of study, estrus detection was carried out by parading a vasectomized bull in all the animals and by critically observing animals for estrus signs.

Ovarian ultrasonographic examination was performed with a battery-operated B-mode ultrasound scanner equipped with inbuilt interchangeable 5/7.5 MHz linear-array rectal transducer (Agroscan). Each ovary was scanned in several planes by moving the transducer along its surface to identify the number and size of the follicles. On the basis of size, the follicles were categorized as small (<5 mm), medium (5–9 mm) and large (>9 mm; Abdoon and Kandil 2001). The stored (–20°C) plasma of the collected blood samples was analyzed

for progesterone concentrations using radioimmunoassay (Ghuman *et al.* 2007). The mean inter- and intra-assay coefficients of variation for the progesterone assay were 7.4 and 8.0% respectively. The sensitivity of the assay was 0.05ng/ml.

Statistical analysis of the data generated on different days regarding the number of follicles or progesterone profile was subjected to generalized linear model ANOVA and *post-hoc* Tukey's method for pairwise comparisons (Fig. 1A-D). In

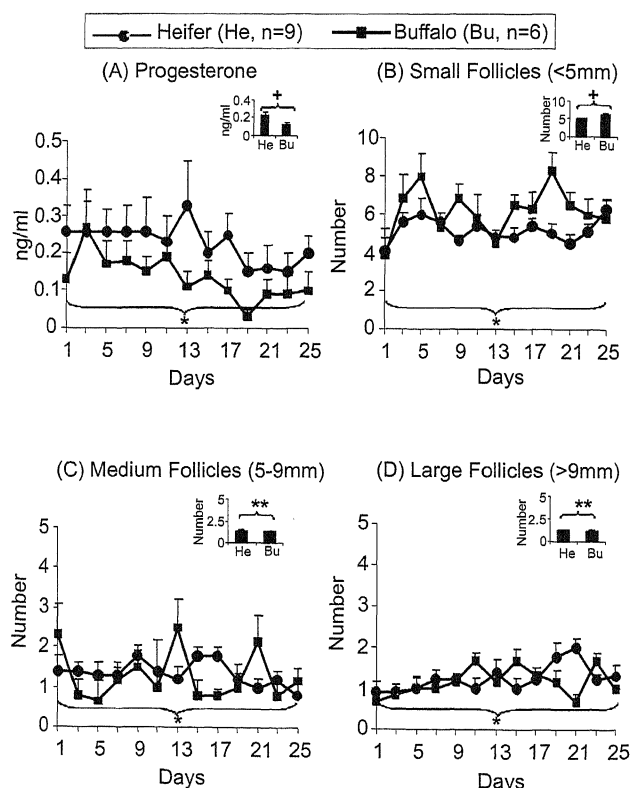


Fig. 1. Ovarian activity and plasma progesterone levels in anestrus heifers and pluriparous buffaloes subjected to repeated per-rectal massage of ovaries. Insets represent mean (±SEM) of collective data of study period in anestrus heifers or pluriparous buffaloes. *P<0.05; *, **P>0.05

Present address: ^{1,3}Assistant Gynaecologist, ²Associate Professor, ⁴Senior Gynaecologist, Department of Animal Reproduction, Gynaecology and Obstetrics ⁵Professor, Department of Veterinary Clinical Services Complex, ⁶Assistant Scientist (Reproduction), Department of Animal Breeding and Genetics.

addition, data of different days for various parameters were pooled and mean values calculated for heifers or pluriparous buffaloes and the differences were examined *post-hoc* using Student's paired t-test (two tail, Fig. 1A-D inset; Dytham 1999). Probabilities of <0.05 were considered significant. Data were expressed as mean values $\pm$ SEM. Statistical analyses were performed using Minitab release 14.2 statistical software.

During the period of study, none of the anestrus heifers or pluriparous buffaloes was detected in estrus by bull parading and failed to show visible signs of estrus, viz. cervico-vaginal mucus discharge and vulvar swelling. Throughout the period of study, plasma progesterone on different days in both the groups remained <0.4 ng/ml (Fig. 1A), which is indicative of true anestrus. Comparison of different days of study period showed that number of small (<5 mm), medium (5–9 mm) or large (>9 mm) follicles were not different between days either in heifers or in pluriparous buffaloes (Fig. 1B-D). In addition, none of the animals ovulated during the study period. These results showed that repeated rectal massage of ovaries does not alter ovarian activity in anestrus buffaloes. These results are in agreement with previous studies in cattle where repeated handling of ovaries at the time of daily ultrasound was not found to modify the course of estrous cycle though ovarian massage was not carried out (Sirois and Fortune 1988).

In the present study, plasma progesterone and number of small follicles recorded in anestrus heifers (0.23 ± 0.04 ng/ml and 5.19 ± 0.15 respectively) were significantly ($P < 0.05$) different compared to those observed in anestrus pluriparous buffaloes (0.13 ± 0.15 ng/ml and 6.20 ± 0.28 , respectively, Fig. 1A, 1B inset), indicating that low plasma progesterone recorded in later might be related with recruitment of higher number of small follicles. However, there was no significant ($P > 0.05$) difference in the number of medium (Fig. 1C inset) or large (Fig. 1D inset) follicles observed in anestrus heifers (1.37 ± 0.83 and 1.25 ± 0.07 respectively) and anestrus pluriparous buffaloes (1.30 ± 0.14 and 1.15 ± 0.07 respectively).

Overall, the results concluded that though differential ovarian activity exists in anestrus heifers and pluriparous

buffaloes, their ovarian activity is not influenced by repeated rectal massage of ovaries.

SUMMARY

Heifers (9) and buffaloes (6) with the history of anestrus were subjected to per-rectal ovarian massage followed by ultrasonography and blood sampling on alternate day for 25 days. During the period of study, none of the heifers or buffaloes was detected in estrus as well as plasma progesterone in both the groups on different days remained <0.4 ng/ml which is indicative of true anestrus. Number of small (<5 mm), medium (5–9 mm) or large (>9 mm) follicles were not different between days and none of the animals ovulated. Plasma progesterone and number of small follicles were different ($P < 0.05$) between the anoestrus heifers and pluriparous buffaloes. This study demonstrated differential ovarian activity in anoestrus heifers and pluriparous buffaloes. Further, repeated per-rectal ovarian massage does not influence ovarian activity in buffaloes.

REFERENCES

- Abdoon A S S and Kandil O M. 2001. Factors affecting number of surface ovarian follicles and oocytes yield and quality in Egyptian buffaloes. *Reproduction, Nutrition and Development* **41**: 71–77.
- Dytham C. 1999. *Choosing and Using Statistics, A Biologist's Guide*. Vol. 2. Blackwell Science Press, London.
- Ghuman S P S, Dinesh D and Dhaliwal G S. 2007. *Radioimmunoassay Laboratory Handbook*. ICAR Centre of Advanced Studies, Department of Animal Reproduction, Gynaecology and Obstetrics, Guru Angad Dev Veterinary and Animal Sciences University, Ludhiana, Punjab.
- Mwaanga A A, Zdunczyk S and Janowski T. 2004. Comparative study on the efficacy of hormonal and non hormonal treatment methods in ovarian afuction affected dairy cow. *Bulletin Veterinary Institute Pullawy* **48**: 265–67.
- Nanda A S, Brar P S and Prabhakar S. 2003. Enhancing reproductive performance in dairy buffalo: major constraints and achievements. *Reproduction Supplement* **61**: 27–36.
- Sirois J and Fortune J E. 1988. Ovarian follicular dynamics during the estrous cycle in heifers monitored by real-time ultrasonography. *Biology of Reproduction* **39**: 308–17.