ADENOSINE DEAMINASE AND BIOCHEMICAL LIVER FUNCTION TESTS IN THE DERMATOPHYTIC CATTLE

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Abstract

Adenosine deaminase (ADA) activity and liver function tests were examined in dermatophytic cattle. In addition, the possible effect/s of the disease on the activities of liver enzymes were investigated. Ten dermatophytic and 10 healthy animals aged between 10 and 12 months were used. Blood samples were obtained from each animal. The samples were left for 30 min at room temperature, and then centrifuged at 3000 x g for 10 min. Serum ADA, lactate dehydrogenase (LDH), alanine aminotransferase (ALT), aspartate aminotransferase (AST), gammaglutamyl transferase (GGT) were analysed colorimetrically. Serum ADA, LDH, and ALT activities were significantly higher in dermatophytic cattle, compared to those of healthy group (P<0.01). In addition, GGT and AST activities in dermatophytic cattle were statistically compared higher to those of the healthy ones. In conclusion, the results of the present study show that the increases in ADA and liver enzyme activities can be an indication of the loss of liver function. The increase in enzyme activity in the current study might have been caused by metabolic products of the fungi.

Key words: cattle, trichophytosis, adenosine deaminase, liver, biochemistry.

Ringworm or dermatophytosis of cattle is exclusively caused by Trichophyton verrucosum. This skin disease is worldwide present in cattle and is responsible for high economic losses in cattle farming. T. verrucosum may also be responsible for severe skin diseases in man. Dermatophytosis is a zoonotic disease that can be transmitted between animals and humans (24). The infection is common at the time of weaning and occurs through the year with a higher incidence during wintertime due to proper conditions for the growth of spores. Additionally, the disease is closely related to immunity and skin pH (19). Dermatophytes are keratinophilic fungi that are able to invade the stratum corneum of the skin and other keratinized structures. The pathogenic interactions between host and fungus are poorly understood (20). Dermatophytes exhibit activity of extracellular enzymes including elastase, keratinase, protease (gelatinase), lipase, and phospholipase (17).

Adenosine deaminase (ADA, adenosine aminohydrolase, E.C.3.5.4.4) catalyses hydrolytic deamination of either adenosine or deoxyadenosine to produce inosine and deoxyinosine, respectively. Serum ADA activity has been found higher in diseases that stimulate immune response in host such as chronic hepatitis, liver cirrhosis, hepatocellular carcinoma. It is purposed that analyses of serum ADA levels in humans and animals are important determinants for the diagnosis of liver diseases (1, 8, 22).

Alanine aminotransferase, a sensitive indicator of liver cell injury, has been used to identify patients with liver disease. This cytosolic enzyme, which is found in many organs, catalyses the transfer of the α-amino group from alanine to α-ketoglutaric acid. Alanine aminotransferase levels are particularly high in the liver. For the detection of liver diseases, ALT is thought to be a more specific indicator than AST, an enzyme found in cytosol and mitochondria (13). LDH and GGT are also commonly used parameters for the diagnosis of liver diseases (16, 21, 23).

Although it has been reported that liver tissue can be influenced by some skin disease such as superficial necrotic dermatitis, metabolic epidermal necrosis or atopic dermatitis (4, 10, 12, 15), not many studies have been done to investigate the possible effect(s) of dermatophytosis on the liver. In the present study, ADA activity and liver function tests that were known to be indicators of liver damage were examined. It was also aimed whether dermatophytosis has an effect on liver function.
Material and Methods

Animals. Ten dermatophytic and 10 healthy beef cattle aged between 10 and 12 months were chosen as the study material. The skin and complete clinical examination of all the animals were carried out including general state, pulse, temperature, respiratory rate, shape, position, and distribution of lesions as well. Ten millilitre blood samples were obtained from each animal. The samples were left for 30 min at room temperature and then centrifuged at 3000 x g for 10 min. The samples were kept frozen (-25 °C) until analysis. All serum samples were analysed within 1 month.

Biochemical analysis. Serum ADA activity was determined at 37°C according to the method of Giusti and Galanti (9) based on the Bertholet reaction, that is, the formation of coloured indophenol complex from ammonia liberated from adenosine and quantified colorimetrically on a spectrophotometer (UV-1201, Shimadzu, Japan). One unit of ADA is defined as the amount of enzyme required to release 1 mmol of ammonia/min from adenosine at standard assay condition. Results were expressed as international unit of enzyme activity of serum.

The activities of serum AST (EC 2.6.1.1), ALT (EC 2.6.1.2), GGT (EC 2.3.2.2) and LDH (EC 1.1.1.27) were assayed colorimetrically according to the standard procedures using commercially available diagnostic kits obtained from Diasis Diagnostic Systems (Holzheim, Germany)

Mycological analysis. After cleaning the area with a cotton swab, soaked with 70% ethyl alcohol, samples were collected by scraping the lesion using a sterile scalpel blade and put into sterile Petri dishes. Hairs were also collected from the lesion. The collected samples were divided into two portions. One of them was used for direct microscopic examination and the other one for culture on mycobiotic agar (DIFCO), incubated at 28°C for 2-6 weeks for the colony formation. In order to identify the pathogenic fungi, macroscopic and microscopic examinations were carried out and appearance of the growth, colony morphology, and colour, shape, size, and colony reverse side morphology were examined. Microscopic examination for positive fungi cultures was done using the Lactophenol cotton blue wet mount method (11). For the identification of the fungi, selective agar medium for Trichophyton (DIFCO) was used.

Statistical analysis was performed using SPSS 10.0. The data for biochemical parameters were analysed using analysis of variance (ANOVA). All the data are presented as mean ± SE. Values were considered statistically significant when P < 0.05.

Results

Serum ADA, AST, ALT, GGT, LDH activities of healthy and trichophytic animals and results of microscopic examination are presented in Table 1.

As can be seen from the Table 1 all the enzymes showed increased activities in dermatophytic calves compared to those of healthy group. This increase was highly significant in the case of ADA, LDH, ALT, and AST. While dermatophytic calves showed positive results in microscopic examination, healthy ones were negative.

Table 1

Mean enzyme activities in healthy and dermatophytic calves

<table>
<thead>
<tr>
<th>Enzyme</th>
<th>Dermatophytic calves</th>
<th>Healthy calves</th>
<th>p</th>
</tr>
</thead>
<tbody>
<tr>
<td>GGT (U/L)</td>
<td>7.98±0.40</td>
<td>6.63±0.11</td>
<td>&lt;0.05</td>
</tr>
<tr>
<td>LDH (U/L)</td>
<td>550.57±7.63</td>
<td>503.13±6.70</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td>AST (U/L)</td>
<td>30.12±0.81</td>
<td>23.23±1.11</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>ALT (U/L)</td>
<td>13.27±0.25</td>
<td>11.95±0.24</td>
<td>&lt;0.01</td>
</tr>
<tr>
<td>ADA (U/L)</td>
<td>10.29±0.47</td>
<td>6.73±0.56</td>
<td>&lt;0.01</td>
</tr>
</tbody>
</table>

Discussion

Dermatophyte is generally a cutaneous pathogen and restricted to the non-living cornified layers. Reaction to a dermatophyte infection may range from mild to severe as a consequence of the host’s reactions to the metabolic products of the fungus, the virulence of the infecting strain or species, the anatomic location of the infection, and local environmental factors. The immunology of the dermatophytosis is a continuously developing field. The immunobiology of the skin is a subject rich with possibilities for advancing our understanding of disease processes (6). Fungi produce different types of proteolytic enzymes specially keratinases that have key roles in fungal invasion and pathogenesis (17, 20). During the invasion the metabolic products of the fungi may penetrate the liver tissue by passing through the skin and blood circulation.

An increase in ADA activity has been reported in animals (1) and humans with liver diseases. In addition, it has been reported that increased serum ADA activity in skin leishmaniosis and cattle dermatophytosis may be a reflection of phagocytic activity of macrophages. As a marker of cellular immunity, the plasma activity of ADA can be found to be elevated in diseases showing a cell-mediated immune response (5). In our opinion, the increased serum ADA activity in dermatophytic cattle may be a reflection of phagocytic activity of macrophages and may provide useful additional diagnostic information on the pathogenesis of hepatitis.

AST and ALT are cytosolic enzymes having wide tissue distribution with the highest concentrations found in the liver. Thus, they were the first enzymes to
be used in diagnostic enzymology when liver damage occurred. Due to their intracellular location in the cytosol, toxicity or disease affecting the liver with subsequent breakdown in membrane structure of hepatocytes, leads to their spillage into plasma and their concentration rises in the latter (7). Although Kumar et al. (14) did not find any statistical difference in AST and ALT levels between healthy and dermatophytic cattle, levels of both enzymes increased in dermatophytic cattle in this study.

LDH is a cytosolic enzyme, which is essentially present in all tissues involved in glycolysis. With any destructive process of these tissues, the enzyme leaks into extracellular fluids and then into body fluids. Hence detection of elevated concentration of this enzyme released into the blood stream from the damaged tissues has become a definitive diagnostic and prognostic criterion for various diseases and disorders (2, 3, 18).

Abnormally high activity of serum GGT appears to be specific for diseases of the liver, biliary tract, and pancreas. Interest in GGT has focused on its appearance to be specific for diseases of the liver, biliary tract, and pancreas. Interest in GGT has focused on its value in the diagnosis of various liver diseases. The severity of fatty liver can be evaluated using GGT value in the diagnosis of various liver diseases. The tract, and pancreas. Interest in GGT has focused on its activity in dogs. Hence detection of elevated concentration of this enzyme released into the blood stream from the damaged tissues has become a definitive diagnostic and prognostic criterion for various diseases and disorders (2, 3, 18).

In conclusion, the results of the present study show that the increases in ADA and liver enzyme activities can be an indication of loss of the liver function. These increases in the enzyme activities in the current study might have been caused by metabolic products of the fungi.

References