

## Hormonal induction of oestrus in anoestrous buffaloes

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### ABSTRACT

The efficacy of a 9 day progestagen treatment in combination with eCG was evaluated for induction of ovulatory oestrus, restoration of ovarian cyclicity and fertility in rural anoestrous buffaloes maintained either on paddy straw plus dried jowar (group I) or with urea molasses mineral mixture block (UMMB) supplement (group II) during their non-breeding season (summer). Hormonal therapy was quite effective in the induction of oestrus but the conception rate at the induced oestrus was very poor (8.3% vs 13.5%). The UMMB supplement improved the energy balance of animals (serum glucose value; 24.8 vs 50.3 mg %) and consequently the incidence of ovulatory oestrus (16.7 vs 53.3%) and the establishment of ovarian cyclicity (25.0 vs 60.0%). Further studies are needed to delineate the role of nutrition and its interaction with season in assessing the response to hormonal therapy for treatment of true anoestrus in rural buffaloes.

**Key words :** Anoestrous, buffalo, hormonal therapy, serum glucose, progesterone, fertility

### INTRODUCTION

In the buffalo, true anoestrus is a major economic problem, which leads to a long calving interval (Rao and Rao, 1970; Pant & Roy, 1972). In our previous study, treatment of rural anoestrous buffaloes with norgestomet ear implant and equine chorionic gonadotrophin (eCG) was found to be highly effective in inducing oestrus but conception rate at induced oestrus and establishment of ovarian cyclicity were low (Pant, 2000). This was attributed to extremely poor plane of nutrition as the animals were maintained only on paddy straw and dried jowar (*Sorghum vulgare*) without any salt and mineral supplement. Therefore, the present study was designed to ascertain if provision of urea molasses mineral mixture block (UMMB) for at least two months prior to hormonal intervention could be of value in improving conception rate at the induced oestrus.

### MATERIAL AND METHODS

**Animals :** Thirty, parous, non-lactating, anoestrous animals, aged approximately 5 to 8 years which belonged to small-holder farmers of Sabarkantha District were employed during non-breeding season (Summer). They

were confirmed to be functionally anoestrous for > 6 months based on their clinical history and on per rectal palpation of internal genitalia and ovaries.

All animals were dewormed by administration of Albendazole (Kalbend, Karnatak Antibiotic and Pharmaceutical Ltd.). They were maintained on a basal ration of paddy straw and dried jowar. A sub-group of 18 animals was supplemented with Urea Molasses Mineral Mixture Block (UMMB) free choice, which was made available during the night. All farmers whose animals were selected for treatment were also given a two months supply of mineral mixture (Kalmin, Cadilla) at the rate of 30 g per animal per day.

**Treatment :** After two months on basal diet (group I) or on UMMB supplement (group II), all animals received a subcutaneous ear implant (Crestar; Intervet International, Boxmeer, The Netherlands) consisting of a silicone implant containing 3 mg norgestomet. The implant was removed after nine days when all animals were given an intramuscular injection of 500 i.u. of equine chorionic gonadotrophin (eCG) (Folligon, Intervet) and of a luteolytic dose of PGF<sub>2</sub>α (Prosolin, Intervet). Following the treatment, the oestrus response was checked by the owner and verified by a veterinarian at the time of AI. Fixed time insemination with frozen thawed semen from a fertile bull was done thrice at approximately 48, 60 and

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72 hours after implant withdrawal. All animals were given an intramuscular injection of 2.5 ml of a synthetic gonadotrophin releasing hormone preparation (Receptal, Hoechst) at the time of first AI.

Animals were examined for pregnancy at 45-60 days after AI at the induced oestrus. Animals which failed to conceive at the induced oestrus were followed for exhibition of first natural oestrus.

Establishment of ovarian cyclicity was calculated as :

$$\frac{\text{No. conceived at induced oestrus} + \text{No. exhibiting first natural oestrus}}{\text{Total number employed}} \times 100$$

Samples of jugular venous blood were collected in non-heparinized vacutainer tubes prior to implant insertion and between days 10-12 after induced oestrus. Progesterone was estimated in serum by a direct solid phase <sup>125</sup>I radioimmunoassay method validated earlier

(Misra *et al.*, 1998). Serum glucose and urea levels were estimated by an autoanalyser.

**Statistical analysis :** The data for oestrous response and conception rate were analysed by Chi-square test and means were compared by student's 't' test. (Steel and Torrie, 1960).

## RESULTS AND DISCUSSION

Results indicated that 27 animals which retained ear implant exhibited oestrus within 36 to 48 hour of implant removal (Table 1). This is in agreement with earlier reports (Rao and Sreemannarayan, 1983; Singh *et al.*, 1983; Shah *et al.*, 1987; Misra, 1996). The conception rate following AI at the induced oestrus was 1/12 (8.3%) and 2/15 (13.3%) in the two groups; the 5.0% increase in the group with UMMB supplement was not significant. The overall conception rate was 3/27 (11.1%) which was very low compared to some earlier reports of 46.2% to 53.3% (Rao and Sreemannarayan, 1983; Singh *et al.*, 1983; Shah *et al.*, 1987). This may be due to low plane of nutrition alongwith heat stress

**Table 1. Reproductive performance of anoestrous buffaloes treated with norgestomet ear implant with or without UMMB supplement**

	Group I without UMMB	Group II with UMMB	Total
Animals (n)	12	18	30
Animals retaining implant (%)	12 (100.0)	15 (83.3)	27 (90.0)
Animals of induced oestrus (%)	12 (100.0)	15 (100.0)	27 (100.0)
Animals conceived at induced oestrous (%)	1/12 (8.3)	2/15 (13.3)	3/27 (11.1)
Animals exhibiting first natural oestrus (%)	2/11 (18.2)	7/13 (53.8)	9/24 (37.5)
Animals relapsing into anoestrous state (%)	9/11 (81.8)	6/13 (46.1)	15/24 (62.5)
Establishment of ovarian activity (%)	3/12 (25.0)	9/15 (60.0)	12/27 (44.4)

**Table 2. Concentrations of glucose (mg/100 ml) and urea in the serum of animals (mean±S.E.M.)**

Attributes	Group I without UMMB	Group II with UMMB
No. of animals	10	18
Glucose	24.8±3.5 <sup>a</sup>	50.3±2.4 <sup>b</sup>
Urea	31.9±3.8	30.1±1.6

Means with in a row with different superscripts differ significantly (P<0.01; Students unpaired "t" test)

(maximum ambient temperature 45-50°C). Some authors have shown that fertility in buffalo cows was compromised on low feeding levels during period of high ambient temperature (Raizada *et al.*, 1969; Kaura and Arora, 1982). Although, the provision of UMMB supplement did improve the energy balance as evidenced by a significant increase in serum glucose (Table 2), however, it was probably not adequate in correcting the nutritional imbalance completely and in enabling the buffaloes to sustain pregnancy. Detailed metabolic profile tests (Payne, 1970) need to be done in well fed as well as undernourished village buffaloes to establish "normal" values and confidence limits for formulating feed supplementation strategies.

Measurement of serum progesterone revealed that prior to implant insertion all animals had basal progesterone levels of <0.2 ng/ml, which confirmed their anoestrous state. However, following induced oestrus elevated progesterone values (>1.0 ng/ml) were encountered in only 16.7 and 53.3% animals in two groups, respectively indicating that hormonal therapy was more effective in inducing ovulation in animals given UMMB supplement. In an earlier study Shah *et al.* (1987) reported this hormonal therapy was effective in inducing ovulation in 70% of acyclic buffaloes kept under high standards of management and nutrition.

Persual of Table 1 further reveals the beneficial effect of UMMB supplement in that in group I of II animals which failed to conceive at the induced oestrus 2 (18.2%) exhibited first natural oestrus, while in group II of 13 animals which did not conceive at the induced oestrus 7 (53.8%) exhibited first natural oestrus; the difference between groups approached significance ( $X^2 = 3.17$ ;  $P < 0.10$ ). Also, the ovarian cyclicity was established in 25.0% and 60.0% animals in the two groups, respectively and the difference approached significance ( $X^2 = 3.53$ ;  $P < 0.10$ ).

In conclusion, present results clearly indicate that under existing managerial conditions, hormonal therapy for improving fertility of rural anoestrous buffaloes seems to be valueless and can not be a substitute for good management. Further studies are needed to

delineate the role of nutrition and its interaction with season in assessing the response to hormonal intervention for treatment of true anoestrous in rural buffaloes.

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