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## Evaluation of altered erythrocytic oxidative stress indices in association with canine pyoderma

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### Abstract

Canine pyoderma is a major skin disease of dog that may lead to increased risk of oxidative stress. The following study was carried out in 40 dogs out of which twenty were pyoderma affected taken as Group 2 and twenty were healthy dogs (Group 1) treated as control. This study was done to estimate oxidative stress biomarkers (Lipid peroxidase, Super oxide dismutase, Catalase). Analysis of these erythrocytic oxidative stress indices revealed significantly high ( $p < 0.05$ ) Lipid peroxidase (LPO) value and significant low ( $p < 0.05$ ) Super oxide dismutase (SOD) and Catalase values at day 0 of experiment. Bacterial isolation and culture was done where Coagulase positive *staphylococcus aureus* species was isolated. In antibiotic sensitivity test azithromycin was found to be highest sensitive. After antibiotic therapy the altered oxidative stress indices are subsequently found to be normalised at day 30 of the experiment.

**Keywords:** Canine pyoderma, LPO, sod, catalase

### 1. Introduction

The skin which functions like a protective barrier to external elements, reflects the health condition of dog. Pyoderma is one of the most common dermatological problem in dogs. Dogs are more prone to pyoderma due to the unique characteristics of their skin consisting of a thin stratum corneum, lack of lipid plug in the hair follicles and high skin pH which possess a risk for bacterial invasion, subsequent growth and over colonization [1]. Bacterial pyoderma caused by gram positive bacteria (*Staphylococcus*) [2] which may be superficial, affecting the epidermis or surface or deeper structures of the skin characterized by pustules, papules, pruritus, and alopecia [3].

Reactive oxygen species (ROS) are a group of oxygen based free radicals which gets elevated during oxidative stress, causes biomolecular damage manifested by lipid peroxidation [4]. ROS-induced cellular damage can be prevented by antioxidants [5]. Antioxidants include high-molecular-weight antioxidant enzymes represented by glutathione peroxidase, superoxide dismutase, and catalase enzymes and antioxidant vitamins like vit-A, E, C [4].

### 2. Materials and Methods

#### 2.1. Ethical approval

The experimental procedures have been conducted in accordance with the guidelines laid down by the Institutional Ethics Committee.

#### 2.2 Area of study

The present study was carried out in the Teaching Veterinary Clinical Complex (TVCC), College of Veterinary Science and Animal Husbandry, OUAT, Bhubaneswar.

#### 2.3 Experimental design

Twenty healthy dogs were selected and grouped as Group 1 as healthy control. Another twenty dogs with clinical signs of pyoderma like erythema, alopecia, pruritus, papules, crusts, pustules, epidermal collarettes, Pus in skin etc were selected and taken as Group 2. Blood sample (5 ml) from each dog on day 0, day 15 and day 30 of treatment was collected in heparinised vials for preparation of RBC hemolysate for estimation of erythrocytic oxidative indices. The Group 2 dogs were treated with Azithromycin @ 10 mg/kg.b.wt once daily for three consecutive days in a week [6] for four weeks and topically mupirocin was applied till

scar appeared, whereas the Group 1 animals were given no treatment.

## 2.4 Parameters studied

Erythrocytic oxidative enzymes like SOD, Catalase and LPO were estimated from the 10% RBC haemolysate prepared from heparinised blood by the manual method using double beam UV-VIS spectrophotometer [7, 8].

## 2.5. Statistical analysis

All the data generated in the above experiments were statistically analyzed using SPSS (1996) computer package. For comparison of groups, Generalized Linear Model, ANOVA procedure and Duncan's multiple range tests were used [9].

## 3. Results

The results of oxidative stress parameters LPO, SOD and Catalase was shown in Table No 1. A significantly ( $p < 0.05$ ) higher LPO value  $1.53 \pm 0.06$  and significantly lower SOD  $0.69 \pm 0.02$  and Catalase  $0.54 \pm 0.03$  values were recorded in the Group 2 dogs when compared with the healthy control Group 1 dogs on day 0. Subsequently the value of LPO was found to be in a significantly lower ( $p < 0.05$ ) level  $1.08 \pm 0.05$  whereas the values of SOD and Catalase were found to in a significant higher level on day 15 then day 0. On day 30 the altered values of the above three parameters were found to be very much normalized than they were on day 0 of the experiment.

**Table 1:** Erythrocytic oxidative stress enzymes in different groups of different observation period

Parameters	Groups (n=20)	Mean $\pm$ SE		
		0 DAY	15 <sup>th</sup> DAY	30 <sup>th</sup> DAY
LPO (nmol /mg Hb)	G1	$0.71 \pm 0.02A$	$0.71 \pm 0.03A$	$0.72 \pm 0.02A$
	G2	$1.53 \pm 0.06cB$	$1.08 \pm 0.05bB$	$0.84 \pm 0.03aB$
SOD(units/mg Hb)	G1	$1.17 \pm 0.06B$	$1.15 \pm 0.06B$	$1.16 \pm 0.07B$
	G2	$0.69 \pm 0.02aA$	$0.77 \pm 0.02abA$	$0.84 \pm 0.02bA$
CATALASE(units/mg Hb)	G1	$1.31 \pm 0.17B$	$1.31 \pm 0.16B$	$1.28 \pm 0.16B$
	G2	$0.54 \pm 0.03aA$	$0.63 \pm 0.01A$	$0.99 \pm 0.01bA$

(Group 1: healthy control group with no treatment, Group 2: Animals treated with azithromycin. Values (mean  $\pm$  SE) having no common superscripts (small letters in row and capital letters in a column) differ significantly at  $p < 0.05$ ).

Coagulase positive *staphylococcus aureus* species was isolated in the skin swab culture of group 2 dogs. The pattern of sensitivity of different antibiotics are given in Table 2.

**Table 2:** Antibiotic sensitivity pattern of *S. aureus* cultural isolate obtained from pyoderma affected dogs

S. No	Antibiotic Disc	Sensitive	Resistant
1	Azithromycin (15 $\mu$ g/disc)	72.34%	27.66%
2	Amoxycyclavulanate (20+10 $\mu$ g/disc)	68.28%	31.72%
3	Mupirocin	68.10	31.90%
4	Cephalexin (30 $\mu$ g/disc)	66.41%	33.59%
5	Vancomycin (15 $\mu$ g/disc)	34.34%	65.66%
6	Ceftriaxone with sulbactam (30 $\mu$ g/disc)	41.38%	58.62%
7	Amikacin (30 $\mu$ g/disc)	22.41%	77.59%

## 4. Discussion

Among the clinical signs recorded in the present study erythema, alopecia, pruritus, papules, crusts and pustules were more common. These were similar to the findings of Craig [10]; Hillier *et al.* [11]; Kelany and Husein [12] and Beigh *et al.* [13]. Bacterial isolation revealed *staphylococci aureus* as the major pathogen which is in accordance with Senapati *et al.* [14] and Hariharan *et al.* [15] also identified *S. aureus* as the major pathogen in dogs with pyoderma. Regarding oxidative stress marker activity, the pyoderma-infected group showed a significant high LPO value with a significant low SOD and catalase values. This indicates existence of oxidative stress at a significantly higher level in pyoderma affected dogs. There was significant decrease in the LPO value on day 30, which is in agreement with Jewell *et al.* [16], Behera *et al.* [17], Packer *et al.* [18], Saskia *et al.* [19], Rock *et al.* [20]. On day30 of the experiment the significant reduction in the erythrocytic mean LPO values may be due to killing the causative organisms and reducing the altered function in different vital organs through antibiotic therapy. Significant increase in SOD and Catalase values were observed on both day 15 and day 30 which may be due to stimulation of body reticulo-endothelial system

enhancing antioxidative enzyme level and reducing oxidant level in absence of bacteria related stress.

The results of the present study suggested that canine pyoderma is a stress related skin disease that affects the antioxidant mechanism of the body, causes significant changes in the erythrocytic oxidative indices which can be normalised after proper antibiotic therapy.

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