

Electrophoresis profile of serum protein analysis from goat naturally infected with Theileriosis

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Abstract

Blood parasites represent an important causes of loss to goat production and the main clinical finding are anemia, ictruse of mucous membrane and enlargement of pre scapular lymph node and reduced growth rates . the present research aimed to determine the changes in the serum proteins of goat infected with theileriosis. Blood samples were collected from 20 Goats naturally infected with theileriosis all infected show ictrus of mucus membrane and enlargement of lymph node. Blood samples were also collected from 20 non infected goats as controls group. Serum were used to determination of total protein by colorimetric assay and protein fractions were separated by electrophoresis. The results were compared using an unpaired two-tailed T test. The serum levels (g/dl^{-1}) of theileriosis infected goats and control goats were respectively: total proteins (5.20 ± 1.17) and (7.06 ± 1.10), α -globulins (0.57 ± 0.23) and (0.39 ± 0.10), β -1 globulins (0.54 ± 0.22) and (0.45 ± 0.11), β -2 globulins (0.25 ± 0.18) and (0.26 ± 0.13), γ -1 globulins (0.95 ± 0.37) and (0.99 ± 0.45), γ -2 globulins (0.56 ± 0.25) and (0.22 ± 0.11). There statistically result significant differences for total protein, albumin, α -globulins, γ -2 globulins infection with theileriosis in goats intense hypoproteinemia, hypoalbuminemia increased α and γ -2 globulins and decreased albumin/globulin (A/G) ratio.

Keywords: goat, theileria, theileriosis, electrophoresis, albumin, biochemistry serum protein
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تقييم بروتينات مصلى الدم للماعز المصاب بحمى القراد باستخدام جهاز الترحيل الكهربائي

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الخلاصة

تعتبر طفيليات الدم من أهم أسباب الخسارة في إنتاج الماعز والعلامات السريرية الرئيسية هي فقر الدم، تضخم الغدد اللمفية أمام لوح الكتف أضافه لانخفاض معدلات النمو. صمم هذا البحث لتحديد التغيرات التي طرأت على بروتينات مصلى الدم المصاب بحمى القراد. حيث تم جمع عينات دم من 20 ماعز مصابة بشكل طبيعي بالحمى القرادية وكان هذا واضح من خلال العلامات السريرية الواضحة للمرض وكذلك تم جمع عينات الدم من 20 ماعز غير مصابة كمجموعة مقارنة. تم استخدام جهاز الترحيل الكهربائي لقياس نسبة البروتين الكلي والألبومين والكلوبولين. وتمت مقارنة النتائج مع المجموعة السليمة باستخدام التحليل الإحصائي وكانت مستويات البروتين في الماعز المصابة والماعز السيطرة على التوالي: البروتين الكلي (5.20 ± 1.17)، (0.99 ± 4.80) الفا-كلوبولين (0.57 ± 0.23)، (0.10 ± 0.39) بيتا - 1 كلوبولين (0.22 ± 0.54)، (0.11 ± 0.45) بيتا - 2 كلوبولين (0.54 ± 0.22)، (0.13 ± 0.26) غاما - 1 كلوبولين (0.37 ± 0.95)، (0.45 ± 0.99) غاما - 2 كلوبولين (0.25 ± 0.18)، (0.25 ± 0.56)، (0.11 ± 0.22) توجد اختلافات مهمة إحصائياً في المستوى الكلي للبروتين والزلزال والكاما - 2 كلوبين ونسبة الزلال إلى الكلوبيولين الكلي حيث أنها أقل في الماعز المصابة بحمى القراد من الماعز السليمة.

الكلمات المفتاحية: ماعز، مرض حمى القراد، حمى القراد، الترحيل الكهربائي، البومين، كيمياء حياتية، البروتين المصلى.

Introduction

Theileriosis are those tick-borne protozoan diseases associated with *Theileria* spp. In cattle, sheep and goats as well as in wild animals (1) It is represent an important cause of disease and loss of production in goats from tropical and sub tropical regions (2). The infection by theileriosis in goats is responsible by interference on serum protein including decrease in total proteins and albumin levels (3) goats infected by theileriosis show decrease in α and β - globulin levels (4) and Albumin/Globulins ratio and increase in γ -globulin levels (5). Serum electrophoresis is a technique of laboratory diagnosis that provides different charged particles. It is passed on movement of charged particles through a solution when subjected to an electrical field (6,7). Separation of serum protein fraction is very important for the diagnosis of different disease like liver disorders, acute inflammation and proliferative cases tissue damage like trauma and many internal parasite (8). The purpose of this study is to assess the diagnostic significance of serum protein electrophoresis in infected goat with theileriosis and comparing protein electrophoresis with other healthy animals.

Materials and Methods

- **Animals and Blood sampling:** A total of 20 goat naturally infected with theileriosis aged from (1-5) years of age and from both sexes, were examined from local breed and the different region were included in this study. A total of 40 goats was divided to naturally infected with theileriosis $n=20$ and healthy goats $n=20$ diseased animals show increase in the body temperature and icterus of mucus membrane as main clinical signs and theileriosis infection was confirmed by blood smears were prepared from jugular vein and stained with 5% giemsa solution then examined in the immersion objective $\times 100$. Smears were investigated on the microscopically. Piroplasma forms of theileriosis the erythrocytes in smears were evaluated in smears. Blood samples were collected by puncture of the jugular vein in all goats into sterile tubes without anticoagulant. after collected 5 minutes at room temperature 22°C the serum was carefully harvested centrifuged at room temperature 22°C for 5 minutes at 50 rpm and stored at -20°C until analysis.
- **Serum Protein Electrophoresis:** Basically, total protein concentrations have analysed using the biuret method (9). The serum protein fractions were separated using the Helena Lab-Titan III® Serum Protein Electrophoresis device (Cat No. 3023), Helena Lab -Titan III Cellulose acetate cards and Electra HR Buffer (Cat No. 5805) tampon solutions.
- **Statistical analysis:** The data between control and diseased animals were analyzed with a One Way Variance analysis and Statistical analysis were done by using SPSS.(10). was applied for multiple comparisons. Differences were considered as significant when P value was less than 0.05.

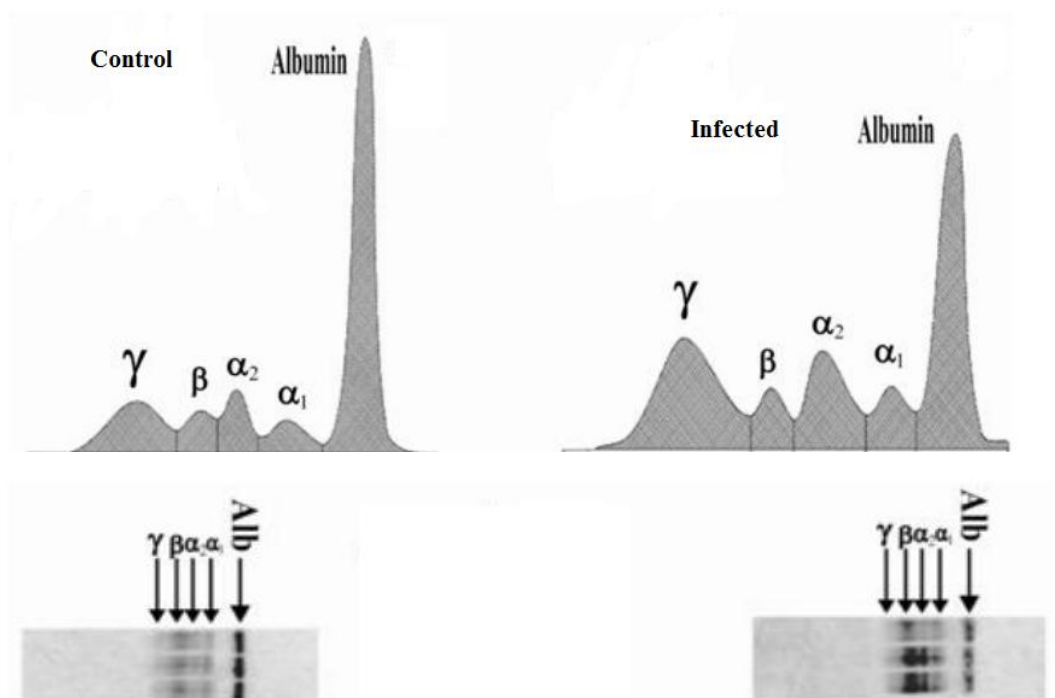
Results

Goats from the group naturally infected with theileriosis presented anemia and icterus of M.M. and weakness. Control goats were clinically healthy with good body condition score. The serum protein levels in (g/dl) of theileriosis infection and control goats are shown in (Table 1, Fig. 1). Goats infected with theileriosis had significantly lower ($p<0.05$) total protein and albumin levels and albumin/globulin ratio and significantly higher ($p<0.005$) levels of α - globulins and γ_2 globulins than controls. No significant differences were found in levels of total globulins, $\beta_{1,2}$ and γ_1 globulins.

Table (1) Serum levels of proteins in goats infected with theileriosis and non infected control group

Components	Infected goats n.(20)	Control goats n.(20)
Total proteins	5.20 ± 1.17*	7.06 ± 1.10
Albumin (g dl ⁻¹)	2.27 ± 0.93*	4.85 ± 0.99
Globulins (g dl ⁻¹)	2.83 ± 0.75	2.25 ± 0.70
α globulins (g dl ⁻¹)	0.57 ± 0.23*	0.39 ± 0.10
β-1 globulins (g dl ⁻¹)	0.54 ± 0.22	0.45 ± 0.11
β-2 globulins (g dl ⁻¹)	0.25 ± 0.18	0.26 ± 0.13
γ-1 globulins (g dl ⁻¹)	0.95 ± 0.37	0.99 ± 0.45
γ-2 globulins (g dl ⁻¹)	0.56 ± 0.25*	0.22 ± 1.11
Albumin/ Globulins ratio	0.89 ± 0.50*	2.37 ± 0.80

* p<0.05.

**Fig. (1) serum levels of proteins in goats infected with theileriosis and non infected control group.**

Discussion

Theileriosis infected are more seen frequently in the summer due to presences of active ticks in summer. It affects cattle, sheep and goats and cause economic losses and anemia and ictruse of m.m and enlargement of prescabular lymphnod (11). The present study showed that proteinemia was detected in the infected goat and confirmed previous observation in infected of theleriosis in cattle which cause reduction of serum levels of total protein and all fractions but without change in albumin/globulins ratio (12, 13, 14). In this study disagree with (15, 16) which show that serum total protein concentration did not change significantly in cattle and calves with natural or experimental theileriosis (17). reported that hypoproteinemia was likely due to hypoalbuminaemia and hypoglobulinaemia probably induced by liver failure. Decrease in albumin concentration was reported in various protozoan and internal parasit; babesia, fasciola (18, 19, 20) and was associated to economic losses, weakness in muscles and disorder in immune system (21). And this study also presented increase in γ globulin levels in the more infected compared with the newly infected because more severe damage tissue

and it was observed in this study that goats infected with theileriosis present abnormalities in serum proteins, characterized by intense hypoproteinemia, hypoalbuminemia increase in α and γ -2 globulins and decreased A/G ratio. Physiologic factors affect serum protein levels including temperature season (22). And blood samples were collected at same season without great variation of temperature and humidity and in the chronic cases show increased loss of large amount of serum proteins because disorder synthesis of albumin and this result agree with (23). Which lead to hypoalbuminemia is decrease production of albumin as a result of acute phase response which is associated with elevated globulin production. albumin is considered to be a negative acute phase protein because it decrease in concentration in response to inflammation (24). **Conclusion:** In this study the infection of goats by theileriosis lead to severe hypoproteinemia, hypoalbuminemia, increased alpha and gamma-2 globulins and decreased A/G ratio.

References

1. Coetzer, J. A. W. & Tustin, R. C. (2004). Infectious diseases of livestock. 2nd ed., Vol. 1, PP.448-501.
2. Radostits, O. M.; Gay, C. C.; Hinchcliff, K. W. & Constable, P. D. (2000). Veterinary Medicine. 10th ed. A textbook of the disease of cattle, sheep, goats, Saunders Company Ltd., London, New York. PP. 1328-1329.
3. Opara, M. N.; Udevi, N. & Okoli, I. C. (2010). Hematological parameters and Blood Chemistry of Apparently Healthy west African Dwarf (Wad) Goats in Owerri south Eastern Nigeria. New York Science J., 8: 68-72.
4. Ismael, A. B.; Swelum, A. A.; Khalaf, F. & Abouheif, A. M. A. (2013). Clinical, Haematological and Biochemical Alterations Associated with an Outbreak of Theileriosis in Dromedaries (*Camelus dromedarius*) in Saudi Arabia. Pak. Vet. J., 34(2): 209-213.
5. Mert, N., Day, H. & October, S. (1997). Different breeds sheep determination by electrophoresis of serum protein level. Ü. Vet. Fake. Bull., 8: 28-30.
6. Thomas, J. S. (2006). Overview of plasma proteins in sahalms veterinary hematology, Feldman, B.F., JG, PP. 891-898.
7. Eckersall, P. D. (2008). Portions in clinical biochemistry of domestic animals. PP. 117-155.
8. Jain, N. C. (1986). Schalm Veterinary hematology. 4th ed. Lea and Fibiger, Philadelphia, PP.610-612.
9. Tiftik, A. M. (1996). Klinik Biyokimya. Mimoza Yay., PP. 155-158.
10. SPSS. (2008). Statistical Package for the social science, version 17 (Win/Mac/linux), users guide SPSS Inc., Chicago, USA. Website, <http://www.spss.com>.
11. Guo, S.; Yuan, Z.; Wu, W.; Ma, D. & Du, H. (2002). Epidemiology of ovine theileriosis in Ganan region, Gansu province, China. Parasitol.
12. Hussein, H. A.; Abd- El-Salam, M. N. & Karama, M. H. (2007). Theileriosis and babesiosis in cattle: haemogram and some biochemical parameters. XIII International Congress of ISAH Tartu, Estonia, PP. 143-150.
13. Singh, A.; Singh, J.; Grewal, A. S. & Brar, R. S. (2001). Studies on some blood parameters of crossbred calves with experimental Theileria Annulata infections. Vet. Res. Comm., 25: 289-300.
14. Soulsby, E. J. L. (1986). Helminthe, Arthropods and Protozoa of domesticated animals. 7th ed. Philadelphia, London, Toronto. PP.718-719.

15. Sandhu, G. S.; Kondal, J. K.; Singh, J. & Brar, R. S. (1998). Haematological and biochemical studies on experimental *Theileria annulata* infection in crossbred calves. *Vet. Res. Comm.*, 22: 347-354.
16. Grewal, A. S. & Singh, A. (1998). *Theileria annulata* infection in crossbred calves. *Vet. Res. Comm.*, 27: 125-139.
17. Sivakumar, T.; Hayashida, K.; Sugimoto, C. & Yokoyama, N. (2014). Evolution and genetic diversity of *Theileria*. *Infection, Genetics and Evolution*, 27: 250-263.
18. Dimopoulos, G. T. (1970). Plasma proteins in clinical biochemistry of domestic animals, Kaneko, J. J., Cornelius C.E. (eds), Second Edition, Academic Press, New-York, London, Vol. 1, PP. 97-129.
19. Meyer, D. J. & Harvy, J. W. (1998). *Veterinary laboratory medicine*. 2th ed. W.B. Saunders Co., PP. 157-199.
20. Erstad, B. L. (1996). Viral infectivity of albumin and plasma protein fraction. *Pharmacotherapy*, 16: 996-1001.
21. Wijnen, P. A. & Van Dieijen-Visser, M. P. (1996). Capillary electrophoresis of serum proteins. Reproducibility, comparison with agarose gel electrophoresis and a review of the literature. *Eur. J. Clin. Chem. Clin. Biochem.*, 34: 535-545.
22. Orag, G. K. (2002). *Vet Lab. Med. Clinical Biochemistry and Hematology*. 2nd ed., Blackwell Sci. PP. 74-130.
23. Thomas, J. S. (2006). Overview of plasma proteins in sahalms veterinary hematology, Feldman, B. F., JG, PP. 891-898.
24. Sharma, D. K.; Chuhan, P. P. P. S. & Agrawal, R. D. (2001). Changes in the levels of serum enzymes and total protein during experimental haemonchosis in goat. *Small Rumin. Res.*, 42:119-123.