EFFECT OF SEASON ON SUPEROVULATORY RESPONSE OF NILI RAVI BUFFALO (*Bubalus bubalis*)

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ABSTRACT

The study was conducted to compare the superovularoty response of buffalo in winter and summer. Four adult buffaloes were treated with follicle stimulating hormone (FSH) during winter (November to January) and three buffaloes in summer (June and July). Superovulatory response was determined by estimating the number of corpora lutea (CL) palpated per rectum on day 6 of the oestrus cycle (oestrus = day 0). The number and quality of embryos collected were also studied. A better ovarian response was observed in winter (5.8±0.3 CL) than in summer (2.7±1.2CL) ($P<0.05$). A mean (±SE) 1.3±0.9 embryos were recovered during winter compared to 0.5±0.4 embryos in summer. All the embryos recovered in winter were of good quality whereas only fair quality embryos were recovered in summer. Two pregnancies were achieved in winter after transfer of three embryos to three synchronized recipient buffaloes. The study showed a better ovarian response of buffalo to FSH in winter.

Keywords: Nili-Ravi buffaloes, Superovulatory response, Season, Follicular stimulating hormone

INTRODUCTION

A low ovarian response and embryo recovery has been the most concerning matter since the studies on multiple ovulation and embryo transfer technique started in buffalo (Alexiev *et al.*, 1988; Zicarelli, 1997). Less number of primordial follicles in buffalo as compared to cow (Samad and Nassari, 1979; Danell, 1987) and anatomy of genital organs in buffalo (Ocampo *et al.*, 1988) have been considered the most likely causes for this poor superovulatory response. Seasonal trend in buffalo breeding may also be one of the factors affecting superovulatory response in this species. Although buffalo continues to breed throughout the year, the peak breeding season is from September to February (Shah and Shah, 1968). Breeding activity decreases with an increase in ambient temperature. Incidentally this is also the period of increasing day light that suggests a possible association of photoperiodism in affecting the breeding efficiency in buffalo. The present experiment reports the superovulatory response of buffalo to FSH during winter and summer.

MATERIALS AND METHODS

Non-pregnant, lactating, Nili Ravi buffaloes maintained at National Agricultural Research Centre (NARC), Islamamabd (latitude 33.7° N; longitude 73.1°E; altitude 5.8 meters) were used in this trial. The buffaloes were in first to fourth lactation, and had calved 3-6 months before treatment with FSH. Four buffaloes were superovulated in winter (November to January) and 3 in summer (June and July). The buffaloes used in winter were not used in summer. Follicle stimulating hormone (20 ml Folltropin-V, Veterepharm, Canada Inc.; equivalent to 400 mg NIH-FSH-P) was used to superovulate the buffaloes. The FSH was given i/m in 8 equally divided doses with 12 hours interval starting from day 10 or 11 of the oestrous cycle. Cloprostenol (250 µg, Estrumate-ICI) was administered at 48 hours after the start of FSH treatment. The buffaloes were inseminated twice (12 and 24 hours after the exhibition of standing heat) with fresh buffalo semen collected from a bull of proven fertility. Embryo recovery in each buffalo was performed on day 6 of the oestrous cycle (oestrus = day 0) using one litre Dulbecc's modified phosphate buffered saline solution. Number of ovulations in response to FSH treatment was estimated by counting the number of corpora lutea (CL) through palpation of the ovaries per rectum at the time of embryo recovery. The number and quality of embryos recovered were also studied. Quality and developmental stage of embryos were determined at 40 X of a steriomicroscope. Three embryos recovered during winter were transferred to buffalo recipients (one embryo per recipient) synchronized by injecting cloprostenol. Animals were offered green oats or its silage during winter, and maize and mott grass during summer. Concentrate supplementation having 15-16% crude protein and 70% total digestible nutrients was fed in both the seasons. Average minimum and maximum temperatures were 2.7 and 20.9°C in the winter months and 23.3 and 36.9°C in summer months. Average humidity was 34% (at 8.00 am) and 84% (at 2.00 pm) during winter, and 46% (at 8.00 am) and 67% (at 2.00 pm) during summer. Means (±SE) were computed for number of CL, embryos and cloprostenol to heat interval. Comparisons were made for the data on number of CL, and cloprostenol to heat interval of the two seasons by student's t test, however no comparison could be made between embryos recovered due to their low number.

RESULTS

Superovulatory response of buffaloes to folicle stimulating hormone during winter and summer is presented in Table 1.

Table 1: Superovulatory response of buffalo to follicle stimulating hormone during winter and summer.

	Winter	Summer
Animals	4	3
Cloprostenol-Heat interval (hours)	53.5±9.8	45.3±7.7
Corpora lutea	5.8±0.3 a	2.7±1.2 b
Embryos recovered	1.3±0.9 (3 animals)	0.5±0.4 (2 animals)

a, b: differ significantly (P<0.05)

A higher number of CL was recovered during winter than in summer (P<0.05). Embryo flushing could not be done in one (out of four animals) in winter due to difficulty in catheter passage through the cervix. Four good quality embryos were recovered from three buffaloes in which 17 CL were palpated on the ovaries (23.5% embryo recovery). In summer, three buffaloes were treated with FSH but embryo recovery could be performed in two responding buffaloes. One fair quality embryo was recovered from these buffaloes in which 7 CL were palpated on the ovaries (14% embryo recovery). The third buffalo had only one CL and was not flushed. This buffalo was found pregnant when palpated per rectum after two months. During winter, three good quality embryos were transferred to three synchronized recipients (±6 hours synchrony). Of three, two recipients were found pregnant at two months after *ET AL.* and delivered two female calves 300 days after embryo transfer.

DISCUSSION

A higher superovulatory response was observed during winter in terms of number of CL palpable per rectum number and quality of emrbyos recovered was also better in winter than in summer. These findings support the results of Matharoo and Singh (1997) who recorded an average of 6 CL during winter and 3 CL in summer in buffaloes superovulated with FSH. Their embryo recovery rate was 2.16 and 0.5 during winter and summer, respectively. Results of 36 superovulation in Italian buffalo also showed that time of the year affects the superovulatory response (Zicarelli, 1997). Summer season has been associated with a lower oestrus activity (Patel *et al.*, 1996) and lower conception rate (Khattab *et al.*, 1996) compared to that in winter season in buffalo. Summer has also ben observed as adversely affecting GnRH stimulated LH release in buffalo (Palta and Madan, 1996). It seems that exogenous FSH is unable to override the seasonal reproduction trend in buffalo.

Although a better ovarian response was recorded in buffalo in winter, the number of embryos recovered was considerable lower than the number of CL palpated per rectum in the present study. Better embryo recovery (31-57%) has been reported in some other studies in buffalo disregard to season of recovery (Drost *et al.*, 1988; Misra, 1993; Samad *et al.*, 1996). Matharoo and Singh (1997) were able to recover 36% embryos in winter and 17% during summer. Differences in non surgical recovery techniques might have resulted in this dissimilarity. In cattle, 40-80% ova can usually be recovered from superovulated intact cows (Hafez, 1993). In an earlier study at our laboratory, eight buffaloes were slaughtered after superovulation which yielded embryos representing 53% of the number f CL (Anwar and Ullah, 1998). Misra *et al.* (1998) recovered 47% eggs from 18 superovulated buffaloes slaughtered for embryo recovery. Although in both aforementioned studies, animals were slaughtered at various time intervals after insemination to study embryonic development, they provide a picture of surgical embryo recovery rate in superovulated buffalo.

The pregnancies were achieved after transfer of three good quality embryos to synchronzed buffaloes. Misra *et al.* (1999) recorded an overall pregnancy rate of 26.4% after 91 embryo transfer in buffalo, however, amongst these transfers, high pregnancy rates were achieved after transfer of better quality embryos (60%) and a closer oestrus synchrony (40.7%).

The limited data of this study indicated a better ovarian response to FSH in winter than in summer.

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REFERENCES

Alexiev, A., K. Vlakvov, Ch. Daraivanov, D. Kacheva, O. Polykhronov, M. Petrov, N. Nikolov, A. drogoev and P. Radev, 1988. Embryo transfer in buffaloes in Bulgaria. Proc. Second Buff. Cong., New Dehli, II: 591-195.

Anwar, M. and N. Ullah, 1998. Early development and location of embryos in the reproductive tract of Nili Ravi burffalo (Babalus babalis): a retrospective analysis, Theriogenology, 49: 1187-1193.

Danell, B., 1987. Oestrus behaviour, ovarian morphology and cyclical variation in follicular system and endocrine pattern in water buffalo heifers. Ph.D. Thesis, Deptt. Obstet. Gynae., Swedish Univ. Agri. Sci., Uppsala, Sweden.

Drost, M., A. Alexiev, K. Vlahov, Ch. Karaivanov, W.S. Cripe, A.P. Leonards, D. Kacheva, O. Polyhronov, N. Nikolov, M. Petrov and A. Dragoev, 1988. Successful non surgical transfer in buffalo (Babalus babalis) in Bulgaria. Theriogenology, 30: 659-668.

Hafez, E.S.E., 1993. Assisted reproductive technology. In: Reproduction in Farm Animals. 6th Ed., Lea and Febriger, Philadelphia, pp: 461-502.

Khattab, R.M., M.A.R. Ibrahim, M.K. Mohsen and A.I.S. El-Shama, 1996.Improvig oestrous expression of Egyptian buffalo using different anallogous of prostaglandin. Annals Agri. Sci., 34: 549-554.

Matharoo, J.S. and M. Singh, 1997. Seasonal variations in superovulatory response and embryo recovery in buffaloes (Babalus bubalis). Indian J. Anim. Reprod., 18: 1-5.

Misra, A.K., 1993. Superovulation and embryo transfer in buffalo; progress problems and future prospects in India. Buffalo J., 9: 13-24.

Misra, A.K., R. Kasiraj, M.M. Rao, N.S. Rangareddy, R.S. Jaiswal and H.C. pant, 1998. Rate of transport and development of preimplantation embryo in the superovulated buffalo (*Bubalus bubalis*). Theriogenology, 50: 637-649.

Misra, A.K., M.M. rao, R. Kasiraj, N.S.R. Reddy and H.C. Pant, 1999. Factors affecting pregnancy rate following nonsurgical embryo transfer in buffalo (*Bubalus bubalis*): a retrospective study. Theriogenology, 52: 1-10.

Ocampo, M.B., R.S. Uenish, C.A. Valder, J. Pastor, L. Cruz and H. Kanagawa, 1988. Non surgical embryo recovery in the water buffalo. Japanese J. Vet. Res., 36: 257-263.

Palta, P. and M.L. Madan, 1996.Seasonal variation in the hypophysial responsiveness to GnRH in cycling buffalo (*Bubalus bubalis*). Asian Australasian J. Anim. Sci., 9: 711-714.

Patel, K.K., F.S. Kavani and J. Ninan, 1996. Effect of season and climatological factors in sexual activity of Mehsani buffaloes. Intl. J. Anim. Sci., 11: 329-332.

Samad, H.A. and A.A. Nassari, 1979. A quantitive study of promordial follicles in buffalo heifer ovaries. Compendium, 13, FAO/SIDa Int. Course Anim. Reprodc., Sweden.

Samad, H.A. Z.I. Qureshi and L.A. Lodhi, 1996. Superovulation response, non-surgical recovery and morphology of Nili Ravi buffalo embryos. Pakistan Vet. J., 16: 54-57.

Shah, N.H. and S.K. Shah, 1968. Seasonal influence on oestrus incidence and conception rate in buffaloes. Pakistan J. Agri Sci., 5: 222-227.

Zicarelli, L., 1997. News on buffalo cow reproduction. Vth Buffalo Congress, Caserta, 13-16 October, Italy, pp: 124-141.