j.Egypt.net.med.Assoc 78, no 4. 601 – 608 (2018)

STATUS OF SOME OXIDATIVE STRESS BIOMARKERS AND LABORATORY FINDINGS IN SHEEP WITH GASTROINTESTINAL PARASITES

By

Shimaa Ghanem Yehia

Departement of Internal Medicine and Infectious Diseases, Faculty of veterinary Medicine, Cairo University, Cairo, Egypt.

Corresponding author email:shimaaghanem2013@gmail.com

ABSTRACT

PGE is a principal cause of losses in sheep manufacture. The present study aimed to assess oxidative stress and serum biochemical status in sheep with PGE. Fifteen adult sheep infested with PGE and seven control adult sheep were involved in this study, clinical signs and fecal examination findings were recorded, as well blood samples were taken to investigate serum biochemical variations and oxidative stress biomarkers status. The most consistent clinical signs recorded in affected sheep include weight loss, diarrhea, and anemia with pale conjunctival mucous membranes and generalized weakness. Significant increase in Hydrogen peroxide activity along with significant decrease in total antioxidant capacity and Catalase activities was detected in diseased group. Significant decrease in total protein, albumin, Zinc and iron was observed in diseased group. In the current investigation, oxidant/antioxidant imbalance along with the alterations in biochemical findings was detected in sheep with parasitic (GE). These findings support that PGE associated with oxidative stress and this procedure may be related to the resulting clinical signs.

Key words:

Ovine, nematodes, minerals, oxidative load, gastrointestinal.

INTRODUCTION

Sheep manufacture is an essential part for meat resource and consumption of human in Egypt and services to improve the rural areas. In Egypt total sheep population was estimated at 5.5 million heads, most of them allocated in Nile-Delta **(Statistics of Live Stocks, 2011).** Helminthosis and nutritional aspects are the two chief issues that may result in depletion of sheep production **(Dorny *et al.*, 1996)**. Many authors stated that certain sheep GI parasites are of public health significance and they were implicated in zoonotic transmission to human

either by direct contact with sheep feces or indirectly through consumption of polluted food or water **(Ralph *et al.*,2006; Byomi *et al.*,2010;Feng and Xiao,2011)**.Clinical manifestations of PGE, as diarrhoea, and mortalities occur mainly in lambs **(Sargison, 2011).** Sub-clinical signs, as weight loss and reduced growth, are possibly more significant to a modern livestock manufacture targeting for enhancement of production **(Taylor, 2009).** Oxidative stress is a consequence of the increase in production of free radicals and reactive oxygen species (ROS), and/or the reduction in antioxidant protection, results in damage of DNA, enzymes and membranes and prompts deviations in the activity of the immune system and in the structure of basic biopolymers which, in sequence, may be related to several health ailments **(Trevisan *et al.*, 2001).** In a number of investigations, the amount of reactive oxygen radicals increased in cells of hosts infected with various species of parasites **(Abd Ellah, 2010).** The purpose of this study is to define the laboratory alterations in parasitic gastroenteritis infected sheep with special emphasis on oxidant-antioxidant status associated with this disease.

MATERIAL AND METHODS

Animals:

Fifteen adult native breed sheep infested with PGE (n =15) with age range from one year to 3 years old were evaluated in this study. The present investigation was carried out in the Teaching Hospital of Faculty of Veterinary medicine and Infectious Diseases, Cairo University; all the fifteen sheep were confirmed positive to have PGE on basis of history, clinical signs and fecal examination. Control sheep (n = 7) with the same age were involved. Selected control group were thoroughly clinically examined, fecal samples were taken from each control sheep and examined microscopically to ensure they are parasitologicaly free. The criteria for inclusion of control group depended on fecal examination, and absence of clinical signs. Each animal was subjected to comprehensive clinical examination and clinical signs were recorded.

Blood sampling and fecal examination:

Blood samples were obtained from jugular vein in EDTA- containing tubes, heparin tubes and plain tube to separate serum and plasma samples .The blood in plain tubes were conserved in slanted position for about 2 h and then refrigerated at 4^0C overnight for separation of serum. Concentration floatation technique using saturated salt solution and microscopic examination were carried out according to **Soulsby (1982).**

Estimation of serum biochemical parameters:

Serum samples were used to determine total protein, albumin, cholesterol, BUN, creatinine, zinc and iron (Bio-Diagnostic Company Egypt, Spectrum Diagnostic Egypt). Assessment of these parameters was performed manually using spectrophotometer (APEL, PD-303S, Japan) as stated by manufacturer instructions.

Estimation of oxidant-antioxidant status:

Plasma samples were used for assessment of catalase, total antioxidant capacity and hydrogen peroxide using respective test kit (Bio-Diagnostic Company-Egypt). Estimation of these parameters was done manually using spectrophotometer (APEL, PD-303S, and Japan) according to manufacturer instructions.

Statistical analysis:

Results of diseased sheep were compared to control data, calculation of mean ± SEM, and data judgment were done using student t-test (STATISTICA for Windows, version 5.1., Stat Soft, Inc.) .$P < 0.05$ considered significant.

RESULTS

The most consistent clinical signs recorded in diseased sheep include weight loss, diarrhea, and anemia with pale conjunctival mucous membranes and generalized weakness.

The examination of fecal samples revealed the presence of parasitic eggs on microscopic examination of diseased group.

Status of oxidative stress biomarkers is shown in (Table 1). Significant increase in Hydrogen peroxide activity along with significant decrease in total antioxidant capacity and Catalase activities were observed in diseased group. Serum biochemical changes are recorded (Table 2). Significant decrease ($P < 0.05$) in total protein, albumin, Zinc and iron was observed in diseased group. No statistical significance were recorded in BUN, cholesterol and creatinine.

Table (1): Status of oxidative stress biomarkers in healthy and diseased groups. P < 0.05 considered significant.

parameters	Healthy group	Parasitic gastroenteritis group	P value
Hydrogen peroxide(mM/L)	0.209 ± 0.096	0.735 ± 0.171	0.0025
Catalase (CAT) U/L	236.11 ± 16.04	140.90 ± 11.6	00.041
Total antioxidant capacity (TAC) mM/L	0.84 ± 0.31	0.40 ± 0.14	0.032

(Table 2): Serum biochemical evaluation in diseased group compared with the apparently healthy group. P < 0.05 considered significant.

parameters	Healthy group	Parasitic gastroenteritis group	P value
Zinc (µg/dl)	63.85 ± 2.92	43.88 ± 3.19	0.0047
Iron (µg/dl)	101.61 ± 8.59	69.90 ± 9.26	0.0327
Total protein (g/dl)	5.60 ± 0.27	3.63 ± 0.15	0.00043
Albumin (g/dl)	2.73 ± 0.26	1.77 ± 0.14	0. 015
Cholesterol (mg/dl)	56.50 ± 7.3	59.87 ± 9.55	0.38
Blood urea nitrogen (mg/dl)	13.88 ± 2.56	12.29 ± 2.06	0.25
Creatinine (mg/dl)	1.44 ± 0.36	1.59 ± 0.13	0. 31

DISCUSSION

Helminthiasis, especially parasitic gastroenteritis, pose a serious health hazard and a restriction to the efficiency of small ruminants due to the accompanying morbidity, mortality, cost of treatment and control procedures **(Nwosu *et al.,* 2007)**. In the present study, anemia and pale mucous membranes were recorded in all the cases as the parasites in the gut depresses absorption and cause various deficiencies resulting in anemia. In some cases, it may be anaplastic due to exhaustion of bone marrow occur due to chronic hemorrhages in the gut via blood sucking parasites **(Sastry 2001).** Parasitic gastroentritis is a chronic disease which

leads to chronic anaemia, weakness, emaciation and progressive weight loss **(Kahn and Line, 2005)**. The reduction in total serum protein and albumin detected in the current study agreed with the earlier reports of **Ashok *et al.* (2005)**, **Jas *et al.* (2008) and Ahmed *et al.* (2015).** Excessive amounts of serum proteins leak into the parasitized intestines as a consequence to increase mucosal permeability is correlated with hypoalbuminemia and therefore the resultant hypoproteinemia in sheep infected by parasitic gastroenteritis **(Chiejina 1987)**. The decrease in total serum protein and serum albumin is a frequent finding of PGE and the resultant protein losing enteropathy **(Soulsby1982)**. In the current study, zinc concentrations were significantly affected by parasitic infestation when compared with control group. This result agreed with the earlier reports of **Siddiqui and Cameron (2005).** Zinc deficiency has been reported to decrease in other intestinal parasites **(Ertan *et al.,* 2002),** the poor intake and impaired absorption of zinc might be factored in zinc deficiency **(Marilyn and Kristine, 2000)**; Analysis of the serum iron revealed significant reduction in diseased group. This reduction of serum iron level in infected sheep could be occur due to the prolonged erythropoiesis to compensate for blood loss leading to reduction of iron stores **(Abakar *et al.* 2000)**. Trace elements have an essential purpose in the inhibition of free-radical-encouraged injury to tissues to keep animal health and productivity **(Oteiza *et al.* 1995)**. low serum trace elements levels have been stated in parasitic diseases **(Tasci *et al.* 1995).**The Depression in serum trace elements concentrations in parasitized animals might attributed to malnutrition and alteration of nutritional absorption **(Seyrek *et al.* 2009).** Oxidative stress is a recent field of investigations in ruminant medicine and has been associated in various disease processes **(Celi, 2011).** Host responds against parasites by a number of ways including the production of reactive oxygen species (ROS) by immune effector cells, which play a role in killing or ejection of parasites from their host and thereby prevent the establishment of infection **(Ben-Smith *et al*.,2002).** Free radicals are formed endlessly by regular metabolic processes, but their frequency of production increases during definite parasitic infestations **(Heidarpour *et al*.,2012)** .The increase in oxidant marker (hydrogen peroxide) together with reduction of antioxidant markers (total antioxidant capacity and catalase), are indicators of oxidative stress. Catalase showed significant decrease in diseased sheep in the present study. Catalase is responsible for H2O2 elimination and thus it afford defense against destructive influence of H2O2 leading to rise RBCs lifetime **(Baghshani *et al* 2011).** Plasma level of ROMs "reactive oxygen metabolites" is a reflection of free radical production **(Miller *et al.,* 1993).** ROMs is a

term that contains not only oxygen-centered free radicals as superoxide anion and hydroxyl radical, but moreover some non-radical derivates of oxygen, as hydrogen peroxide, and hypochlorous acid **(Reilly *et al.*, 1991).** TAC can reveal the total antioxidant capacity of the serum which acts contrary to strong free radical reactions potently leading to oxidative impairment of biomolecules such as lipids, proteins, and DNA **(Kosecik *et al.* 2005).**

CONCLUSION

In the present study, the decreased concentrations of TAC and catalase coupled to the increased concentration of hydrogen peroxide may be related to decreased antioxidant capacity.It can be concluded from the present study that parasitic gastroenteritis in sheep induces changes in oxidant/antioxidant status. These alterations may contribute to the development of clinical signs.

REFERENCES

Abakar A D; El Amin, E A and Osman AY. (2000): Clinical Response to Experimental Haemonchus contortus Infection in Desert Lambs. The Sudan J. Vet. Res (1999 -2000) 16:1-10.

Abd Ellah, M.R. (2010): Involvement of free radicals in animal diseases. Comparative Clinical Pathology, 19: 615- 619.

Ahmed A,. Dar MA,. Bhat AA, Jena B, .Mishra G.K, Tiwari RP. (2015): Study on Haemato Biochemical profile in Goats suffering from Gastrointestinal Parasitism in Jaipur district of Rajasthan. J. Livestock Sci. 6: 52-55.

Ashok K., Vihan V.S., Rana, R. and Vinod K. (2005): Blood Biochemical changes in some important parasitic infestations in goats for clinical appraisal. Indian Journal of Small Ruminants 11: 156-160.

Baghshani H, Razmi GR, Yaghfouri S, Dezaki AA. (2011): Status of some oxidative stress biomarkers in sheep naturally infected with theileriosis. Res Opin Anim Vet Sci; 1:499 - 504.

Ben-Smith, A., Lammas, D.A. and Behnke, J.M. (2002): Effect of oxygen radicals and differential expression of catalase and superoxide dismutase in adult Heligmosomoides polygyrusduring primary infections in mice with differing response phenotypes. Parasite Immunol, 24: 119 - 129.

Byomi M, Samaha H, Zidan S. (2010): Epidemiological studies on some zoonotic enteric protozoa in different areas of Nile Delta. J Am Soc Minn Reclam; 5:199 -207.

Celi, P. (2011): Oxidative stress in ruminants. In: Mandelker, L. and Vajdovich, P. (editors), Studies on veterinary medicine, oxidative stress in applied basic research and clinical practice. 1st (ed.), Humana Press, Pp: 191-231.

Chiejina S N. (1987): Parasitic gastroenteritis in cattle and small ruminants: pathogenesis diagnosis and treatment. Zariya Vet., 2 (2): 45 - 64.

Dorny, P.; Balubara, A.; Iskander, M. and Pandey, V. S. (1996): Helminth infections of sheep in North Sumatra, Indonesia. Vet. Parasitol. 61: 353-358.

Ertan, P., Yereli, K., Kurt, O.; Balcioglu, I.C. and Onag, A. (2002): Serological levels of zinc, copper and iron elements among Giardia lamblia infected children in Turkey.Pediatr.Int., 44 (3): 286 - 288.

Feng Y and Xiao L.(2011): Zoonotic potential and molecular epidemiology of Giardia species and giardiasis. Clin Microbiol Rev; 24:110 - 40.

Heidarpour .M, Mohri .M, Borji .H and Mohandas E. (2013): Oxidant/antioxidant balance and trace elements status in sheep with liver cystic echinococcosis. Comp Clin Pathol., 22, (6): 1043-1049.

Jas, R., Datta. S. and Ghosh, J.D. (2008): Haemato-biochemical impact of gastrointestinal nematodosis in Bengal goat. Journal of Veterinary Parasitology 22: 21 - 26.

Kahn CM, and Line S. (2005): The Merck Veterinary Manual 9th Ed., Merial, USA.

Kosecik M, Erel O, and Sevinc E, Selek S. (2005): Increased oxidative stress in children exposed to passive smoking. Int J Cardiol 100:61- 64.

Marilyn, E.S., and Kristine, G.K. (2000): Zinc deficiency impairs immune responses against parasitic nematode infections at intestinal and systemic sites. J Nutr, 130 (Supple): 1412-1420.

Miller, J.K.; Brzezinska-Slebodzinka, E.; and Madsen, F.C. (1993): Oxidative stress, antioxidants, and animal function .Journal of Dairy Science, v.76, p.2812-2823.

Nwosu, C O, Madu P P, and Richards W S. (2007): Prevalence and seasonal changes in the population of gastrointestinal nematodes of small ruminants in the semi-arid zone of North-Eastern Nigeria. Veterinary Parasitology, 144: 118-124.

Oteiza PI, Olin KL, and Fraga CG, Keen CL. (1995): Zinc deficiency causes oxidative damage to proteins, lipids and DNA in rat testes. J Nutr .125:823 - 829

Ralph A, O'Sullivan V, Sangster C, Walker C. (2006): Abdominal pain and eosinophilia in suburban goat keepers – trichostrongylosis. Med J Aust; 184:467-9.

Reilly, P.M.; Schiller, H.J.; Bulkley, G.B. (1991): Pharmacologic approach to tissue injury mediated by free radicals and other reactive oxygen metabolites. The American Journal of Surgery, V.161, p.488-503.

Sargison, N.D. (2011): Pharmaceutical control of endoparasitic helminth infections in sheep. Veterinary Clinics of North America: Food Animal Practice, 27 (1), p. 139 -156.

SastryGA.(2001): Stomach worm disease. In:Veterinary Pathology.7thedition.CBS publishers, New Delhi, 2001; 731-732.

Seyrek K, Karagenc T, Pasa S, and Kıral F, Atasoy A. (2009): Serum zinc, iron and copper concentrations in dogs infected with Hepatozoon canis. Acta Vet Brno 78:471 - 475.

Siddiqui, H U R and Cameron R D. (2005): Effect of Haemonchus contortus Infection and Nutrition on Glucose and Trace Elements. Int. J. Agri. Biol., Vol. 7, No. 2.

Soulsby, E.J.L.(1982): Helminths, Arthropoda and Protozoa of domesticated animals. 7th ed. Baillier, Tindal and Cassel, London.

Statistics of Live Stocks (2011): Economic affairs sector, Ministry of Agriculture and land reclamation, A.R.E. <http://www.agr-egypt.gov.eg/StudiesAll.aspx>; (In Arabic) [accessed 09.12.12].

Tasci S, Sengil AZ, and Altindis M, Arisoy K. (1995): The effect of zinc supplementation in experimentally induced Toxoplasma gandii infection. J Egypt Soc Parasitol 25:745-75.

Taylor, M. (2009): Changing patterns of parasitism in sheep. In Practice, 31, p. 474 - 483.

Trevisan, M., Browne, R., Ram, M., Muti, P., Freudenheim, J., Carosella, A.N. and Armstrong, D. (2001): Correlates of markers of oxidative status in the general population. American Journal of Epidemiology, 154: 348 -356.