Plasma Malondialdehyde, Thyroid Hormones and Some Blood Profiles in Ovine Babesiosis

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Makale Kodu (Article Code): KVFD-2012-8113

INTRODUCTION

Babesiosis is a tick-borne hemoprotozoan disease with host-specific features carried by members of the Babesia genus. Babesia ovis induces the expression of severe pathogenic traits in sheep and causes sheep babesiosis.

Keywords: Ovine Babesiosis, Malondialdehyde, Thyroid hormones, Blood profiles

Özet


Anahtar sözcükler: Koyun babesiosu, Malondialdehid, Tiroid hormonları, Kan profilleri

Summary

Thyroid hormone and plasma malondialdehyde (MDA) levels as well as other blood parameters were performed in ovine babesiosis. The Babesia genus comprises pathogen parasites that cause economic problems in livestock management. Many published studies have suggested its complications, but few studies have assessed blood biochemistry. Hence, a survey of the changes in blood parameters in animals with babesiosis may be useful. Sheep with acute babesiosis were identified based on clinical signs and the observation of Piroplasms in red blood cells with Giemsa staining of blood smears. Blood samples were obtained from the jugular veins of 46 babesiosis-infected sheep and 46 healthy sheep without babesiosis. Malondialdehyde (MDA), Paraoxonase (PON), low density lipoprotein (LDL), high density lipoprotein (HDL), Cholesterol (Chol), Total plasma protein (TPP) and glucose (GL) in plasma and triiodothyronine (T3), thyroxine (T4) measured in serum. Levels of erythrocyte superoxide dismutase (SOD), glutathione peroxidase (GSH-Px), catalase (CAT) and total antioxidant capacity (TAC) were also measured. The results indicated significant increases (P<0.01) in MDA and GL and significant decreases (P<0.01) in the levels of antioxidant enzymes (SOD, CAT except SOD), TPP, T3, and T4, LDL, HDL, and Chol levels when compared with healthy group. The results suggested that hypothyroidism with concurrent oxidative stress are significant signs of ovine babesiosis.

Keywords: Ovine Babesiosis, Malondialdehyde, Thyroid hormones, Blood values
which is characterized by fever, anemia, icterus and hemoglobinuria. B. ovis has been observed in most regions of Iran. A high prevalence among sheep and goats in north-eastern of Iran was reported by Razmi et al. Studies of blood profiles changes in sheep babesiosis are scarce. Hematological and biochemical alterations in splenectomized sheep with experimentally induced B. motasi infection were investigated by Alani and Herbert. In 2010 Apaydin et al. investigated serum protein fractions in sheep naturally infected with babesiosis.

Anemia is a common consequence of babesiosis, which causes parasitemia and erythrocyte destruction. Anemia results from the physical effects on erythrocytes, activated macrophage-mediated erythrocyte phagocytosis, antibody-affected erythrocytes and enhanced cell membrane permeability. Moreover, severe cases are not always accompanied by parasitemia; while non-parasitic factors may also destroy erythrocytes in animals with parasite infections. Free radicals are thought to play a role in erythrocyte decomposition; the assessment of free radicals in the pathogenesis of parasitic infections garnered increasing attention in recent years.

Free radicals and Reactive Oxygen Species (ROS) are continually produced during metabolic processes; levels may increase under pathological conditions and cause tissue damage. Oxidative stress as a result of the excessive generation of free radicals, which overwhelms the antioxidants available (redox imbalance), has been reported in some animal diseases, such as pneumonia, pig’s sepsis and airway disorders in horses. Oxidative stress causes macromolecular damage, erythrocyte oxidation (cell destruction) and poly unsaturated fatty acid oxidation, which results in lipid peroxidation. Malondialdehyde (MDA) is a by-products of lipid peroxidation. The measurement of MDA levels can be used to indirectly measure the degree of lipid peroxidation and the levels of free radicals. Literature regarding MDA is scarce but includes reports on elevated MDA levels in sheep naturally infected with Babesia ovis and cattle infected with B. bovis. The thyroid gland plays a vital role in several functions, such as, oxygen consumption by cells, energy utilization, biologic processes (metabolism, growth and nervous improvement) and the productive performance of domestic animals (hair, milk, and meat). Hence, thyroid hormone levels can be investigated as indicators of the health status of the animal. The thyroid hormones include of tri-iodothyronine (T3) and tetra-iodothyronine (T4). The chief secretory hormone in adult sheep is T4. Several factors influence thyroid function: e.g. seasonal changes, reproductive status, age and mercury-induced hypothyroidism. Although related research is abundant, investigations of Babesiosis-induced blood profile alterations are scarce.

There are no published literatures on thyroid hormones alterations in sheep babesiosis. Thus, the recent study aimed to evaluate thyroid hormones (T3, T4), MDA as a lipid peroxidation index, some blood profiles and probable inter-relationship of these factors each other in ovine babesiosis.

**MATERIAL and METHODS**

This study was conducted in western Azerbaijan province (North west of Iran) in 2011. The animals were examined for the presence of ticks and clinical signs including hyperthermia (39.9-40.8), anorexia, petechial bleeding, icterus, and hemoglobinuria. Anemia, which is associated with pallor of the mucosal membrane, was observed in 46 sheep. Blood samples were collected via the jugular vein and blood smear staining was performed with Giemsa solution 5%. Microscopic examination in the immersion objective (X100) revealed *Piroplasms* in the same 46 sheep. The other 46 sheep without any clinical or paraclinical signs of babesiosis were selected as the healthy group. Blood samples with and without EDTA were centrifuged at 3000 RPM for 10 minutes at 4°C. MDA and PON activity were detected using the Satoh and Furlong method (spectrophotometer, model Cecil,Italy), respectively. T3 and T4 were measured in serum using a Roche Co. Elecsys 2010 in accordance with the electrochemiluminescence method; GL, TTP, cholesterol, HDL, and LDL (Pars azmoon Co. kits, Tehran, Iran) levels were evaluated in plasma using a Hitachi-917 Auto analyzer (Japan). SOD, GSH-Px, and TAC were measured in isolated and lysed red blood cells (by auto-analyzer, Alcyon-300, USA), (Ransod® and Ransel kits, Randox laboratories Ltd. G.B) and CAT was measured manually according to a previously described method, Abei. Statistical analysis was performed for all of the data completed during the study. The Mean ± SD and the determination of variation between the data points were carried out with Student’s t-test with SAS v9.1 (SAS Institute Inc., Cary, NC, USA). The significance level was specified at P<0.01.

**RESULTS**

All of the altered parameters are presented in Table 1. A significant elevation of MDA (P<0.01) was observed in the infected sheep compared to the healthy ones. In contrast, the level of antioxidant enzymes activity (involving CAT, GSH-Px, TAC or PON) was reduced in sheep with babesiosis compared to the healthy group (P<0.01). No changes in SOD activity were observed. In the case of thyroid hormones, decreases (P<0.01) in T3, T4, lipid profiles, and TPP were observed in infected sheep. Finally, glucose levels were increased in sheep with babesiosis as compared with the healthy group.

**DISCUSSION**

Lipid peroxidation plays an essential role in erythrocyte oxidation and anemia. Several studies have indicated
the importance of lipid peroxidation in blood parasitic diseases. Various others have reported babesiosis-induced MDA elevation in domestic animals such as, sheep naturally infected with Babesia ovis. The same authors demonstrated the important role of B. ovis in the induction of oxidative damage to RBCs and anemia. Moreover, Saleh reported decreases in T3, T4, free T3 and free T4 in dogs and cause lipid peroxidation in crossbred cattle naturally infected with Babesia ovis. Babesia gibsoni infections in sheep are known to result in lipid peroxidation via unknown mechanism(s).

Several studies demonstrated diminishing of antioxidant enzymes (SOD, CAT, GSH-Px, TAC) in hemoprotozoan diseases in domestic animals which is in accordance with our study except for SOD activity. Chaudhuri et al. reported a significant elevation of CAT and SOD in canine babesiosis compared with non-babesiosis dogs, which is also in contrast to the results presented here.

PON is a hydrolase that plays a major role in xenobiotic metabolism such as involving in the detoxification of organophosphorus compounds and inhibiting lipid peroxidation. Various studies have reported an association between PON activity in humans, but reports in the context of veterinary medicine are scarce. PON levels were found to be low in this study. The following represent potential reasons for the decrease: 1) A protective role of PON in the peroxidation of lipids and inhibition of the harmful effects of MDA. 2) Decreased HDL levels. Various studies have reported an association between the N-terminal of paraoxonase and apolipoprotein A1 (Apo A1) of HDL. Hence, a decrease in PON might be simultaneously correlated with HDL decreases.

Hypothyroidism is the most prevalent thyroid disturbances in small ruminants and causes disorders that reduce the animal’s ability to defend against infections and render it vulnerable to ketosis. Diverse factors such as, iodine deficiency and chronic disease can induce hypothyroidism. Certain situations may cause hypothyroidism without clinical signs, as is typical of secondary euthyroid conditions: for example, Pituitary-Thyroid (HPT) axis disorders and the possible effects on hormone-binding proteins in serum, disturbances in hormone metabolism, disorders in the peripheral distribution of hormones, the inhibition of hormone synthesis and an increase in the release of cytokines (e.g., interleukin-1, interleukin-2, interferon gamma and tumor necrosis factor a) that may be caused anorexia as well as the inhibition of secretion by the glandular portion of the thyroid. Issi et al. reported low levels of free T3, free T4, total T3 and total T4 in the pre-treatment of cattle naturally infected with Theileria annulata. A similar study performed by Badiei et al. in crossbred Holstein cattle naturally infected with Theileria annulata revealed low levels of T3, T4, free T3 and free T4. The authors postulated a paramount effect of anorexia.

Kumar et al. reported decreases in T3 and T4 in dogs with concurrent Erlichia canis and Babesia gibsoni infections and hypothesized that the body adapts by decreasing cellular metabolism during the period of disease. In recent
studies the levels of T3 and T4 were low. It is likely that certain factors have induced diminished levels in sheep with babesiosis. These factors include low iodine levels in soil and food, cytokines elevations due to disease that may cause anorexia and thyroid activity suppression, deficiencies of trace elements (e.g. selenium), and bodily adaptations such as decreased cellular metabolism during periods of disease as well as seasonally induced alterations.

Thyroid hormones influence lipid and lipoprotein metabolism in diverse species such as sheep, goat, horse and camel. Moreover, thyroid hormones increase lipolysis in adipose tissue and stimulate lipogenesis and the associated enzyme activity. The relationship between thyroid hormones and cholesterol remains to be elucidated. Notably, triglyceride levels do not correlate with cholesterol or thyroid hormone levels in camels and goats. Barrera et al. demonstrated the inverse correlation of triglycerides, cholesterol and phospholipid concentrations with hyperthyroidism in stricken Nubian goats. Santi et al. reported cholesterol decreases resulting from the catabolic effect of liver mediated by thyroid hormones. Ibrahim et al. demonstrated the inverse association between hypothyroidism and total cholesterol, triglyceride and LDL -cholesterol. Conflicting data exist regarding the relationship between thyroid function and cholesterol and triglyceride levels. We observed decreases in all aspects of the lipid profile, as reported by Bartley. Various factors may have affected this decrease, such as hypothyroidism, malnutrition and/or seasonal effects.

Thyroid hormones are considerable mediators of glucose metabolism and their roles in provoking glucose utilization have been established. Forhead et al. described the effects of thyroid hormones in decreasing liver glycogen storage during late pregnancy in the ovine fetus and suggested a possible influence of hypothyroidism and the related difficulties with euglycemia regulation. Keller et al. reported hypoglycemia in many dogs that suffered from babesiosis and indicated the importance of prompt hypoglycemia detection in canine babesiosis. The significant increase in glucose levels in these animals compared with the non-babesiosis group observed in this study may have been due to disease - the stress induced and/or intra-vascular hemolysis.

Infected animals exhibited a decrease in TPP when compared to healthy ones. Several studies have demonstrated TPP decreases in dog and sheep with babesiosis. Moreover, Barrera et al. suggested that TPP decreases may be due to anorexia in horse babesiosis. Our results generally support this assertion.

In conclusion, on the basis of similar published studies in various species of domestic animals, the results presented here suggest that oxidative stress is a common complication of ovine babesiosis. Moreover, low levels of T3 and T4, which denote hypothyroidism may exacerbate oxidative stress. Thus, it can be concluded that the concurrent occurrence of oxidative stress and hypothyroidism may negatively impact farm animals and thereby meat or dairy production.

REFERENCES


