ISSN 1819-1878

Asian Journal of Animal Sciences

http://knowledgiascientific.com

Knowledgia SCIENTIFIC
A Place to Publish Outstanding Research

Asian Journal of Animal Sciences 5 (2): 153-157, 2011
ISSN 1819-1878 / DOI: 10.3923/ajas.2011.153.157

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, 2006). 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 18-26.4% 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, 1983). 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 $pH \geq 5.8$ 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

Table 1: The concentrations of serum ALP and amylase (mean± SEM) in SARA- affected and the rest of the cows (consisted of marginally affected and healthy cows)

Cow group	No. of cows	Serum ALP activity (IU mL^{-1})	Serum amylase activity (IU mL^{-1})
All sampled cows	159	93.5±3.85	26.38±2.46
SARA- affected cows	45	90.36±6.3	23.30±2.76
The rest of the cows	114	94.70±4.75	27.44±3.17
Early lactation cows	75	87.65±4.97	24.58±4.38
SARA- affected cows	22	76.64±6.4	17.0±10.48
The rest of the cows	53	91.61±6.86	27.48±6.31
Mid lactation cows	84	98.90±5.53	28.03±1.9
SARA- affected cows	23	104.09±10.16	30.00±5.01
The rest of the cows	60	97.17±6.6	27.40±1.94

Table 2: The concentrations of serum ALP and amylase (mean±SEM) in SARA- affected, marginally affected and healthy cows

Cow group	No. of cows	Serum ALP activity (IU mL^{-1})	Serum amylase activity (IU mL^{-1})
All sampled cows	159	93.50±3.85	26.38±2.46
SARA- affected cows	45	96.53±6.58	26.54±2.48
marginally affected cows	34	88.52±7.74	31.55±12.85
Healthy cows	80	93.85±5.85	24.72±1.76
Early lactation cows	75	87.65±4.97	24.58±4.38
SARA- affected cows	22	76.64±6.4	17.00±1.48
marginally affected cows	9	87.63±14.76	21.91±2.6
Healthy cows	44	92.35±7.73	21.55±2.15
Mid lactation cows	84	98.90±5.53	28.03±1.9
SARA- affected cows	23	104.09±10.16	30.00±5.01
marginally affected cows	25	103.24±7.87	24.05±2.03
Healthy cows	36	93.28±9.61	29.45±2.84

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

Bowers, G.N. Jr. and R.B. McComb, 1975. Measurement of total alkaline phosphatase activity in human serum. Clin. Chem., 21: 1988-1995.

Brown, M.S., C.R. Krehbiel, M.L. Galyean, M.D. Remmenga and J.P. Peters *et al*., 2000. Evaluation of models of acute and subacute acidosis on dry matter intake, ruminal fermentation, blood chemistry and endocrine profiles of beef steers. J. Anim. Sci., 78: 3155-3168.

Enemark, J.M.D., 2008. The monitoring, prevention and treatment of sub-acute ruminal acidosis (SARA): A review. Vet. J., 176: 32-43.

Garrett, E.F., K.V. Nordlund, W.J. Goodger and G.R. Oetzel, 1997. A cross-sectional field study investigating the effect of periparturient dietary management on ruminal pH in early lactation dairy cows. J. Dairy Sci., 80: 169-169.

Garrett, E.F., M.N. Perreira, K.V. Nordlund, L.E. Armentano, W.J. Goodger and G.R. Oetzel, 1999. Diagnostic methods for the detection of subacute ruminal acidosis in dairy cows. J. Dairy Sci., 82: 1170-1178.

Hajikolahi, M.R.H., M. Nouri, F.S. Afshar and A.J. Dehkordi, 2006. Effects of experimentally ruminal lactic acidosis on blood pH, Bicarbonate and pCO2 in the sheep. Pak. J. Biol. Sci., 9: 2003-2005.

Harmon, D.L. and R.A. Britton, 1983. Balance and urinary excretion of calcium, magnesium and phosphorus in response to high concentrate feeding and lactate infusion in lambs. J. Anim. Sci., 57: 1306-1315.

Kleen, J.L., G.A. Hooijer, J. Rehage and J.P.T. Noordhuizen, 2003. Subacute ruminal acidosis (SARA): A review. J. Vet. Med. A Physiol. Pathol. Clin. Med. , 50: 406-414.

Kleen, J.L., 2004. Prevalence of subacute ruminal acidosis in Deutch dairy herds-A field study. Ph.D. Thesis, School of Veterinary Medicine Hanover, pp: 93-104.

Kaneko, J.J., J.W. Harvey and M.L. Bruss, 1997. Clinical Biochemistry of Domestic Animals. 5th Edn., Harcourt Brace and Co. Asia Pvt. Ltd., Singapore, ISBN: 981403312X, pp: 893.

Krause, K.M. and G.R. Oetzel, 2005. Inducing subacute ruminal acidosis in lactating dairy cows. J. Dairy Sci., 88: 3633-3639.

Krause, M.K. and G.R. Otzel, 2006. Understanding and preventing subacute ruminal acidosis in dairy herds: A review. Anim. Feed Sci. Technol., 126: 215-236.

Krehbiel, C.R., R.A. Britton, D.L. Harmon, T.J. Wester and R.A. Stock, 1995. The effects of ruminal acidosis on volatile fatty acid absorption and plasma activities of pancreatic enzymes in lambs. J. Anim. Sci., 73: 3111-3121.

Lal, S.B., S.K. Dwivedi, M.C. Sharma and D. Swarup, 1992. Biopathological studies in experimentally induced ruminal acidosis in goats. Indian J. Anim. Sci., 62: 200-204.

Mohamed, T., H. Sato, T. Kurosawa, S. Oikawa and A. Nitanai, 2003. Ultrasonographic imaging of experimentally induced pancreatitis in cattle. Vet. J., 165: 314-324.

Nordlund, K.V., E.F. Garrett and G.R. Oetzel, 1995. Herd-based rumenocentesis-a clinical approach to the diagnosis of subacute rumen acidosis. Compend. Contin. Educ. Pract. Vet., 17: S48-S56.

Oetzel, G.R., 2003. Introduction to ruminal acidosis in dairy cattle. Proceedings of the 36th Annual Conference of American Association of Bovine Practitioners, Sept. 15-17, Columbus, OH.

Tajik, J., M.G. Nadalian, A. Raoofi, G.R. Mohammadi and A.R. Bahonar, 2009. Prevalence of ubacute ruminal acidosis in some dairy herds of Khorasan Razavi Province, Northeast of Iran. Iran. J. Vet. Res., 10: 28-32.

Winn-Deen, E.S., H. David, G. Sigier and R. Chavez, 1988. Development of a direct assay for a-amylase. Clin. Chem., 79: 2005-2008.