Indian Journal of Animal Sciences **84** (5): 507–510, May 2014/Article

Effect of vitamin E and zinc supplementation on liver enzymatic profile of pre- and post-partum Sahiwal cows

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Received: 9 April 2012; Accepted: 18 January 2014

ABSTRACT

The study was conducted to elucidate the changes in circulatory plasma levels of hepatic enzymes in vitamin E and zinc supplemented periparturient Sahiwal cows. Sahiwal cows (18) were selected from the institute herd, apparently healthy and in advanced state of pregnancy. The cows were randomly divided into 3 groups 6 cows in each, viz. control, treatment 1 (T1) and treatment 2 (T2). Control group was fed a control diet as practiced for pregnant cows in NDRI dairy farm. The treatment 1 (T1) group was supplemented with zinc @ 60 ppm/day/cow and treatment 2 (T2) group was supplemented a mixture of vitamin E @ 1000 IU/day/animal and zinc @ 60 ppm/day/cow with control diet during day 60 prepartum to day 90 postpartum. Plasma was analyzed for alkalline phosphatase (ALP), alanine aminotransferase (ALT) and aspartate aminotransferase (AST). ALP levels were differing significantly in all 3 groups. ALT level was significantly lower in treatment 2 as comparison to its level of both control and treatment 1 animals. AST level was also significantly lower in treatment 2 as comparison to its level of both control and treatment 1 animals. The study indicated that vitamin E and zinc supplementation lowers these hepatic enzymes in blood plasma, prevents oxidative damage of liver and improves the health condition of liver.

Key words: Alanine aminotransferase, Alkaline phosphatase, Aspartate aminotransferase, Sahiwal cow

Pregnancy and early lactation are stressful stages accompanied with increased metabolic activities and energy demand. Physiological changes during this period impose considerable challenge to homeostasis by altering normal metabolism and production of stressors resulting in metabolic disorders (Valocky *et al*. 2007). Alanine aminotransferase (ALT) and aspartate aminotransferase (AST) increase during pregnancy and early lactation. AST and ALT are normally found in cells, liver, heart, muscle tissue, pancreas and kidneys. Low levels of AST are normally found in the blood. Increase in plasma AST is associated with cell necrosis of liver and skeletal or cardiac muscle, starvation and lacking vitamin E. The amount of AST in the blood is directly related to the extent of the tissue damage. Aspartate aminotransferases enzymes are found in liver, striated and cardiac muscle, and is a good marker of soft tissue damage (Otto *et al*. 2000, Abutarbush and Radostits 2003). ALT, found mainly in liver and in smaller amounts in kidneys, heart, muscles, pancreas, is normally present in blood in low levels. But on liver damage it is released into the bloodstream, increasing its levels. Plasma alanine aminotransferase is a good marker for an acute hepatic damage. Alkaline phosphatase (ALP) belongs to hydrolases that cause hydrolysis of monoesters of phosphoric acid (Sato *et al*. 2005, Mohri *et al*. 2007). ALP is localized mainly in the cellular membrane of hepatocytes (Valocky *et al*. 2007). Shinde *et al*. (2009) and Berg *et al*. (2006) reported high levels of bone ALP in growing calves and in late pregnancy.

The plasma α-tocopherol and zinc level decreases around calving (Rajiv 2001, Chandra and Aggarwal 2009). ALP, ALT and AST levels increase because chances of liver damage and metabolic disorders increased during this period. Vitamin E and zinc decrease elevation in the plasma AST and ALT activities by decreasing hepatocellular damage (Naziroglu *et al*. 1999). However, very little information is available on effect of vitamin E and zinc supplementation on hepatic enzymes profile in pre- and post-partum Sahiwal cows. Therefore, the present study was designed to explore the effect of vitamin E and zinc supplementation on liver enzyme profiles (ALP, ALT and AST) during peripartum in Sahiwal cows.

MATERIALS AND METHODS

The experiment was conducted on apparently healthy and in advanced state of pregnancy 18 Sahiwal cows (415±12.56 kg body weight), selected from NDRI herd.

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The cows were randomly divided into 3 groups of 6 cows in each, viz. control, treatment 1 (T1) and treatment 2 (T2). Cows of all 3 groups were in nearly similar parity (2.8±1.10). Control group was fed a control diet as practiced for pregnant cows in NDRI dairy farm. The treatment 1 (T1) group was supplemented with zinc @ 60 ppm/day/cow and treatment 2 (T2) group was supplemented a mixture of vitamin E @ 1,000 IU/day/animal and zinc @ 60ppm/day/cow with control diet during day 60 prepartum to day 90 postpartum. All animals were kept in loose housing system as practised in NDRI dairy farm.

Blood samples from animals were collected by jugular venipuncture on –60, -45, –30, -15, –7, –3, 0, 3, 7, 15, 30, 45, 60, 90 and 120 day in relation to expected date of calving with use of vacutainer tubes containing heparin as an anticoagulating agent at 7.30 AM. Samples were brought to the laboratory in chilled iceboxes soon after collection and centrifuge at 1200×g at 4°C for 20 min to separate the plasma from packed erythrocytes. Plasma samples were stored at —20°C in different aliquots for the estimation of plasma alkaline phosphatase (ALP), plasma alanine aminotransferase (ALT) and plasma aspartate aminotransferase (AST) activities.

Plasma ALP activity was estimated as per Kind and King (1954) and ALT and AST activities in blood plasma according to Reitman and Frankel (1957).

Data for all measured variables were analyzed as repeated measures using the MIXED procedure of SPSS version 19. The model included the main effects of vitamin E and zinc treatment (groups), days around calving and their interactions.

RESULTS AND DISCUSSION

The ALP level was higher in prepartum than its levels in postpartum period in all groups but its levels were lowest in T2 in comparison to T1 and control cows (Table 1). At day –15, –7 and –3 (prepartum) ALP levels differed significantly (P<0.05) in all 3 groups (Table 1). Plasma ALP level was observed significantly lower (P<0.01) in treatment 2 (T2) in comparison to treatment 1 (T1) and control group (Fig. 1).

In present experiment ALP levels increased during pregnancy, which is supported by Sato *et al*. (2005). An increase in ALP activity is directly proportional to the degree of damage suffered by an organ (Kawashima *et al*. 2007). In present experiment concentration of ALP was lower in treatment than that in control group; it is in agreement with Obianime *et al*. (2010).

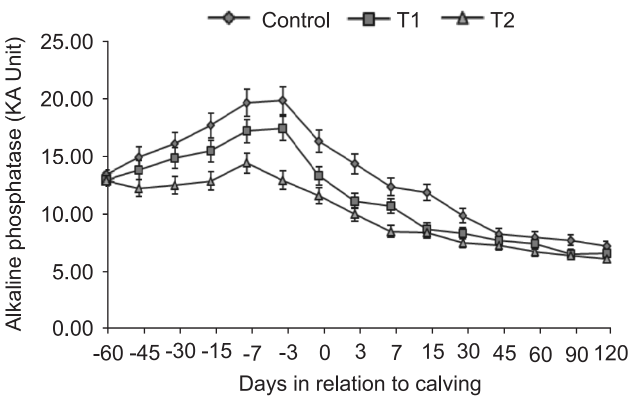

Fig. 1. Changes in plasma alkaline phosphatase (ALP) activity in pre- and post-partum Sahiwal cow treated with vitamin E and zinc.

ALT level began to increase from last 3 days of pregnancy and reached maximum at the day 3 postpartum in relation to calving, which was maximum (P<0.05) in control than T1 and T2 (Table 1). ALT levels in treatment groups were lower than its level on all the days of pre- and post-partum of control group cows (Table 1). Overall value of ALT (Fig. 2) was significantly lower (P<0.05) in T2 than that in T1 and control group.

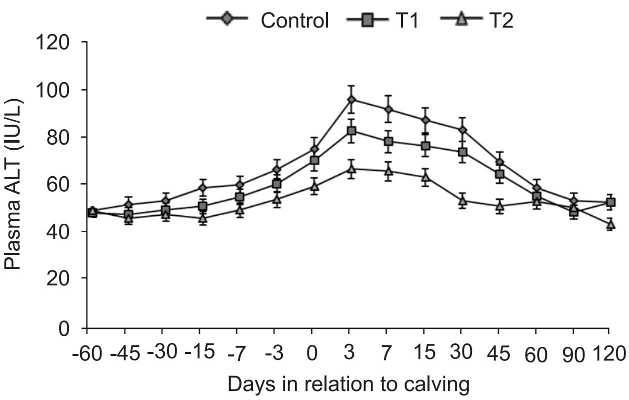

Fig. 2. Changes in plasma alanine aminotransferase (ALT) activity in pre and postpartum Sahiwal cow treated with vitamin E and zinc.

AST differed significantly (P<0.01) in groups (Table 1). In T2, days 0 (calving), 3 and 7 (postpartum) plasma AST levels were significantly (P<0.05) lower than corresponding values of both control and T1 animals (Table 1). Overall value of ALT was significantly lower in T2 than that in both control and T1 animals. AST levels are presented in Fig. 3.

Table 1. Overall mean of various parameters along with P-values in Sahiwal cows treated with vitamin E and zinc

Parameter	Mean±SEM			SEM	P-values		
	Control	T1	T2		Treatment	Day	Treatment x Day
ALP (KA Unit)	13.26	11.57	10.12	0.66	0.010	<0.001	0.015
ALT (IU/L)	68.62	62.30	54.39	3.05	<0.001	0.010	0.010
AST (IU/L)	278.19	248.78	229.11	9.96	<0.010	<0.001	<0.001

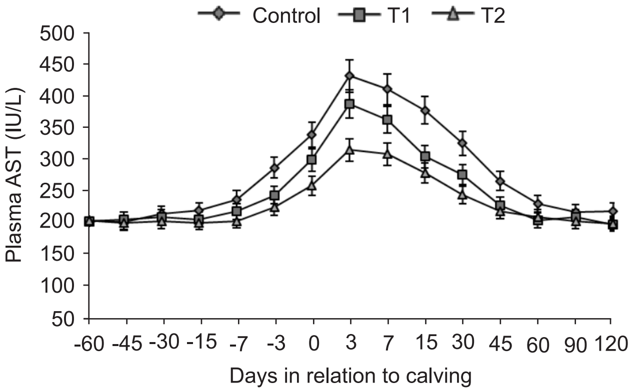

Fig. 3. Changes in plasma aspartate aminotransferase (AST) activity in pre- and post-partum Sahiwal cow treated with vitamin E and zinc.

The present study confirmed that during pregnancy, there are changes in plasma AST, ALP and ALP which are normally associated with an increased risk of liver disease. Numerous publications reported on the specific causes of abnormal liver function test in pregnancy (Castro *et al*. 1999). Iribhogbe *et al*. (2011) showed that vitamin E and its combination with zinc, both supplementations successfully revert the hyper-hepatic state of pregnancy.

Increased liver enzyme (ALT and AST) activities in cow during transition period in this study were in accord with the recorded association to the fatty liver observed in cows during this period (Semacan and Sevinc 2005). The activities of plasma ALT and AST are commonly used as indicators of liver cell damage and death. Concentration of ALT and AST were found higher in postpartum period than that in prepartum. During postpartum, metabolic activity and oxidative stress were higher in comparison to prepartum. So chances of liver damage are more during this period resulting in increase in the concentration of ALT and AST (Rezaeisaber and Nazer 2011) as observed in our experiment. Measurement of liver enzymes in serum is useful for evaluating fatty liver disease (Rezaeisaber and Nazer 2011). Mild to moderate fatty liver might result in a liver dysfunction without hepatocytes destruction and a subsequent increase in liver enzyme activities (Bulent *et al*. 2006)

Vitamin E is a protective antioxidant (Stohs *et al*. 2001, Das and King 2007); and inhibits peroxidation, mop up free oxygen radicals and disorganize and break of peroxidation chain reactions by an inhibition of reactive oxygen species (Das and King 2007). Vitamin E supplementation lowers ALT and AST around the parturition (Gaafar *et al*. 2011) as reported in our results.

In conclusions, vitamin E supplementation lowers the concentration of the ALP, ALT and AST because it prevents the metabolic and oxidative stress by neutralizing the reactive oxygen species during peripartum and improves the condition of liver.

ACKNOWLEDGEMENTS

The authors thank Dr A K Srivastava, Director, National Dairy Research Institute, Karnal, Haryana (ICAR, New Delhi) for providing necessary facilities for successful completion of the research.

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