

## Effect of oxytocin and some herbal preparations on induction of uterine contraction and expulsion of foetal membranes in buffaloes\*

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Received : June 6, 2001

Accepted : April 20, 2002

### ABSTRACT

Experiments were designed to study the efficacy of oxytocin and some herbal preparations in inducing uterine muscle contraction *in vitro* (experiment I), expulsion of fetal membranes for prevention of retention of fetal membranes (experiment II) and treatment of buffaloes with RFM (experiment III). The result revealed dose dependent uterine contraction of estrus goat uteri, and reduction in the time required for expulsion of fetal membranes in buffaloes treated with uterotone, exapar, replanta and involon than the control. The difference for expulsion of fetal membranes between control and uterotone, exapar involon group was significant. There was no significant difference for onset of first post partum estrus in control and treatment groups. Experiment III revealed no significant difference on time required for expulsion of placenta and onset of first partum estrus.

**Key words :** Herbal preparation, foetal membranes, oxytocin, uterine contraction

Retention of fetal membranes in cows and buffaloes constitutes one of the major postpartum complications which often lead to severe endometritis, pyometra, perimetritis, ovaritis (Roberts, 1971), cystitis and peritonitis (Wetherill, 1965) and vaginal prolapse. All these conditions result into drastic reduction in milk yield and affect the fertility (Laven and Peters, 1996) characterized mainly by prolonged calving interval (Joosten *et al.*, 1988; Arthur *et al.*, 1996) and repeat breeding (Narsimhan and Deopurkar, 1994). Various myometrial stimulants like ergonovine, stilbesterol and oxytocin (Roberts, 1971; Sharma, 1976) have been tried for the treatment of retained fetal membranes in cows and buffaloes. A combination of prostaglandin F<sub>2</sub>, alpha plus oxytocin may prove to be of immense value in expelling retained fetal membranes (Ediqvist *et al.*, 1975; Patil *et al.*, 1980; Shaw, 1938). Emphasis has also been claimed to have therapeutic efficacy. The drugs capable of inducing myometrial contraction may be useful for hastening the expulsion of fetal membranes or in prevention of retention of fetal membranes. However, systematic study to evaluate the efficacy of these products in stimulating contraction of uterine muscle and expulsion of fetal membranes is lacking. Hence, experiments were designed to evaluate the efficacy of herbal products in induction of uterine muscle contraction *in vitro*,

prevention of fetal membranes retention and treatment of retained fetal membranes in buffaloes.

### MATERIALS AND METHODS

#### *In vitro* studies (Experiment I)

**Collection and preparation of tissues :** Estrus goat uteri were obtained from local slaughter house. The estrus goat uteri were confirmed by the presence of estrual fluid in uterine lumen, relaxed external os and presence of mature graafian follicles on the ovary. The uteri thus obtained were collected immediately after slaughter in the De-Jalon solution (composition in mM-NaCl-154, KCl-5.6, CaCl<sub>2</sub>-0.55, NaHCO<sub>3</sub>-6.0 and Glucose-2.78) at 2-8°C.

Small pieces (25 x 2 mm<sup>2</sup>) of tissues were cut from the greater curvature and taken into a petridish containing De-Jalon solution at 29-30°C with continuous aeration. Then the tissues were cleaned and mounted on a tissue bath with 30 ml De-Jalon solution at 29-30°C. A tension of 1 gm weight was put to the tissues. They were continuously airted and allowed to equilibrated for 60 minutes. During the whole experimental period the bath solution was changed every 10 minutes.

After equilibrium, tissues preparations were treated with drugs viz., oxytocin, uterotone, exapar, replanta and involon. The contraction induced by the drug was recorded on a chymograph.

\* Part of M.V.Sc. thesis

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After every treatment the tissue were allowed to come to their normal resting state to record the effect of next drug. For each drug, records of six tissue preparations were taken to find out mean and standard error (mean $\pm$ S.E.).

#### Preparation of herbal products and drugs for *in vitro* study

Oxytocin (Syntocinon-Novartis India Limited) was diluted to 5 mu/ml, 10 mu/ml, 20 mu/ml and 40 mu/ml in De-Jalon solution.

Uterotone (Cattle Remedies) and Exapar (Dabur Ayurved) 1 ml, 2 ml, 4 ml liquid preparation as such was added in bath solution to make 31 ml. Replanta (Natural Remedies) 1 gm powder was dissolved in 5 ml of De-Jalon solution. Involon (Natural Remedies) 2 bolus were dissolved in 31 ml of De-Jalon solution to get concentration of 1.0 g/5 ml. These solutions were filtered with Whatman filter paper number 40 and filtrate was used in the study. Difference doses of replanta solution (1 ml, 2 ml, 4 ml) and involon solution (2 ml, 3 ml, 4 ml) were added in bath solution to make a total volume of 31 ml with different concentration of drugs.

#### *In vivo* studies

**Prevention (Experiment II) :** Buffaloes after calving from different dairies near to Drug and Bhilai were randomly divided into 5 groups. Each group was having 6 animals. Buffaloes in group I were kept as control while buffaloes in treatment groups were treated with one of the herbal preparation viz., Uterotone (125 ml two times a day at 12 hrs interval for 5 days). Exapar (100 ml two times a day on day of calving and then 50 ml two times a day for 3 days), Replanta (100 gms two times a day on day of calving and then 50 gms a day for 3 days), Involon (1 bolus two times a day for 3 days), orally after calving.

Observations were recorded in control and treatment groups for 12 hrs for average time (hours) required for shedding of fetal membranes after calving and for 3 months for appearance of first post partum estrus.

**Treatment (Experiment III) :** Buffaloes after calving with retained placenta for 12 hrs were randomly divided into 5 groups. Each group was having 6 animals. Buffaloes in group I were kept as control while buffaloes in treatment groups were treated with one of the herbal preparation viz., uterotone (300 ml at 3 hrs interval for 5 days) and Exapar, Replanta, Involon in the dose schedule mentioned for prevention. Buffaloes in all groups were observed for 12 hrs for expulsion of fetal membranes and 3 months for appearance of first post partum estrus.

### RESULTS AND DISCUSSION

**Experiment I :** The mean contraction of uterine muscle *in vitro* was 42.66 $\pm$ 3.58 mm, 79.05 $\pm$ 1.77 mm, 65.77 $\pm$ 1.63 mm, 48.33 $\pm$ 4.59

mm and 64.55 $\pm$ 1.67 mm with oxytocin, uterotone, exapar, replanta and involon, respectively. The mean contraction of uterine tissues in different doses of oxytocin viz., 0.5 x 10<sup>-2</sup> IU/ml, 1 x 10<sup>-2</sup> IU/ml, 2 x 10<sup>-2</sup> IU/ml and 4 x 10<sup>-2</sup> IU/ml was 16.00 $\pm$ 1.73, 29.33 $\pm$ 3.08, 49.83 $\pm$ 6.23 and 75.5 $\pm$ 3.31 mm, respectively. The mean contraction of uterine tissues *in vitro* with 1 ml, 2 ml and 4 ml of uterotone was 23.67 $\pm$ 2.01, 67.33 $\pm$ 3.11 and 114.67 $\pm$ 0.21 mm, respectively. Exapar given at the dose rate of 1 ml, 2 ml and 4 ml resulted mean uterine muscle contraction of 37.33 $\pm$ 1.48, 73.67 $\pm$ 1.28 and 86.33 $\pm$ 2.14 mm, respectively. 1 ml, 2 ml and 4 ml filtrate of replanta powder resulted uterine contraction of 24.83 $\pm$ 3.41, 56.33 $\pm$ 5.18 and 63.83 $\pm$ 5.18 mm respectively. the 2 ml, 3 ml and 4 ml of involon solution in bath solution resulted uterine contraction of 33.53 $\pm$ 1.47, 72.33 $\pm$ 0.92 and 86.00 $\pm$ 2.69 mm, respectively. Statistically all the drugs has dose dependent significant ( $P < 0.05$ ) effect on induction of uterine contraction.

In the available literature there is no report regarding to the *in vitro* effect of herbal preparations on uterine contraction. Edqvist *et al.* (1975) mentioned the *in vitro* effect of oxytocin on tubular genital tract of ewes. The present studies established the effect of oxytocin and herbal preparations namely uterotone, exapar, replanta and involon on inducing contraction in estrus goat uteri.

**Experiment II :** The mean time (hrs) interval required for expulsion of fetal membranes in buffaloes in control group (without any treatment) was higher (7.33 $\pm$ 0.74 hrs) as compared to group treated with uterotone (5.29 $\pm$ 0.48 hrs), exapar (4.88 $\pm$ 0.51 hrs), replanta (5.56 $\pm$ 0.65 hrs) and involon (4.71 $\pm$ 0.52 hrs). The difference in the mean time required for expulsion of fetal membranes between control and uterotone, exapar, involon treated groups was significant. However, there was no significant difference between control and replanta treatment group (Table 2).

The onset of first post partum estrus in control group was 64.00 $\pm$ 4.08 days. The onset of first post partum estrus in uterotone, exapar, replanta and involon treated group was 57.00 $\pm$ 3.61, 72.50 $\pm$ 6.83, 68.83 $\pm$ 6.24 and 64.33 $\pm$ 3.18 days respectively. Statistically there was no significant difference between control and treatment groups. The findings of present experiment confirm the findings of several workers that the herbal products like uterotone (Sharma *et al.*, 1972), exapar (Khanna *et al.*, 1997; Sharda *et al.*, 1998) involon (Sonwane, 1999; Tiwari, 1999) and replanta (Chakraborty and Paul, 1989) and can be used immediately after parturition for prevention of fetal membranes retention in buffaloes.

**Experiment III :** The time interval from 12 hrs after calvings i.e. after giving treatment to expulsion of fetal membranes in uterotone, exapar, replanta involon and control groups was

Table 1. Uterine contraction (*in vitro*) using different drugs

S.No.	Drugs	Doses	Number	Uterine contraction (mm)	
				Mean	± SE
1.	Oxytocin	i. $0.5 \times 10^{-2}$ IU/ml	6	16.00	±1.73
		ii. $1.0 \times 10^{-2}$ IU/ml	6	29.33	±3.08
		iii. $2.0 \times 10^{-2}$ IU/ml	6	49.83	±6.23
		iv. $4.0 \times 10^{-2}$ IU/ml	6	75.50	±3.31
2.	Uterotone	1 ml	6	23.67	±2.01
		2 ml	6	67.33	±3.11
		4 ml	6	114.67	±0.21
3.	Exapar	1 ml	6	37.33	±1.48
		2 ml	6	73.67	±1.28
		4 ml	6	86.33	±2.14
4.	Replanta**	1 ml	6	24.83	±3.41
		2 ml	6	56.33	±5.18
		4 ml	6	63.83	±5.18
5.	Involon***	2 ml	6	35.33	±1.47
		3 ml	6	72.33	±0.92
		4 ml	6	86.00	±2.63

\*\* (1 gm/5 ml in bath solution)

\*\*\* (1 gm/5 ml in bath solution)

Table 2. Effect of herbal preparations on expulsion of foetal membranes and onset of first post partum estrus

Groups	Prevention		Treatment	
	Time required for expulsion of fetal membranes (hrs)	Appearance of first post partum estrus (days)	Time required for expulsion of fetal membranes (hrs)	Appearance of first post partum estrus (days)
Control	7.33±0.74 (4 to 11)	64.00±4.08 (48 to 78)	8.37±1.41 (2 to 12)	63.66±5.20 (45 to 84)
Uterotone	5.29±0.48* (4 to 8)	57.00±3.61 (43 to 66)	6.00±0.68 (4 to 8)	57.33±4.19 (42 to 68)
Exapar	4.88±0.51* (3 to 7)	72.50±6.83 (54 to 100)	9.50±0.96 (6 to 12)	73.16±8.20 (52 to 104)
Replanta	5.56±0.65 (4 to 10)	68.83±6.24 (52 to 88)	9.66±0.66 (8 to 12)	69.33±6.87 (48 to 92)
Involon	4.71±0.52* (3 to 7)	64.33±3.18 (50 to 72)	7.33±0.71 (5 to 10)	68.83±3.62 (48 to 73)

\* Significant (P &lt; 0.05); Figures in parenthesis indicate range

6.00±0.68, 9.50±0.96, 9.66±0.66 and 7.33±0.71 and 8.37±1.41 hrs respectively (Table 2). Statistically there was no significant difference between control and treatment groups. Mean time interval from calving to onset of first post partum estrus in control group was 63.66±5.20 days. Onset of first post partum estrus in uterotone, exapar, replanta and involon treated group was 57.33±4.19, 73.16±8.20, 69.33±6.87, 63.83±3.62 days respectively (Table 2). Statistically there was no significant difference between control and treatment groups.

The findings of present experiment indicate that the use of above herbal products are not effective in treating the established cases of retained fetal membranes in buffaloes.

#### ACKNOWLEDGEMENT

Authors are thankful to the Dean, College of Veterinary Science and Animal Husbandry, Anjora, Durg (C.G.) for providing necessary facilities.

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