

ALTERED OXIDATIVE STRESS BIOMARKERS IN ASSOCIATION WITH GASTROENTERITIS OF DOGS

By

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ABSTRACT

Gastroenteritis cases are commonly encountered in small animal practice. This study aimed to estimate oxidant-antioxidant status in gastroenteritis affected dogs. This study was applied on seventeen (17) dogs, 10 of them had sign of gastroenteritis and the reminders (7) were apparently healthy dogs enrolled as a control. The they undergo thorough physical, clinical examination and blood samples were withdrawn from each animal for estimation of hematologic alterations, SOD, GPx, Catalase, TAC, MDA, H₂O₂, cortisol, Zn, Cu, Cl, K and Fe levels. Reduction in hemoglobin, PCV, catalase and GPx along with significant elevation in SOD and cortisol were recorded. Gastroenteritis has an impact on oxidant-antioxidant equilibrium in dogs

Keywords:

Oxidative stress biomarker, Malondialdehyde, SOD, GPx, catalase, intestinal parasites.

INTRODUCTION

Gastroenteritis is a medical term means inflammation of the gastrointestinal tract (stomach and intestine); numerous causes have been implicated for example sudden change of diet, toxicity, or infection with virus, bacteria and parasite are among most common causes.

Signs are usually vomiting either accompanied with/without diarrhea, abdominal pain and consequently dehydration especially if fluid loss through vomiting and diarrhea persist for more than one day (**Tilley and Smith 2011**).

In young dogs, diarrhea accompanied by vomiting can be seen in association with parasitic causes, in which, *Toxocara canis* most common parasite affecting young animals and usually associated with digestive disturbance in puppies (**Merrill, 2012**).

Confirmation of diagnosis usually depends on microscopic detection from fecal sample **(Bellwood and Andrasik - Catton 2014)**.

Oxidative stress is a condition occurs when oxidant substances exceed antioxidant system capacity to scavenge **(Sies and Cadenas 1985)**. Free radicals or reactive oxygen species (ROS) are normally generated by cellular metabolism **(Dallaqua and Damasceno, 2011)**. Over production of ROS have reported in different diseases in dogs **(Kiral et al., 2005; Dimri et al., 2012)**. The second effector arm is antioxidant; which are substances that protect the body against toxic effect of the oxidants. Superoxide dismutase SOD, glutathione peroxidase (GPx), and catalase are enzymatic antioxidant considered as first line defense to suppress damage effect of oxidant substance **(Rubio et al., 2016 Aytakin et al., 2010)**.

This study aimed to estimate oxidant-antioxidant status in gastroenteritis affected dogs with parasitic burden.

MATERIAL AND METHODS

Animals:

Seventeen dogs were divided into two groups including control group and seven apparently healthy dogs) and diseased group including ten dogs presented with sign of vomiting and diarrhea). Both groups were subjected to physical and clinical examination. These were of different ages ranged between 2 months to 6 months and both genders. Clinical cases and control dogs were admitted to small animal clinic; faculty of Veterinary Medicine Cairo University and Alabasya pet's hospital.

Samples:

Fecal samples were collected from each animal and examined under microscope for confirmation of parasitic involvement according to **(Bellwood and Andrasik - Catton 2014)**. Blood samples were collected using EDTA and plain tubes. Blood samples with anticoagulant were used for hematological analysis using automated Hematology analyzer (Hemolyzer 3 pro, Germany). catalase and hydrogen peroxide estimated in separated plasma, erythrocyte lysate were used for estimation of SOD and GPx. Serum samples were used for determination of MDA, TAC, K, Cl, Zn, Cu and iron using specific commercial kits (spectrum diagnostics and Bio-diagnostic Egypt) manually using Spectrophotometer (APLE, japan). Cortisol was estimated in serum samples via (ADVIA Centaur CP Immunoassay system, Germany).

Statistical analysis:

Comparing control group with diseased group by using Student t-test (IBM SPSS Statistics v20), P value of ≤ 0.05 considered significant.

RESULTS

For clinical picture, the diseased dogs had signs of vomiting, diarrhea, anorexia and dullness. Alterations in hematologic values of diseased dogs compared to control dogs were recorded in (Table 1). Nonsignificant decrease in RBC, Hg, HCT, MCV, MCH, MCHC and Granulocytes along with non-significant increase in WBCs and Lymphocytes were recorded in affected dogs compared with control dogs.

Alterations in oxidant-antioxidant status are recorded in (Table 2). Significant increase in SOD and cortisol associated with significant decrease in GPx and catalase level were recorded. Decrease in TAC, Cu, Zn, iron and Cl, H₂O₂ and K were recorded, indicated no statistically significant changes. (Table 2).

Table(1): Hematological findings in apparently healthy dogs (control) and gastroenteritis diseased dogs.

Parameter	Control (n=7) Mean ± SE	Diseased (n=10) Mean ± SE	P value
RBC ($\times 10^{12}/l$)	7.17 ± 0.2	5.71 ± 0.55	0.2
Hb (g/dl)	17.3 ± 0.6	13.7 ± 0.9	0.2
HCT (%)	48.2 ± 2.04	36.9 ± 3.5	0.24
MCV (fl)	67.4 ± 1.8	64.8 ± 1.38	0.6
MCH (pg)	24.1 ± 0.5	23.1 ± 0.4	0.5
MCHC (g/dl)	35.9 ± 0.2	35.6 ± 0.4	0.1
WBC ($\times 10^9/l$)	13.67 ± 2.12	15.7 ± 8.4	0.1
LYM ($\times 10^9/l$)	2.4 ± 1.03	10.3 ± 7.6	0.08
MONO ($\times 10^9/l$)	3.1 ± 1.8	3.7 ± 1.3	0.5
GRAN ($\times 10^9/l$)	8.01 ± 3.00	4.8 ± 1.3	0.14

Table (2): oxidative stress biomarkers in apparently healthy dogs (control) and gastroenteritis diseased dogs.

Parameter	Control (n=7) Mean ± SE	Diseased (n=10) Mean ± SE	P value
SOD U/MI	14.68 ± 0.26	51.05 ± 13.63*	0.002
Gpx mu/ml	14.96 ± 0.21	9.06 ± 2.54*	0.01
Catalase (U/L)	596.8 ± 73.8	446.3 ± 20.9*	0.03
Hydrogen peroxide (m M/L)	0.0037 ± 0.001	0.0035 ± 0.002	0.8
Malondialdehyde (nmol/ ml)	3.44 ± 0.4	3.43 ± 0.7	0.08
Total antioxidant capacity (mM/L)	0.95 ± 0.07	0.75 ± 0.1	0.2
Cortisol ug/dl	0.43 ± 0.09	3.77 ± 0.81*	0.01
Cu (µg/dl)	127.85 ± 10.5	103.8 ± 6.9	0.5
IRON (µg/dl)	96.85 ± 5.0	81.37 ± 5.6	0.9
Zn (µg/dl)	115.2 ± 7.21	110.3 ± 16.3	0.1
CL (m M L)	108.2 ± 4.19	106.0 ± 2.8	0.3
K (m M L)	4.4 ± 0.3	4.3 ± 0.2	0.2

*Significant with (p ≤ 0.05).

DISCUSSION

Gastroenteritis is one of the major causes that initiates pet owner to visit veterinary clinic. Hematological alteration showed decrease in hemoglobin, PCV, the reduction in such values might be associated with presence of *Toxocara canis* (Mahadappa and Dey 2018) and a mild degree of anemia could be observed (Tvedten and Weiss 2000). The parasite is thought to consume blood from intestine and depleted important nutrient for body to produce RBCs (Koski and Scott 2003) and hence the reduction. Elevated Cortisol level was significantly increased indicating that stress condition was associated with stomach inflammation (Monika et al., 2018). Changes of minerals were recorded in this study with reduction of iron, zinc and copper in diseased dogs. The increased production of antioxidant might be correlated with low iron levels (Evans and Halliwell 2001); moreover, the effect of lipid peroxidation of erythrocyte wall was linked to with reduction in iron level (Ranjan et al., 2006). Cu and zinc are known to be cofactor for SOD synthesis, so their reductions might be associated with increased the consumption of synthesized SOD to counteract resultant oxidation process (Cristiana et al., 2014). Significant increase in SOD was recorded, SOD is considered as first line defense in antioxidant mechanism, it is important to catalyze the dismutation of superoxide radicals in to H₂O₂ (Dringen et al.,2005). Reduction of catalase was recorded in

diseases patients compared to control group Increase consumption of catalase to neutralize production of H₂O₂ by break it down to H₂O will resulted in decrease catalase level (**Clemen and Waller 1987**). Reduction in GPx was also recorded in this study. GPx is an important antioxidant enzyme which decrease the end products of lipid peroxidation and convert H₂O₂ to water (**Rotruck *et al.*, 1973**). Reduction in Gpx might be linked to increase utilization in converting H₂O₂ to water (**Taylor *et al.*, 1993**).

CONCLUSION

The present study study concluded that parasitic burden in young puppies cause stress manifested by elevation in cortisol along with alterations in oxidant-antioxidant balance and these alterations support notion that gastroenteritis caused by parasite associated with oxidative process

Acknowledgments:

Author likes to thank Prof Dr: Tahir Baraka internal medicine. Department of medicine and infectious diseases, faculty of Vet. Med, Cairo University.

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