



Therapeutic management versus biological constituents of sub-oestrous crossbred cows of Asom

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ABSTRACT

The present work was undertaken to observe efficacy of some drugs for therapeutic management of sub-oestrous crossbred cows. A total of 60 confirmed sub-oestrous cows were considered. Cows were divided into 6 groups each group comprising 10 cows. Respective cyclic group of cows were treated with prostaglandin 500 µg on day 10, estradiol-valerate 30 mg on day 10 and at early oestrus, laboratory made mineral mixture (aauvetmin), aauvetmin plus multivitamins and a herbal drug stenot bolus. The mean time interval from last treatment to onset of induced oestrus was shortest (3.25 ± 0.25 days) with highest conception on first insemination after treating with cloprostenol injection. Oral administration of balanced mineral mixture along with multivitamins was also found to be cost effective. Regular monitoring of vulvar swelling might be useful from breeding point of view. The serum calcium, inorganic phosphorus, sodium, potassium, magnesium, cobalt, total protein were not much altered whereas, glucose level was dropped to lower level on the day of onset of oestrus. The level of cholesterol and alkaline phosphatase was elevated whereas when zinc concentration was increased, the iron and copper decreased on the day oestrus in all responded cows. Moreover, when 17β -estradiol in blood was reached highest concentration, progesterone concentration decreased to the lowest but not below 0.50 ng/ml, while cortisol was at higher level on the day of induced oestrus than at before treatment in all responded cows.

Key words: Biological constituents, Sub-oestrous crossbred cows, Therapeutic management

Sub-oestrus is one of the infertility problems affecting large population of cows in dairy herds though they cycle normally (Bhatt *et al.* 1993 and Bonia 2009). The both urban and rural dairy farmers frequently said that their most of the high yielding crossbred cows are suffering from this reproductive failure leading to a big economic loss mostly to new dairy entrepreneurs. The relative contribution of various physiological, genetic and reproductive management of sub-oestrus condition are to be looked after seriously (Connor *et al.* 2010). It appeared that minerals interaction is very essential for body function (Lucy 2001) and comparative studies on different means require to find out a suitable therapeutic management sub-oestrous cows. Keeping these in view the present experiment was carried out to study efficacy of administration of prostaglandin, oestrogen, minerals, vitamins and anti-stress drug as therapeutic management and changes of biological constituents in serum of crossbred cows of Asom suffering from sub-oestrus condition.

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MATERIALS AND METHODS

Post-partum crossbred cows (1000) from 115 dairy farms of 3 districts, viz. Kamrup, Darrang and Morigaon of Asom were investigated clinico-gynaecologically per rectum to determine the reproductive status based on genital changes to confirm sub-oestrous cows. Confirmed sub-oestrous cows (60) selected and divided in to 6 groups each comprising 10 cows. These were treated with different treatments regimen (Table 1). Cows of first to fifth lactation with milk yield ranging from 10–20 litres were considered for the study. Blood samples were collected from all cows just before treatment and on the day of induced oestrus for estimation of biological constituents like calcium, inorganic phosphorus, sodium, potassium, magnesium, total protein, cholesterol, glucose, alkaline phosphatase, iron, copper, zinc, 17β -estradiol, progesterone and cortisol by using standard methods of quality kits. Cobalt was determined by using atomic-absorption spectrophotometer.

RESULTS AND DISCUSSION

Out of 1000 randomly selected high yielding crossbred cattle 75 crossbred cows (7.50%) reported to be anoestrous were identified as sub-oestrus on the basis of presence of

Table 1. Response and fertility of sub-oestrous cows after administration of different treatments

Group	Treatment (starting day of treatment during the cycle)	Number of animals treated	Dose level	Route of administration	Number of responded cows (%)	Interval between last treatment and onset of oestrus mean $\pm$ S E (days)	Number of cows conceived
A	Cloprostenol inj. (day 10)	10	500 mg	Intra-muscularly	8 (80%)	3.25 $\pm$ 0.02	6
B	Estradiol valerate (day 10)	10	30 mg	Intra-muscularly	2 (20%)	6.00 $\pm$ 0.00	0
C	Estradiol valerate (at early oestrus)	10	30 mg	Intra-muscularly	2 (2%)	6.00 $\pm$ 0.00	0
D	Mineral mix.aauvetmin (ICAR NWP recommended) (day 10)	10	30 gm	Orally	2 (20%)	10.00 $\pm$ 0.00	2
E	Mineral mix.aauvetmin (ICAR NWP recommended) + supplevit-M (day 10)	10	30 gm	Orally	4 (40%)	9.75 $\pm$ 0.00	3
F	Stenot (day 10)	10	2 bolus	Orally	3 (30%)	9.75 $\pm$ 0.00	2

uterine tone with corpus luteum along with other genital changes on rectal palpation.

Effect of different treatment in sub-oestrous cows: Typical genital changes observed in vulva, vulvar mucous membrane, quantity of vulvar mucous, cervix, uterus, ovaries. Typical oestrus behaviours like mounting, bellowing, restlessness, reduced milk yield, mucous discharge and loss of appetite were the common sign of oestrus in responded cows under different treatment groups are similar report (Robert 1971, Arthur 1973, McDonald 1980).

The mean time interval from last treatment to onset of induced oestrus was shortest in prostaglandin treated group with highest conception rate. Similarly, prostaglandin or its analogue like cloprostenol was used effectively in sub-oestrus crossbred cows with effective luteolysis of corpora lutea with decreasing circulatory progesterone allowing resumption of gonadotrophic stimulation and follicular maturation so that responding females shown oestrus signs within 72 to 96 h after treatment and conception (Barr 1977, Nair and Madhavan 1984, Ajitkumar *et al.* 1995 and Deshpande *et al.* 2008) whereas, lower conception in dairy cattle (Mane *et al.* 2010). Although administration estradiol-valerate intra-muscularly on day 10 of the sub-oestrous cycle or at early oestrus able to induce heat only but failed to conceive as was not effective as said (Arthur 1973). The oral feeding of the combination of Aauvetmin and Supplevit-M was comparatively better for fertility when Aauvetmin alone or Stenot proved to be inferior. Supplementation with complexed trace minerals elements like cobalt, copper, manganese and zinc improved fertility of post-partum dairy cows (Campbell *et al.* 1999). However, administration of balanced mineral mixture along with vitamins was cost effective treatment of sub-oestrus condition. It is also suggestive to dairy farmers for regular monitoring of vulvar swelling might be useful for sub-oestrus detection and subsequent breeding.

Biological constituents in serum of sub-oestrous cows responding to different treatments viz. variation of biological constituents in serum of sub-oestrus cows responded to different treatments are presented in Table 1. After administration of cloprostenol, no much difference serum concentration of all biochemical constituents but glucose, cholesterol, alkaline phosphatase, 17 β - estradiol and cortisol were comparatively higher on day of induced oestrus except progesterone of responded sub-oestrous cows of the present study. Calcium plays an important role in gonadotropic regulation of ovarian steroidogenesis (Carnegie and Tsang 1984) where marginal deficiency of phosphorus causing disturbance in the pituitary-ovarian-axis including ovulation (Bhaskaran and Abdullakhan 1981). In present study lower level of sodium in blood estimated (Roussel *et al.* 1983, Small *et al.* 1997) observed nonsignificant difference of serum potassium with significantly ($P < 0.05$) higher sodium in cattle. No variation of serum magnesium level in sub-oestrous cows on the day of induced oestrus was found. Although magnesium did not appeared to have any direct role in reproduction but enzymes, which influence absorption of calcium and inorganic phosphorus and therefore, magnesium imbalance may influence reproductive efficiency indirectly (Sharma *et al.* 2004). The serum protein concentration before treatment and on the day of onset of oestrus were found to be almost stable and this finding was in agreement with McLennan and Willoughby (1973) who stated that the serum total protein had little contribution during oestrus of cows. Comparatively lower mean serum glucose concentration was observed on the day of onset of induced oestrus treated with different experimental drugs, when similar lower glucose during oestrus was reported by Singh and Dutt (1974). Ahlwat and Derashri (2009) found significantly higher glucose in cyclic cows. The increases of ovulation rate by high-energy inputs would be caused by an enhancement in the developmental competence of preovulatory follicles (Letelier

et al. 2008). Clifton and Meyer (1956) found that injection of oestrogen depressed the appetite of rats and Wynn and Doar (1969) had also reported a decrease in blood glucose in women receiving oestrogen therapy. Letelier *et al.* (2008) observed lower glucose level on the day of induced oestrus in all responding treated sub-oestrous cows and glucogenic supply increased ovulation rate by modifying follicle

recruitment and subsequent development of pre- ovulatory follicles without effecting ghrelin secretion. Higher serum cholesterol concentration on the day of induced oestrus of responded cows were similar to reports (Dhoble *et al.* 2004, Ahlwat and Derashri 2009). The present higher mean serum alkaline phosphatase values on the day onset of oestrus of the experimental cows were might be indicative of cellular

Table 2. Mean levels of different biochemical constituents of blood before treatment and on the day of induced oestrus

Biochemical constituents	Group A		Group B		Group C	
	Before treatment	At induced oestrus	Before treatment	At induced oestrus	Before treatment	At induced oestrus
Calcium (mg%)	10.23±0.43 (8)	10.92±0.57 (8)	9.48 (2)	9.77 (2)	10.43 (2)	11.09 (2)
Inorganic phosphorus (mg%)	5.90±22 (8)	5.90±0.26 (8)	5.80 (2)	5.70 (2)	5.97 (2)	6.60 (2)
Sodium (mmol)	159.67±6.31 (8)	149.66±3.22 (8)	157.49 (2)	149.12 (2)	146.15 (2)	147.82 (2)
Potassium (mmol)	5.57±0.37 (8)	5.45±0.32 (8)	5.66 (2)	5.44 (2)	5.55 (2)	5.58 (2)
Magnesium (mg/dl)	3.60±0.54 (8)	3.41±0.37 (8)	3.24 (2)	4.38 (2)	3.44 (2)	3.51 (2)
Total protein (g/dl)	8.15±0.29 (8)	9.41±0.38 (8)	8.82 (2)	9.61 (2)	8.50 (2)	9.29 (2)
Glucose (mg/dl)	62.11±3.34 (8)	59.94±4.08 (8)	63.89 (2)	59.11 (2)	56.86 (2)	56.06 (2)
Cholesterol (mg/dl)	97.60±8.75 (8)	105.74±9.66 (8)	87.59 (2)	104.58 (2)	100.97 (2)	101.01 (2)
Alkaline phosphatase (IU/ml)	342.83±12.89 (8)	402.70±15.63 (8)	349.44 (2)	398.46 (2)	394.90 (2)	398.75 (2)
Iron (mg/dl)	199.47±8.75 (8)	289.88±9.66 (8)	199.55 (2)	288.86 (2)	245.10 (2)	245.55 (2)
Copper (mg/dl)	88.14±7.46 (8)	71.17±5.66 (8)	84.46 (2)	63.32 (2)	61.90 (2)	62.18 (2)
Zinc (mg/dl)	136.38±7.46 (8)	141.37±5.66 (8)	135.79 (2)	142.99 (2)	142.50 (2)	142.56 (2)
Cobalt (ppm)	0.40±0.05 (8)	0.43±0.03 (8)	0.38 (2)	0.45 (2)	0.45 (2)	0.45 (2)
17 β -estradiol (pg/ml)	13.63±1.44 (8)	42.68±2.22 (8)	14.75 (2)	46.35 (2)	39.48 (2)	39.59 (2)
Progesterone (ng/ml)	4.67±0.56 (8)	0.51±0.05 (8)	5.75 (2)	0.52 (2)	0.49 (2)	0.50 (2)
Corstisol (nM/L)	22.28±1.43 (8)	26.13±0.65 (8)	24.00 (2)	29.50 (2)	26.00 (2)	29.00 (2)

Figures within parentheses indicated number of observation.

Table 3. Mean levels of different biochemical constituents of blood before treatment and on the day of induced oestrus

Biochemical constituents	Group A		Group B		Group C	
	Before treatment	At induced oestrus	Before treatment	At induced oestrus	Before treatment	At induced oestrus
Calcium (mg%)	10.10 (2)	10.25 (2)	11.25±0.51 (4)	11.44±0.47 (4)	10.38±0.73 (3)	11.49±0.59 (3)
Inorganic phosphorus (mg%)	5.91 (2)	6.48 (2)	5.67±0.22 (4)	6.24±0.26 (4)	5.58±0.34 (3)	5.89±0.37 (3)
Sodium (mmol)	158.60 (2)	147.10 (2)	152.31±6.47 (4)	153.88±5.76 (4)	157.25±5.76 (3)	146.35±4.89 (3)
Potassium (mmol)	5.49 (2)	5.72 (2)	4.72±0.45 (4)	5.25±0.15 (4)	6.03±0.24 (3)	5.99±0.31 (3)
Magnesium (mg/dl)	3.60±0.54 (8)	3.41±0.37 (8)	3.24 (2)	4.38 (2)	3.44 (2)	3.51 (2)
Total protein (g/dl)	9.09 (2)	10.75 (2)	8.79±0.45 (4)	10.16±0.43 (4)	9.07±0.45 (3)	10.45±0.22 (3)
Glucose (mg/dl)	62.49 (2)	60.09 (2)	63.28±5.50 (4)	50.87±3.75 (4)	65.42±3.45 (3)	60.80±3.89 (3)
Cholesterol (mg/dl)	97.01 (2)	105.18 (2)	97.69±5.55 (4)	115.94±4.66 (4)	93.87±6.21 (3)	118.38±5.32 (3)
Alkaline phosphatase (IU/ml)	339.69 (2)	390.63 (2)	314.67±14.34 (4)	420.87±13.46 (4)	343.05±10.29 (3)	393.81±9.52 (3)
Iron (mg/dl)	199.47±8.75 (8)	289.88±9.66 (8)	199.55 (2)	288.86 (2)	245.10 (2)	245.55 (2)
Copper (mg/dl)	88.14±7.46 (8)	71.17±5.66 (8)	84.46 (2)	63.32 (2)	61.90 (2)	62.18 (2)
Zinc (mg/dl)	136.38±7.46 (8)	141.37±5.66 (8)	135.79 (2)	142.99 (2)	142.50 (2)	142.56 (2)
Cobalt (ppm)	0.40±0.05 (8)	0.43±0.03 (8)	0.38 (2)	0.45 (2)	0.45 (2)	0.45 (2)
17 β -estradiol (pg/ml)	16.75 (2)	49.63 (2)	15.53±2.23 (4)	43.56±2.46 (4)	16.25±2.72 (3)	45.60±2.58 (3)
Progesterone (ng/ml)	3.55 (2)	0.58 (2)	4.04±0.62 (4)	0.62±0.06 (4)	3.09±0.45 (3)	0.52±0.04 (3)
Corstisol (nM/L)	23.50 (2)	26.50 (2)	22.50±2.35 (4)	25.25±1.11 (4)	23.00±1.22 (3)	28.00±1.45 (3)

Figures within parentheses indicated number of observation.

changes in female genitalia during the oestrus period (Rowland 1977, Mc Donald 1980). It was evident from the study that when iron concentration was higher, the copper concentration lowered down in the induced oestrus of all responded cows and this trend of variation was well supported by Singh (2007). Small *et al.* (1997) observed nonsignificant difference of serum iron and zinc concentration with significantly ($P < 0.05$) higher copper in nulliparous cattle. The mean blood levels of cobalt at before treatment and on the day of induced oestrus of sub-oestrous cows remained static. Campbell *et al.* (1999) reported that supplementation trace elements like cobalt, copper, manganese and zinc improved reproduction with production of dairy cows but cobalt injection did not alleviate condition rather vitamin B₁₂ injection cured symptoms of this vitamins though cobalt associated with synthesis of this vitamin in ruminants (Ranjan 1977). Hence, it was opined that minerals interact with each other in the body and many interactions can result in mineral elements tying up or making other elements available or unavailable for essential body functions (Parish 2008).

17 β -estradiol, progesterone and cortisol level in serum of sub-oestrous cows treated with different drugs: The result indicated that 17 β -estradiol and cortisol levels were highest with lowest concentration of progesterone level on the day of induced oestrus. The result indicated that 17 β -estradiol and cortisol levels were highest with lowest concentration of progesterone level on the day of induced oestrous like that of normal cyclic cows. As similar to lower progesterone on the day of induced oestrus of present study, Kang *et al.* (1994) measured significantly lower blood progesterone concentration at initial period of silent heat. But when Zdunczyk *et al.* (1994) used cloprostenol i/m on 10th day cycle of sub-oestrous cows came into oestrus after treatment along with decreased progesterone to < 3.18 nmol/litre. Tasi and Wiltblank (1998) opined that prostaglandin distinctly regulated physiological changes in early and mid-cycle bovine corpora lutea. Wynn and Doar (1969) pointed out a decrease in blood glucose in women receiving oestrogen therapy. They were unable to conclude whether this change resulted from altered rates of production and/or removal of glucose. The mean blood levels of cortisol before treatment and on the day of induced oestrus revealed that this hormone indicating the oestrus as a stress condition where cortisol was at elevated level. Adeymo *et al.* (1981) reported that low cortisol in plasma might had reflected adaptation to the hot climate and mechanism to prevent high metabolic heat generated. It was cited that cortisol under normal conditions did not involve in the regulation of cycle (Rao and Pandey 2005, Bonia 2009).

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