Serum Alkaline Phosphatase and Amylase Activities in Subacute Ruminal Acidosis in Dairy Cows

J. Tajik and S. Tahvili
Department of Clinical Sciences, School of Veterinary Medicine, Shiraz University, Shiraz, Iran

Corresponding Author: J. Tajik, Department of Clinical Sciences, School of Veterinary Medicine, Shiraz University, Shiraz, Iran Tel: 00987116138810, 00989171897203

ABSTRACT
Ruminal fluid and blood samples were drawn from 159 dairy cattle (75 early lactation and 84 mid lactation cows from 10 dairy herds) to compare blood serum alkaline phosphatase and amylase activities in subacute ruminal acidosis. No significant differences were detected for blood alkaline phosphatase and amylase activities in subacute ruminal acidosis affected and the rest of the cows. The differences between animals with subacute ruminal acidosis and those with marginal pH values and healthy cows were not significant. The blood alkaline phosphatase and amylase activities had no significant correlations with ruminal pH.

Key words: Subacute ruminal acidosis, dairy cattle, serum alkaline phosphatase, amylase

INTRODUCTION
Subacute ruminal acidosis (SARA) is characterized by daily episodes of low ruminal pH between 5.5 and 5.0 (Krause and Oetzel, 2005). This digestive disorder is the consequence of feeding high grain diets to dairy cows, which are adapted to forage diets (Oetzel, 2003; Hajikolahi et al., 2006). In a dairy herd, two groups of cows are most susceptible to SARA, early lactation and mid lactation cows (Kleen et al., 2003). Field studies revealed the presence of SARA in 11-29.3% early lactation cows and in 18-23.4% mid-lactation cows (Garrett et al., 1997; Kleen, 2004; Tajik et al., 2009). Although, SARA is suggested as the most important nutritional disorder of dairy cattle (Enemark, 2008), yet the mechanism of its pathogenesis is not completely known. Furthermore, the signs of SARA are not completely known and the diagnosis is often difficult in the field due to the variable and subtle signs.

The recommended protocol for the diagnosis of SARA is collection of ruminal fluid by rumenocentesis (Nordlund et al., 1995; Garrett et al., 1999). Although the complex etiology and subclinical course of SARA complicate its diagnosis and necessitate its routine monitoring, rumenocentesis complications (haematomas and abscess formation at the puncture site and septic peritonitis) render routine monitoring of rumen pH by rumenocentesis unpractical. Therefore, finding a non invasive and simple method for detection of SARA in dairy herds will be most valuable.

The negative balance of calcium due to decreased feed intake and increased urinary excretion seemed to be the cause of increased plasma alkaline phosphatase (ALP) activity in rumen acidosis affected cows (Harmon and Britton, 1988). On the other hand, decrease of dry matter intake is the more consistent sign of SARA in affected cows (Kleen et al., 2003).

Plasma amylase activity was observed to increase in ruminal acidosis affected goats and was believed to be due to pancreatic tissue damage (Lal et al., 1992). However, Krehbiel et al. (1995)
did not report blood amylase increase in ruminal acidosis affected lambs and suggested that the severity of the acidosis and rumen pH decrement was the determinant factor of pancreatic tissue damage. SARA has to be defined as an intermittent fall of the ruminal pH to non physiological levels and the severity of rumen pH decrement is less than acute ruminal acidosis. Despite this fact, Brown et al. (2000) observed increased serum amylase activity in SARA-affected cows. It seems there is some controversy concerning the change of amylase in SARA-affected cows.

This research was designed to study blood serum ALP and amylase activities in SARA-affected and compared to non-affected cows.

MATERIALS AND METHODS

From September 2007 to November 2007, 7 Holstein dairy herds in Khorasan Razavi province, northeast of Iran, were selected according to willing for participation in the study. All herds were fed Total Mixed Rations (TMR), which were formulated to meet all NRC nutrient recommendations. In all herds, the ration was consisted of alfalfa and corn silage as the forage and different ratios of barley and maize milled grains as the concentrate.

Two groups of 12 cows were selected randomly in each herd. One group consisted of early lactation cows (3-20 days in milk) while the other consisted of mid-lactation cows (60-150 days in milk). Four to six hours following morning TMR feeding, ruminal fluid collection was carried out by means of rumenocentesis (Nordlund et al., 1995) from selected cows. Ruminal fluid pH was determined immediately with a portable pH-meter (Horiba, B-213, Kyoto, Japan). Also, jugular vein blood samples were collected at the time of rumenocentesis from all cows. The blood serum was separated after centrifugation at 1800 g for 10 min and stored at -18°C until analysis.

Alkaline phosphatase (Bowers and McComb, 1975) and amylase (Winn-Deen et al., 1988) activities were measured in the stored serum samples by commercial kits (ZiestChem Diagnostics Tehran, Iran) using an autoanalyser (Ependorf, EPOS analyzer 5060, Germany). Control serum (ZiestChem Diagnostics Tehran, Iran) was used for controlling measurement accuracy.

Statistical analysis was preformed using SPSS12 (Illinois, Chicago). Two sample t-tests were used to compare the serum ALP and amylase activities between SARA-affected cows and the rest of the cows and to detect differences between early lactation and mid-lactation groups. Correlations of ALP and amylase with the ruminal pH were analyzed by Pearson's correlation tests. Analysis of variance (ANOVA) test was used for comparison of ALP activity between animals with SARA, animals with a marginal pH and the rest of the cows. Because of unequal variances, Kruskal-Wallis test were used to compare amylase between SARA-affected, marginally affected and healthy cows. With the same reason, Kruskal-Wallis tests were used to compare amylase when early and mid lactation cows were evaluated separately. Differences were considered significant at p<0.05.

RESULTS

It was possible to draw a ruminal fluid from 159 out of 168 initially selected animals (75 early lactation and 84 mid lactation cows). Cows with a rumen pH of 5.5 or less at the time of rumenocentesis, considered to be experiencing SARA and a ruminal pH≤5.8 considered as a non affected cow. Animals with rumen pH values between 5.6 and 5.8 were considered to be marginally acidotic. If three or more cows in a rational group have rumen pH of 5.5 or less, the group was considered to be experiencing SARA (Nordlund et al., 1995; Garrett et al., 1999).

The concentrations of serum ALP and amylase in SARA-affected and the rest of the cows are shown in Table 1. There were no significant difference between SARA-affected and the rest of the cows in blood ALP and amylase. Also, the differences were not significant when early lactation and
mid lactation cows were evaluated separately. No significant difference was found between early and mid lactation cows in the blood ALP and amylase activities. At the time of rumenocentesis, early lactation cows in 4 farms and mid-lactation cows in 5 farms were found to be experiencing SARA. The comparison of mean ALP and amylase between SARA affected and non affected rational groups using student's t- tests showed no significant difference.

The blood ALP and amylase activities had no significant correlations with the rumen pH (r = -0.057 and r = -0.04, respectively), But there was a significant correlation between blood ALP and amylase activities (r = 0.239, p = 0.006). When early and mid lactation cows were evaluated separately, the correlations of the blood ALP and amylase activities with rumen pH were not significant. But, the correlation between the blood ALP and amylase activities was significant in early lactation cows (r = 0.331, p = 0.009). The concentrations of serum ALP and amylase in SARA-affected, marginally affected and healthy cows are shown in Table 2, no significant difference was detected between animals with SARA and those with marginal pH values and healthy cows for blood ALP and amylase activities. The differences were not significant when early and mid lactation cows were evaluated separately.

**DISCUSSION**

The normal range of serum ALP activity in cattle is wide, 0-488 IU mL⁻¹ (Kaneko et al., 1997). So, in this study, serum ALP activities of all sampled cows were normal. Harmon and Britton (1983) believed that the increase in the serum ALP activity of rumen acidosis affected cows was due to feed
intake decrement and urinary excretion of calcium. Despite the decrease of dry matter intake in SARA-affected cows (Kleen et al., 2003), no serum ALP change was detected. On the other hand, Brown et al. (2000) reported that the change in blood pH of SARA affected cows was small, hence, urinary excretion of calcium like that observed by Harmon and Britton (1983) in acute acidosis, did not happen during SARA. In Harmon and Britton (1983) experiment, the ruminal pH of wethers, 6 hours after feeding a high concentrate diet, was between 5.2 and 5.5. According to definition of SARA by Krause and Oetzel (2006), it seems that the wethers involved in this study were suffered from SARA. In Harmon and Britton (1983) study, blood ALP increased significantly after feeding 90% concentrate diets for 10 days. In Brown et al. (2000) study, steers received a SARA inducing meal and blood ALP was measured for 14 days. Brown et al. (2000) did not observe any change in the serum ALP activity during the experimentally induced SARA in beef steers which is same to our results.

Blood activity of amylase is routinely used as a clinical laboratory test for diagnosis of acute pancreatitis (Krehbiel et al., 1995). Krehbiel et al. (1995) believe that the severity of rumen pH drop in rumen acidosis is the determinant factor of pancreatic tissue damage. Although decrease of rumen pH in SARA is intermittent, Brown et al. (2000) reported increased serum amylase activity in SARA-affected cows. But, according to our results, there was no significant difference between SARA-affected and non affected cows and rational groups in the blood serum amylase activity and no significant correlation was found between the rumen pH and the serum amylase activity. Mohamed et al. (2003) found that the serum amylase values in cows with chronic pancreatitis remain normal. Therefore pancreatic damages in SARA affected cows can not be rejected.

Brown et al. (2000) suggested that the serum amylase and ALP activities might be useful in distinguishing between SARA-affected and non affected steers. But, present results did not confirm this. It seems that there are some differences between experimentally controlled studies of SARA and farm condition and the results of the experimental studies may be different from what is happening in dairy farms. Also, pathologic study and comparison of pancreatic lesions between SARA-affected and non affected cows in dairy farms is recommendable.

REFERENCES


