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CLINICAL INVESTIGATION

Serum Adenosine Deaminase Levels in Patients with Brucellosis and in Healthy Subjects*

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Abstract: The aim of this study was to determine the serum adenosine deaminase (ADA) levels in patients with brucellosis and compare them with the results of healthy individuals. Forty- eight subject were enrolled in this study : 34 patients with brucellosis and 14 healthy individuals. Serum ADA activity was assessed by spectrophotometer in patients with brucellosis and statistically compared with that of healthy individuals. A normal range of serum ADA was considered 5-35 IU/l. Serum ADA levels were also compared with *Brucella* agglutinations titers and serum C reactive protein (CRP).

Serum ADA levels were found to be significantly higher in patients with brucellosis than in healthy individuals (43.45 ± 24.19 IU/l and 27.5 ± 9.3 IU/l, respectively) ($P < 0.01$). Serum ADA activity did not show any correlation between the *Brucella* agglutination titer and CRP level.

Serum ADA level showed significant alterations in patients with brucellosis compared to healthy subjects.

We concluded that serum ADA level may be used in the follow up of patients with brucellosis together with clinical and other laboratory findings.

Key Words: Brucellosis, adenosine deaminase, activity.

Introduction

Adenosine deaminase (ADA) is an essential enzyme for the differentiation and proliferation of T lymphocytes and the monocyte-macrophage systems (1).

ADA is involved in the purine metabolism and plays a significant role in the mechanisms of the immune system (2). Increased serum ADA activity, mainly associated with tuberculosis, can also occur in other infectious and non-infectious diseases such as brucellosis and rheumatoid arthritis (2,3). The basic immunological response of brucellosis is cellular (4). In this study, serum ADA activity was measured in brucellosis and serum ADA levels were compared between patients with brucellosis and healthy individuals.

Materials and Methods

Forty- eight subjects were enrolled in this study : 34 patients with brucellosis and 14 healthy individuals. In individuals having clinical symptoms and findings relevant to brucellosis, a serological diagnosis was made by

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standard agglutination test (SAT) giving positive results above or equal to 1/160 titration. The control group was selected from healthy volunteers who had no disease and who were negative for SAT. Serum ADA activity was assessed by a colorimetric method in patients with brucellosis and statistically compared to that of healthy individuals.

Serum ADA levels were measured by spectrophotometer in the sera of patients with brucellosis and healthy subjects. ADA activity was determined according to Bergmeyer's methods (2,5). A normal range for ADA was considered 5-35 IU/l.

Serum CRP was measured by nephelometric methods (N High Sensitivity CRP, Dode Behring, Germany). A normal range of CRP was considered 0-5 mg/l. Serum ADA levels were compared with SAT titers and CRP level. Statistical analysis was perfrmed with SPSS for Windows version 6. program. The mean values obtained in the 2 groups were compared by chi-square test and Student's t test.

A Kruskal – Wallis test was run to find whether the determination of ADA levels in sera might differ with *Brucella* agglutinations and CRP. Pearson's correlation test was used to evaluate correlations amongst the parameters.

All results are expressed as mean values + standard deviation (SD), and statistical significance is defined as $P < 0.01$.

Table 1. Comparison of mean serum ADA levels in patients with brucellosis and healthy subjects.

	Mean serum ADA level (IU/l)
Patients with brucellosis	43.45 ± 24.19
Healthy subjects	27.5 ± 9.3

Results

Serum ADA levels were significantly higher in patients with brucellosis than those in healthy individuals ($P < 0.01$). The results are shown in Table 1.

Serum ADA activity was compared to normal reference values in patients with brucellosis and healthy subjects. The results are shown in Table 2.

Thirty-one patients with brucellosis had a high level of CRP and 3 patients of the healthy controls had a high level of CRP. Serum ADA activity did not show any correlation between the *Brucellla* agglutination titer and CRP level (data not shown).

Discussion

Brucellosis is a zoonotic disease that often becomes chronic with a high rate of recurrence. Turkey is an endemic area for brucellosis and the incidence of brucellosis here is rising alarmingly (6). The intracellular survival of this microorganism in the reticuloendothelial system cells determines the chronic course of the disease, as well as the inability of antibiotic therapy to eradicate the infection completely. Protective immunity against intracellular bacteria depends on the interplay between various T cell subsets and cytokines. It has been established in a murine model that protective acquired immunity to brucellae involves T-cell dependent activation of macrophages in which both CD4+ and CD8+ T cells contribute to protection against *Brucella* infection. Cell-mediated immunity to brucellosis therefore is likely to include the production of cytokines that activate macrophages and lymphocytes for production of anti-*Brucella* activities (4). CD-26 is a lymhocyte marker that anchors ADA on the T cell surface. It is found that ADA is regulated by cytokines on the cell surface during T cell activation. Interleukin (IL-2) and IL-12 up- regulate ecto-

Table 2. Distribution of ADA activity in patients with brucellosis and healthy controls compared with normal reference values of ADA.

	Low level of ADA (<5 IU/l)		High level of ADA (>35 IU/l)		Normal reference value of ADA (5-35 IU/l)		Total	
	n	(%)	n	(%)	n	(%)	n	(%)
Patients with brucellosis	7	(21.2)	26	(78.8)	1	(2.9)	34	(100)
Healthy controls	11	(78.6)	3	(21.4)	0	(0)	14	(100)

ADA and CD26 expression. In contrast, IL-4 led to down-regulation of lyphocyte surface ADA without modifiying the level of CD26 (7).

ADA is present in serum and most tissues, particularly lymphoid tissues, and is essential for the maturation and function of lymphocytes, especially those of T lineage, and is required for the maturation of human blood monocytes to macrophages. The serum ADA activity was significantly higher in the sera of patients with brucellosis than in those of healthy controls in this study. High serum ADA activity has been reported in various diseases, including infection, malignancy and liver disease (8). Viciana et al. (1) assessed serum ADA activity in 67 patients with brucellosis before therapy and at 1, 3 and 6 months of follow up. They found that serum ADA activity in brucellosis was higher than that in healthy controls, both in those with acute febrile noncomplicated form and those with focal symptoms from one organ . They did not find any differences between the brucellosis groups.

A negative correlation between the duration of the disease and ADA activity was also found. They concluded that ADA activity is increased during the active stage of brucellosis ; therefore it can be considered a biochemical follow up marker of the disease and, probably, a marker of relapses. Ahmed et al. (4) reported a high level of IL-12 and interferon (IFN)-gamma and an undetectable level of IL-4 in the serum of patients with brucellosis compared with the controls. Similarly, Demirdag et al. (9) reported that IFN-gamma and TNF-alpha levels were significantly higher in acute brucellosis patients compared to after treatment and the control group. They concluded that IFN-gamma and TNF-alpha may be used for monitoring brucellosis. Serum ADA level is correlated with some cytokines. Interleukin (IL-2) and IL-12 led to up - regulation of ADA , but IL-4 led to down- regulation of ADA (8). Karadenizli et al. (10) reported that serum ADA activity was higher in brucellosis patients . In contrast to the study by Viciana et al. (1), they reported that ADA activity is not informative in the discrimination of acute and chronic brucellosis. Similarly, Dikensoy et al. (11) reported that increased pleural fluid adenosine deaminase in brucellosis is difficult to differentiate from tuberculosis. In our study, serum ADA levels were significantly higher in the patients with brucellosis than those of healthy individuals (43.45 ± 24.19 IU/l and 27.5 ± 9.3 IU/l, respectively) ($P < 0.01$). Serum ADA activity did not show any correlation between *Brucella* agglutination titer and CRP. Serum ADA level showed significant alterations in patients with brucellosis compared to healthy subjects.

It is our conclusion that serum ADA and some cytokines such as IL-12 and INF-gamma may be useful in the follow up of patients with brucellosis before and after treatment together with clinical and laboratory findings. Further controlled studies are necessary to elucidate the role of ADA and cytokines in patients with brucellosis.

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