



Original Article

Biochemical parameters changes in infected dogs with *Babesia canis* in Urmia, Iran

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ABSTRACT

The aim of this study was to evaluate the changes in the biochemical parameters in infected dogs with babesiosis in 3 levels. The *Babesia* genus, as pathogen blood parasites, cause financial problems in livestock industry. Many published papers have suggested babesiosis-induced biochemical parameters, but no study has determined these parameters at four levels of parasitemia (less than 1%, 1%, 2% and 3%). Dogs with babesiosis were identified based on clinical signs and the observation in red blood cells. After blood sampling from 38 infected and 38 healthy dogs, biochemical parameters glucose paraoxonase, superoxide dismutase, catalase, glutathione peroxidase and adenosinedeaminas were measured in plasma. The results indicated significant increase and decrease in biochemical parameters. In conclusion, the results suggested that dog babesiosis causes cellular damage, especially in kidney and anemia in infected dogs. Furthermore, babesiosis caused kidney damage. This infection affected the activity of liver enzymes. The obtained results revealed a highly significant increase in serum Aspartate aminotransferase and Alanine aminotransferase.

تغییرات پارامترهای بیوشیمیایی در سگ های آلوده به بابزیا کانیس در ارومیه، ایران

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چکیده

هدف از این مطالعه بررسی تغییرات پارامترهای بیوشیمیایی در سگ های آلوده به بابزیز در سه سطح بود. جنس بابزیا به عنوان یک انگل خونی بیماری زا باعث ایجاد مشکلات مالی در صنعت دامپروری می شود. بسیاری از مقالات منتشر شده پارامترهای بیوشیمیایی ناشی از بابزیز را پیشنهاد کرده اند، اما هیچ مطالعه ای این پارامترها را در چهار سطح از پارازیتمی (کمتر از ۱٪، ۱٪، ۲٪ و ۳٪) تعیین نکرده است. سگ های مبتلا به بابزیز بر اساس علائم بالینی و مشاهده گلبول های قرمز شناسایی شدند. پس از خونگیری از ۳۸ سگ آلوده و ۳۸ سگ سالم، پارامترهای بیوشیمیایی گلوکز پاراکسوناز، سوپراکسید دیسموتاز، کاتالاز، گلووتاتیون پراکسیداز و آدنوزین دامینا در پلاسما اندازه گیری شد. نتایج حاکی از افزایش و کاهش معنی داری در پارامترهای بیوشیمیایی بود. در نتیجه، نتایج نشان داد که بابزیز سگ باعث آسیب سلولی، به ویژه در کلیه و کم خونی در سگ های آلوده می شود. علاوه بر این، بابزیز باعث صدمات کلیه شد. این عفونت بر فعالیت آنزیم های کبدی تأثیر می گذارد. نتایج به دست آمده نشان دهنده افزایش بسیار معنی دار آسپاراتات آمینوترانسفراز و آلانین آمینوترانسفراز سرم بود.

واژه های کلیدی: بابزیز سگ، آسپاراتات آمینوترانسفراز، آلانین آمینوترانسفراز، مالون دی آلدیاید، ارومیه

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## INTRODUCTION

Babesiosis is an important parasitic infectious disease in the livestock industry throughout the world [1]. More than 110 species have been identified for the genus *Babesia* so far [2]. *B. canis* is a common cause of death in dogs [3]. Babesiosis is also known by other names including piroplasmosis, biliary fever, red water and Texas fever. *Babesia* species are intraerythrocytic and cause symptoms, for instance, anaemia, high fever, jaundice, enlarged spleen and liver, neutropenia, anisocytosis, thrombocytopenia, hematuria and hemoglobinuria [4]. Adenosine deaminase is an important enzyme in the maturation and differentiation of T lymphocytes and its activity is higher in T cell than B cell. Adenosine deaminase activity increases during disease by stimulating the immune response [5]. Malondialdehyde is a byproduct of lipid peroxidation. It is one of the most abundant and reliable biomarkers that plays an important role in assessing lipid peroxidation or indirect detection of free radicals. Malondialdehyde is integrated with the cell membrane, it binds to phospholipids and enzymes and causes intracellular damage [6]. Paraoxonase (PON) is an enzyme that can hydrolyze organic phosphate stresses; this enzyme circulates in the blood along with HDL and decreased activity increases the risk of coronary artery disease [7]. Superoxide dismutase, specifically named SOD, is a byproduct of oxygen metabolism that causes various cellular damage and is broken down by other enzymes such as catalase [8]. Therefore, superoxide dismutase, catalase and glutathione peroxidase are present in almost all cells that are exposed to oxygen [9]. Alanine aminotransferase ALT is an enzyme found mainly in liver and kidney cells and is a marker of liver disease. The striated muscles, myocardium, and liver tissues are the main sources of AST. So far,

studies have been conducted on changes in some biochemical and haematological parameters in the world and Iran: Therefore, this study aimed to evaluate the changes in the biochemical parameters in infected dogs with Babesiosis in Urmia.

## MATERIALS AND METHODS

In this study, which was conducted in June 2020, out of 62 dogs studied in Urmia, 38 dogs with an average weight of  $1.38 \pm 22.14$  in the age range of 1-1.5 years, after blood sampling and staining of blood smear with 5% Giemsa, *Babesia canis* was identified and no parasites were observed in the other dogs. *Babesia canis* was identified and no blood parasites were observed in the other dogs. In terms of clinical examination, the absence of ticks was performed on the surface of the skin and clinical symptoms were observed, including hyperthermia, anorexia, submucosal haemorrhage, jaundice, hemoglobinuria and anaemia accompanied by pale mucous membrane. Parasitic determination (based on percentage) was performed based on the number of infected red blood cells under a percentage of a microscopic field with a magnification of 100 (>1- 1%, 2%, 3%) [10]. After blood sampling and transfer to tubes containing anticoagulant solution (EDTA), the samples were centrifuged at 600 rpm for 5 minutes and finally, plasma was separated from the blood. MDA with (Satoh) method, cardiac troponin I by ELISA method (Cobas co, 1978) Antioxidant enzymes (PON, CAT, GSH, SOD) by colourimetric method and parameters of glucose, adenosine deaminase, creatinine, urea, total protein and enzyme Liver alanine aminotransferase and aspartate aminotransferase were measured with a spectrophotometer using diagnostic kits (Pars Azmoun Company, Tehran).

**RESULTS**

The tables 1, 2, 3 and 4 show the comparison of changes in biochemical parameters at levels less

than 1%, 1%, 2% and 3% in the parasite-infected group with the group without parasitic infection. Accordingly, the results showed that the mean parameters of malondialdehyde, creatinine, urea,

**Table 1.** Comparing biochemical parameters in infected dogs to healthy dog at level <1%

Biochemical parameters	Control group	Patient group
Glutathione peroxidase (U/ mgHb)	191.50±11.18	68.20±11.94 <sup>†</sup>
Malondialdehyde (nmol / ml)	3.14±0.89	10.18±1.70 <sup>†</sup>
Adenosine deaminase (U / L)	59.08±2.15	16.56±1.80 <sup>†</sup>
Creatinine (mg / dl)	0.68±0.072	1.86±0.064 <sup>†</sup>
Urea (mg / dl)	0.47±1.08	5.22±0.89 <sup>†</sup>
Total protein (mg / dl)	2.44±0.39	5.68±0.46 <sup>†</sup>
Aspartate aminotransferase (U / L)	39.13±2.77	112.69±4.18 <sup>†</sup>
Alanine aminotransferase (U/L)	13.86±2.79	41.70±3.17 <sup>†</sup>
Glucose (mg / dl)	96.55±4.70	125.18±3.84 <sup>†</sup>
Cardiac troponin I (ng / ml)	0.28±0.018	0.129±0.016 <sup>†</sup>
Paraoxonase (U / L)	82.59±6.49	32.01±3.76 <sup>†</sup>
Superoxide dismutase (U / gHb)	764.26±31.12	380.12±15.90 <sup>†</sup>
Catalase (k / gHb)	294.70±3.12	89.63±5.19 <sup>†</sup>

Note: Numbers expressed based on mean ± and standard deviation † that shows significant difference with control group.

**Table 2.** Comparing biochemical parameters in infected dogs to healthy dog at level 1%

Biochemical parameters	Control group	Patient group
Glutathione peroxidase (U/ mgHb)	186.36±10.66	31.72±4.40 <sup>†</sup>
Malondialdehyde (nmol / ml)	2.82±1.09	15.64±1.3 <sup>†</sup>
Adenosine deaminase (U / L)	44.32±3.08	7.61±1.19 <sup>†</sup>
Creatinine (mg / dl)	0.53±0.024	2.91±0.045 <sup>†</sup>
Urea (mg / dl)	1.12±0.6	7.09±1.24 <sup>†</sup>
Total protein (mg / dl)	6.12±0.15	7.26±0.74 <sup>†</sup>
Aspartate aminotransferase (U / L)	45.80±3.12	164.08±9.18 <sup>†</sup>
Alanine aminotransferase (U/L)	17.51±1.61	54.29±2.10 <sup>†</sup>
Glucose (mg / dl)	102.70±3.66	122.86±4.12 <sup>†</sup>
Cardiac troponin I (ng / ml)	0.32±0.044	0.174±0.018 <sup>†</sup>
Paraoxonase (U / L)	82.52±6.49	18.50±1.35 <sup>†</sup>
Superoxide dismutase (U / gHb)	870.94±31.12	236.24±18.90 <sup>†</sup>
Catalase (k / gHb)	394.01±12.85	56.61±7.20 <sup>†</sup>

Note: Numbers expressed based on mean ± and standard deviation † that shows significant difference with control group.

**Table 3.** Comparing biochemical parameters in infected dogs to healthy dog at level 2%

Biochemical parameters	Control group	Patient group
Glutathione peroxidase (U/ mgHb)	134.99±12.58	8.66±1.70 <sup>†</sup>
Malondialdehyde (nmol / ml)	4.13±1.55	29.36±6.12 <sup>†</sup>
Adenosine deaminase (U / L)	39.08±2.19	7.54±0.83 <sup>†</sup>
Creatinine (mg / dl)	0.79±0.023	5.08±0.038 <sup>†</sup>
Urea (mg / dl)	2.06±0.31	9.87±1.30 <sup>†</sup>
Total protein (mg / dl)	5.71±0.29	8.96±0.75 <sup>†</sup>
Aspartate aminotransferase (U / L)	38.10±3.49	362.18±9.25 <sup>†</sup>
Alanine aminotransferase (U/L)	24.63±5.01	41.70±3.17 <sup>†</sup>
Glucose (mg / dl)	98.13±6.94	159.12±4.60 <sup>†</sup>
Cardiac troponin I (ng / ml)	0.032±0.013	4.76±0.071 <sup>†</sup>
Paraoxonase (U / L)	94.75±9.28	6.30±1.11 <sup>†</sup>
Superoxide dismutase (U / gHb)	374.26±29.46	89.39±11.12 <sup>†</sup>
Catalase (k / gHb)	351.43±24.82	22.08±2.10 <sup>†</sup>

Note: Numbers expressed based on mean ± and standard deviation † that shows significant difference with control group.

**Table 4.** Comparing biochemical parameters in infected dogs to healthy dog at level 3%

Biochemical parameters	Control group	Patient group
Glutathione peroxidase (U/ mgHb)	150.22±9.44	11.32±2.15 <sup>†</sup>
Malondialdehyde (nmol / ml)	3.54±2.12	21.87±9.61 <sup>†</sup>
Adenosine deaminase (U / L)	46.01±5.39	29.0±8.36 <sup>†</sup>
Creatinine (mg / dl)	0.61±0.011	4.30±0.021 <sup>†</sup>
Urea (mg / dl)	9.10±2.10	1.89±0.15 <sup>†</sup>
Total protein (mg / dl)	6.04±0.19	8.46±0.91 <sup>†</sup>
Aspartate aminotransferase (U / L)	42.39±2.22	214.05±11.60 <sup>†</sup>
Alanine aminotransferase (U/L)	18.73±3.80	76.94±4.50 <sup>†</sup>
Glucose (mg / dl)	92.72±4.38	141.28±6.67 <sup>†</sup>
Cardiac troponin I (ng / ml)	0.046±0.067	0.239±0.029 <sup>†</sup>
Paraoxonase (U / L)	125.49±10.08	9.77±0.88 <sup>†</sup>
Superoxide dismutase (U / gHb)	857.91±38.59	174.36±12.77 <sup>†</sup>
Catalase (k / gHb)	294.52±18.06	35.08±4.77 <sup>†</sup>

Note: Numbers expressed based on mean ± and standard deviation † that shows significant difference with control group.

total protein, cardiac troponin, aspartate aminotransferase, alanine aminotransferase and glucose in dogs infected with babesiosis had a significant increase compared to control dogs ( $P < 0.01$ ). In contrast, the levels of paraoxonase, superoxide dismutase, catalase, glutathione peroxidase, adenosine deaminase in the parasite-infected group were decreased compared to the group without parasitic infection ( $P < 0.01$ ).

## DISCUSSION

Babesiosis is an important disease of worldwide significance. Its effects range from relatively mild to fatal. A total of 38 dogs infected with canine babesiosis were diagnosed by clinical examination. The findings revealed that infected dogs had the significant increase of parameters malondialdehyde, creatinine, urea, total protein, cardiac troponin I, aspartate aminotransferase, alanine aminotransferase and glucose at the level of less than 1%, level 1%, level 2% and level 3% ( $P < 0.01$ ). In contrast, the levels of paraoxonase, superoxide dismutase, catalase, glutathione peroxidase, adenosine deaminase in the parasite-infected group reduced at 4 levels (Tables 1, 2, 3 and 4). The goal of this study was to evaluate the changes in biochemical parameters in infected dogs with babesiosis. Increased level of malondialdehyde has been reported in *B. gibsoni* infection [11] also in study of malondialdehyde levels in serum of dogs infected with *Babesia canis* [12]. The result demonstrated that the level of this parameter has increased. These results are in line with our findings. According to the other studies,

the level of malondialdehyde began to increase remarkably in proportion to the degree of anaemia [13]. Aspartate aminotransferase and Alanine aminotransferase level increasing could be seen with hepatic hypoxia in infected dogs [14]. The result of study about haemato-biochemical changes in natural cases of canine babesiosis indicated that changes in biochemical parameters were obvious. Our study revealed the same results.

## CONCLUSION

*Babesia* infection in dogs caused anemia and thrombocytopenia due to the increase in enzymes activity that might be attributed to severe anemia leading to hypoxic and toxic liver damages. Babesiosis caused kidney damage by forming immune complexes. The infection affects the activity of liver enzymes. The obtained results revealed a highly significant increase in serum Aspartate aminotransferase and Alanine aminotransferase.

## ETHICS

All ethical standards have been respected in this study.

## CONFLICT OF INTEREST

None declared.

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