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Research Article

EFFECT OF PARENTERAL ADMINISTRATION OF VITAMIN E AND SELENIUM DURING PERIPARTURIENT PERIOD ON CORTISOL PROFILE IN SURTI BUFFALOES

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ABSTRACT

A study was conducted on Surti buffaloes during their transient period. The animals were divided into two groups comprising of ten animals in each group as: Treatment group of Surti buffaloes treated with Inj. Vitamin E and Selenium at the dose rate of 10 ml on 60th, 45th, 30th and 15th day before expected date of parturition and after parturition on 15th, 30th day intramuscular (IM) and Control group of Surti buffaloes given 10 ml of Inj. Normal saline solution (IM). Blood samples were collected on same days before injection as well as on the day of parturition, 45 and 60 days postpartum in serum clotting vacutainer. The mean serum cortisol concentration did not differ significantly at any of the intervals between treatment and control groups. The mean serum cortisol concentration was also non-significantly ($p>0.05$) different period-wise between pregnant and non-pregnant groups, but the overall pooled mean level was significantly ($p<0.05$) lower in the pregnant group as compared to non-pregnant groups of buffaloes.

Keywords: Cortisol, Periparturient, Selenium, Surti buffalo, Vitamin E

The transition or periparturient period, from 3 weeks before to 3 weeks after parturition, is a stressful time for dairy cows (Drackley, 1999). During the transition period, immunosuppression commonly occurs and cows exhibit great susceptibility to a number of diseases (Mallard *et al.*, 1998). Vitamin E is an important antioxidant that has been shown to play an important role in immuno responsiveness and health in dairy cows (Weiss and Spears, 2006). In Vitamin E and Selenium deficiency condition, free radicals accumulate and not only damage cell membranes, but also disrupt several processes linked to the synthesis of steroids (Seagerson and Libby, 1982). It is not surprising therefore that negative impacts of Vitamin E and Selenium deficiencies have been observed on various components of the reproductive events. Estimation of cortisol is a well-accepted indicator for stress in animals and men and its estimation gives idea by throwing the light on its level during periparturient period.

MATERIALS AND METHODS

The present research work was undertaken on twenty (20) Surti buffaloes during their transient period i.e. two month before their expected date of parturition to two month after parturition, divided into treatment (n=10) & control (n=10)) groups, at Livestock Research Station, Navsari Agricultural University, Navsari, Gujarat. The animals were fed green fodder, hay and compounded concentrate, as per the standard feeding schedule followed on the farm. The animals had free access to drinking water. The animals were also washed and sprinkled with water twice daily or were allowed to wallow in the pond during hot noon hours of summer season to reduce heat stress. The treatment protocol is given in Table 1.

Pregnancy diagnosis was carried out per rectally at 90 days post breeding. Again the two groups were made from all 20 animals irrespective of treatment and control group on the base of its conception in pregnant (n=13) and non-pregnant (n=7) groups.

Blood samples were collected from all those selected animals aseptically by jugular vein puncture in serum clotting vaccutainers. The vaccutainers were kept in

Table 1
Treatment protocol

Group of Animals	Treatment Schedule/Protocol (n=10)	Dose & Route	Blood collection schedule
Treatment	Inj. Vitamin E and Selenium (E-CARE Se*) was Injected on day 60, 45, 30 and 15 before expected date of parturition and after parturition on day 15, 30.	10 ml (500 mg Vit. E & 15 mg Selenium), I/M	On day 60, 45, 30, 15 before the expected date of parturition, on the day of parturition and 15, 30, 45 and 60 day after parturition
Control	Inj. Normal saline was Injected on day 60, 45, 30 and 15 before expected date of parturition and after parturition on day 15, 30.	10 ml Normal saline, I/M	

*DL-α Tocopheryl Acetate I.P. equivalent to Tocopherol (Vitamin E) Base -50mg, Sodium Selenite U.S.P. equivalent to Selenium Base -1.5mg in each ml: Inj. E-CARE Se 10 ml vial, Provimi Animal Nutrition India Pvt. Ltd. Bangalore, India.

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slanting position at room temperature for 1-2 hours. Finally, serum was separated by centrifugation at 3000 rpm for 5 minutes and stored in 4.5 ml plastic storage vial and stored at –20 °C in deep freezer until analysis. Serum cortisol concentration was measured by standard Enzyme Linked Immuno Sorbent Assay technique using assay kits (Intra Assay Variation: ≤ 9.0 %; Inter Assay Variation: ≤ 9.8%) and procedure described by Product No. DNOV001, Nova Tec Immundiagnostica GmbH Technologie & Waldpark, Germany. The tests of significance for pregnant vs. non-pregnant and treatment vs. control groups were made by Standard Student's paired 't' test. The fortnight-wise variation within the group was tested for each trait by using completely randomized design as well as the mean differences between and within the groups were tested using Duncan's New Multiple Range Test (DNMRT) at 1 per cent and 5 per cent level of significance.

RESULTS AND DISCUSSION

The mean serum cortisol concentration was observed non-significantly differing between treatment and control groups at different peripartum intervals. The mean serum cortisol concentration was non-significantly higher at 60th day and non-significantly lower at 45th day, 30th day and 15th day before parturition and on the day of calving as well as 15th day, 30th day but mean serum cortisol concentration was observed non-significantly higher at 45th day and 60th day after parturition in treatment group as compared to control group, respectively (Table 2).

The prepartum mean serum cortisol levels were found non-significantly lower when compared between prepartum and postpartum at various intervals, viz., at 60, 45, 30 day prepartum than that of 60, 45, 30 day postpartum, respectively and non-significantly higher at 15th day prepartum (119.477±9.165 ng/ml) than that of 15th day postpartum (116.152±10.141 ng/ml) in the treatment group. Whereas, in control group, prepartum values were lower when compared between prepartum and postpartum at different time intervals in Surti buffaloes (Table 2).

The pooled mean serum cortisol concentration was found to be significantly ($p<0.05$) increased from 60th day to 45th day, 30th day, 15th day prepartum and on the day of calving and there after significantly ($p<0.05$) decreased at 15th day, 30th day and non-significantly ($p>0.05$) at 45th day and 60th day postpartum. Similar were the trend for treatment and control groups (Table 2).

The mean serum cortisol concentrations were found non-significantly lower in pregnant than non-pregnant group at different intervals. Moreover, overall prepartum mean serum cortisol levels in the present study were found non-significantly lower as compared to postpartum levels in pregnant (90.201±3.641 vs. 95.653±3.492 ng/ml) and non-pregnant (98.720±4.974 vs. 105.851±6.035 ng/ml) group (Table 2).

Similarly, the overall mean serum cortisol values in the pregnant group were found non-significantly lower as compared to non-pregnant group at prepartum and postpartum phase, and hence in the overall pooled means. The mean serum cortisol level was found non-significantly lower on the day of calving in pregnant group as compared to non-pregnant group (Table 2).

The mean serum cortisol level gradually increased in trend from 60th day prepartum to significantly ($p<0.05$) on the day of calving and it was gradually decreased in trend from day of parturition to non-significantly ($p>0.05$) at 15th day and significantly ($p<0.05$) and non-significantly ($p>0.05$) at 30th day in the pregnant and non-pregnant group, respectively and there after non-significantly ($p>0.05$) decreased at 45th day and 60th day postpartum in pregnant and non-pregnant groups of Surti buffaloes, respectively (Table 2).

The present results showed that cortisol concentration during pregnancy reached maximum level at parturition and thereafter declined significantly ($p<0.05$) at 15th day postpartum followed by insignificant fall in the treatment group and significant fall ($p<0.05$) in the control group up to 60th day postpartum, and lower levels of cortisol were observed in the vit-E Selenium treated group as compared to control group.

The results of present study were in accordance with the findings of Kulberg *et al.* (2002), who reported that serum cortisol level increased as parturition approached, with peak level at parturition and slightly decreasing level were found postpartum.

Similarly, Djokovic *et al.* (2014) found serum cortisol concentrations insignificantly increased to a peak (108.58±69.55 nmol/l) before calving, thereafter dropped (34.59±31.50 nmol/l) to an approximately steady level in clinically healthy puerperal cows; Teama and Gad (2014) found that the prepartum period had a greater mean of cortisol and decrease in serum cortisol sustain from time immediately following parturition until the end of experiment in cows, which was also in agreement with our findings.

Gupta *et al.* (2005) also recorded significantly ($p<0.05$) higher cortisol on the day of parturition, the cortisol level gradually increased from 21st day before calving to the day of calving in vitamin E and selenium treated and control group. Further, they reported lower

Table 2

Mean serum Cortisol levels (ng/ml) at different fortnightly intervals peripartum in antioxidant treated and control groups as well as pregnant and non-pregnant groups of Surti buffaloes (Mean±SEM)

Peripartum Phases	Days	Cortisol ng/ml						
		Treatment (n=10)	Control (n=10)	't'-Value	Pooled(n=20)	Pregnant(n=13)	Non-pregnant(n=7)	't'-value
Prepartum	60	68.496±4.169^{a}	65.711±2.980^{a}	0.543	67.104± 2.514^{a}	64.216±3.357^{a}	72.466±2.825^{a}	1.632
	45	82.109±3.198^{ab}	85.230±3.834^{b}	0.625	83.670± 2.456^{b}	82.290±3.734^{bc}	86.231±1.060^{ab}	0.757
	30	100.305±6.379^{bcd}	100.526±5.141^{cd}	0.027	100.416± 3.987^{c}	99.472±5.935^{cde}	102.169±3.499^{b}	0.315
	15	119.477±9.165^{de}	123.606±6.883^{e}	0.360	121.542± 5.598^{d}	114.826±6.767^{ef}	134.013±8.580^{cd}	1.716
	Overall	92.597±4.269	93.768±4.139	0.197	93.183±2.955	90.201±3.641	98.720±4.974	1.383
Day of Parturition	0	135.109±12.570^{f}	144.618±6.116^{f}	0.680	139.864± 6.890^{e}	133.146±8.725^{g}	152.339±10.338^{d}	1.358
Postpartum	15	116.152±10.141^{cde}	128.272±5.882^{e}	1.034	122.212±5.872^{d}	118.129±7.649^{fg}	129.794±8.943^{cd}	0.945
	30	102.322±10.518^{bcd}	104.871±3.967^{d}	0.227	103.597± 5.479^{c}	100.537±4.654^{de}	109.279±13.522^{bc}	0.752
	45	91.013±10.994^{abc}	89.594±3.227^{bc}	0.124	90.304± 5.579^{bc}	86.830±5.373^{bcd}	96.754±12.794^{ab}	0.842
	60	81.365±7.445^{ab}	80.192±3.471^{b}	0.143	80.779± 4.000^{ab}	77.118±4.368^{ab}	87.577±7.875^{ab}	1.267
	Overall	97.713±5.177	100.732±3.560	0.481	99.223± 3.126	95.653±3.492	105.851±6.035	1.570
Overall	't'-Value	0.762	1.276	—	1.404	1.081	0.912	—
	P-Value	0.448	0.206	—	0.162	0.282	0.366	—
	Pooled	99.594±3.519	102.513± 2.972	0.634	101.054±2.299	97.396±2.699$_{y}$	107.847±4.142$_{x}$	-2.191*

Means bearing different superscripts (a,b,c) within a column (between phase intervals) differ significantly ($p<0.05$).Means bearing different subscripts (x,y,z) within a row (between groups) differ significantly ($p<0.05$).

cortisol level in vitamin E and selenium treated group as compared to control group at 21st, 7th, 3rd, 2nd, 1st day before calving and on the day of parturition, support the present findings. Sathya *et al.* (2007) also suggested supplementation with antioxidants like vitamin E and Se to reduce oxidative stress in the immediate postpartum period in the group of dystocia-affected buffaloes based on lower level of cortisol seen in the treated group of affected buffaloes.

On the contrary, Chaiyabutr *et al.* (2000) reported non-significantly higher 29.9±24.9 ng/ml cortisol level in early lactation as compared to 23.5±14.5 ng/ml in late pregnancy in crossbred cows. Ahmed *et al.* (2013) recorded the lowest concentrations of cortisol on 1st and 6th day after parturition in Holstein cows.

Cortisol was stated to regulate the dynamics of ovulatory follicles (Acosta *et al.* 2005) and have a positive influence on the luteal function during estrous or pregnancy (Myers *et al.* 2007). Therefore, the present study could support the previous studies for the importance of cortisol in the maintenance and function of the luteal life span, and that cholesterol is relevant indicator of luteal function during the puerperium period in cattle which requires more research. Moreover, these findings might also be supported by our observations in which we noted higher level of cortisol at 45th and 60th day postpartum in vitamin E and selenium treated group with resumption of ovarian activity as compared to control group of animals.

In general, the highest blood cortisol level found in Surti buffaloes immediately before calving suggested that buffalo-cows undergo severe stress immediately before and during calving, which leads to hormonal changes manifested as significant ($p<0.05$) increase in glucocorticosteroid (cortisol) level in the circulation.

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