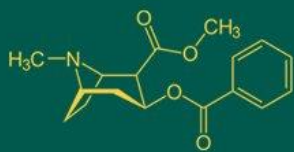


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Antioxidant effects of vitamin E as an adjunct to Marbofloxacin in management of pyoderma in dogs

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Abstract

Skin diseases are a common problem in dogs that may lead to increased risk of oxidative stress. Oxidative stress arises when cells metabolize oxygen and generate free radicals, known as reactive oxygen species (ROS). Under normal conditions, ROS are neutralized by antioxidant defenses; however, excessive ROS production overwhelms these defenses, leading to cellular damage. This damage is reflected by the release of lipid and enzyme biomarkers and contributes to tissue inflammation, infections, obesity. The following study was carried out in 18 dogs in which six dogs were used as control, twelve dogs were affected with pyoderma. This study was done to estimate oxidative stress biomarkers (Lipid peroxidase, Super oxide dismutase, Catalase). Diagnosis was confirmed by characteristic lesions and microscopic findings of degenerated neutrophils with cocci and rod bacteria in the impression smears. Common clinical lesions comprised of pruritus, erythematous papules, furuncles with edema, hot spots with moist erosive surfaces, ruptured pustular moist lesions, generalized alopecia, exfoliative superficial pyoderma, pyo-traumatic lesions, pustules, interdigital furunculosis, excoriations, erosions, and ulcers. Analysis of these erythrocytic oxidative stress marker revealed significantly high ($P \leq 0.05$) Lipid peroxidase (LPO) value and significant low ($P \leq 0.05$) Super oxide dismutase (SOD) and Catalase values at day 0 of experiment. After antibiotic treatment enhanced with vitamin E Group (T_2) therapy the altered oxidative stress LPO was subsequently found to be normalised at day 14 of the experiment. while antibiotic treated group (T_1) found decreased significantly by 7 indicating reduced oxidative stress but remained elevated and failed to normalize even at day 21. SOD activity showed recovery from day 14, approaching control values in the Combination group (Antibiotic + Vit E) (T_2), while in the antibiotic-only group (T_1) it increased slightly by day 21 but remained lower than control. Catalase activity surpassed control by day 14 in T_2 group, whereas in T_1 group it neared normal by day 21.

Keywords: Canine pyoderma, oxidative stress, LPO, SOD and catalase

Introduction

Canine pyoderma, is one of the most frequently diagnosed dermatopathy in dogs, is characterized by a bacterial skin infection. It is widely recognized as a secondary condition caused by underlying factors that disrupt the skin barrier or immune defenses (Bajwa, 2016) [1]. Dogs are particularly predisposed 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 (Sasaki *et al.*, 2007) [18]. The condition is most commonly caused by gram positive bacteria (Staphylococcus) (Devriese *et al.* 2005) [6] which may be superficial, affecting the epidermis or surface or deeper structures of the skin characterized by pustules, papules, pruritus, and alopecia (Bourguignon *et al.* 2013) [4].

Oxidative stress can be defined as the phenomenon of cell and tissue damage as a result of the imbalance between oxidant/antioxidant substances in body (Puppel *et al.*, 2015) [15]. 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 (Meher *et al.*, 2018) [11]. ROS-induced cellular damage was delayed or prevented by antioxidants (Paulsen and Carroll 2013) [14]. ROS-induced cellular damage was delayed or prevented by antioxidants (Paulsen and Carroll 2013) [14]. Antioxidants include high-molecular-weight antioxidant enzymes represented by glutathione peroxidase, superoxide dismutase, and catalase enzymes (Trouba *et al.* 2002) [21].

Vitamin E, the most powerful antioxidant, is concentrated in cell membranes. It is considered to be the first line of defence against lipid peroxidation, protecting cell membranes at an early stage of free radical attack through its free radical quenching activity (Kapun *et al.* 2014) [9]. It is an important constituent of sebum and is constantly secreted to the surface of the skin. Due to the high concentration of fats in the keratinous layer, this lipophilic antioxidant is expected to play an important part in removing ROS in the skin during oxidative stress.

This work aimed to assess oxidative stress levels in pyoderma infected dogs and therapeutic response to Vitamin E supplementation as an adjunct treatment along with systemic antibiotic.

Materials and Methods

The proposed work was carried out on the clinical cases of dogs suffering with pyoderma, presented to the Veterinary Clinical Complex, College of Veterinary Science and Animal Husbandry, Rewa, and the Divisional Veterinary Hospital, Bichhiya, Rewa along with private clinics for various dermatological conditions.

Experimental design

A total of 18 dogs diagnosed with pyoderma and exhibiting clinical signs of pyoderma like erythema, alopecia, pruritus, papules, crusts, pustules, epidermal collarettes, Pus in skin etc were randomly allocated into Two therapeutic groups (T₁, T₂) to evaluate the efficacy of systemic, and systemic plus vitamin E, and In addition, six apparently healthy dogs were included as the control group (T₀) and did not receive any treatment. In therapeutic Group (T₁) Marbofloxacin was prescribed @ 2mg/kg b.wt. OD, PO for 21 days. Group (T₂) Marbofloxacin was prescribed @ 2mg/kg b.wt. OD, PO for 21 days plus vitamin E @ 5mg/kg b.wt. OD, PO for 21 days. In the study, all animals included in the therapeutic trial were provided supportive treatment i.e., Hydroxyzine hydrochloride @ 2 mg/kg, PO OD for one week. and Syrup vitabest derm administered orally at 5-10 ml once daily for one week.

Diagnosing canine pyoderma

Clinical diagnosis of canine pyoderma was based on the presence of erythema, alopecia, pruritus, papules, crusts, pustules, epidermal collarettes, Pus within the skin was assessed, and impression smear examination were taken and examined microscopically by Mendelsohn *et al.* (2006) [12]

Parameters studied

The blood of examined animals was drawn using heparinized anticoagulant tubes to prepare plasma and erythrocytes lysates. Briefly, the red cells were collected by centrifugation (3000 rpm for 15 min) and plasma was drawn off, then cells were washed with three times with cold saline, with centrifugation after each wash. The resultant red cell pellets was used for estimation of the levels of LPO (lipid peroxidase) enzymes using method suggested by Rehman (1984) [16]. The resultant red cell pellets were lysed by adding (1:10 dilution) of cold distilled water to the estimated pellet volume and red cell stroma was removed by centrifuging. The resulting clarified supernatant was used for estimation of the levels of superoxide dismutase and catalase enzymes using Madesh and Balasubramanian (1998) [10] and Brannan *et al.* (1981) [5], respectively.

Statistical analysis

The data generated during the study were analysed using standard statistical procedures as described by Snedecor and Cochran (1994) [20]. Results were expressed as mean $\pm$ standard error, and differences between groups and treatment days were evaluated for statistical significance.

Results and Discussion

Skin lesions in pyoderma

Multiple pustular lesions on the medial aspect of the thigh (Plate 01:A), Extensive alopecia on the head and thorax (Plate 01:B), Superficial pyoderma with red inflamed patches (Plate 01:C), Interdigital lesions in dog with pyoderma (Plate 01:D), Multiple raised skin lesions on the shoulder, chest and forelimb (Plate 01:E), Multifactorial alopecia with crusted papules (Plate 01:F), Moth eaten alopecia affecting the dorsal region (Plate 01:G), There are deep, irregular ulcers on the tail and perineal region (deep pyoderma) (Plate 01:H), are common skin lesions in cases of pyoderma in dogs. In case of papules and pustules, the bacterial invasion (eg. *Staphylococcus pseudintermedius*) triggers neutrophilic inflammations leading to pustule formation which rupture and form crusts. Bacterial toxins and cytokines (IL-1 and TNF alpha) causes vasodilatation leading to redness (erythema) and warmth (Hillier and Griffin, 2001) [8]. Folliculitis damages hair follicles causing alopecia (Santaro *et al.*, 2015) [17] and is exacerbated by pruritus and self-trauma. Pustules rupture oozing serum and WBCs along with bacteria forming dried crusts and further perpetuating the infection (Beco *et al.*, 2013). Healing pustules leave circular scaling as epidermis regenerates causing formation of epidermal collarettes (Millier *et al.*, 2013). Deep bacterial invasion may lead to tissue necrosis and further ulcer formation. In chronic inflammations, it may stimulate melanocytes causing post inflammatory hyper-pigmentation (Santaro *et al.*, 2015) [17].

Impression smear examination

Impression smears from the lesions of superficial pyoderma affected dogs were stained with Giemsa stain and EMB (Eosin methylene blue) and observed under oil immersion (100 x) which revealed the presence of cocci and rod bacteria in most of the cases along with degenerated neutrophils (Plate 02 A 100x). impression smear revealing large no. of rods (bacilli) (Plate no. 02 B 100x), were recorded during microscopic examination of stained impression smears Similar observations as in the present study have been previously recorded by Bloom and Rosser (2001) [3], Hiller *et al.* (2006) [7] and Veeranki (2015) [22].

Oxidative stress parameters

Lipid peroxidase ($\mu\text{mol/L}$)

The Mean $\pm$ S.E. values of LPO before and after treatment in the control and different treatment groups across different days are presented in Table 1 A significantly ($P \leq 0.05$) higher LPO value was recorded in group T₁ ($4.29 \pm 0.09^{\text{bc}}$) and group T₂ ($4.56 \pm 0.06^{\text{cd}}$) compared with the healthy control group (T₀) on day 0. These findings are consistent with Mehar *et al.* (2018) [11] and Sivel *et al.* (2022) [19]. Who found a significant increase in mean values of LPO in dogs affected with pyoderma when compared with healthy control animals. Bacterial infection may lead to increased liberation of reactive oxygen species (ROS) causing lipid peroxidation reflected in form of increased lipid peroxidase

activity (Miller *et al.*, 2013) [13]. Similarly, cytokines and neutrophils might also release ROS causing oxidative damage. Skin inflammation also leads to cellular membrane

damage and increased lipid peroxidation. (Santoro *et al.*, 2015) [17].

Table 1: Levels of LPO ($\mu\text{mol/L}$) in the different treatments and control groups

Parameters	Groups (n = 06)	Mean $\pm$ SE			
		0 DAY	7 th DAY	14 th DAY	21 st DAY
LPO ($\mu\text{mol/L}$)	T ₀	2.15 $\pm$ 0.08 ^{aA}	2.22 $\pm$ 0.06 ^{aA}	2.16 $\pm$ 0.06 ^{aA}	2.17 $\pm$ 0.06 ^{aA}
	T ₁	4.29 $\pm$ 0.09 ^{bC}	3.71 $\pm$ 0.08 ^{bC}	3.38 $\pm$ 0.08 ^{bA}	3.35 $\pm$ 0.08 ^{cA}
	T ₂	4.56 $\pm$ 0.06 ^{cD}	3.44 $\pm$ 0.05 ^{bC}	2.77 $\pm$ 0.06 ^{abB}	2.62 $\pm$ 0.04 ^{bA}

Means with different superscript in capital letter in a row and small letter in a column differ significantly ($P \leq 0.05$)

Treatment groups (T₁-T₂) exhibited progressive declines in LPO levels from Day 7 onwards ($P \leq 0.05$), attaining values approach to the control group by Day 21, which were significant ($P \leq 0.05$) when compared to Day 0 values. Group T₂ showed the most rapid reduction by Day 14, stabilizing thereafter and becoming nearly comparable to controls. Vitamin supplemented antibiotic group (T₂) performed best in a synergistic fashion as compared to other groups in reducing the oxidative stress and resultant skin healing in pyoderma by reducing bacterial load along with reduction in oxidative stress. Similarly, Verma (2023) [23] also recorded improvements after different treatment with

like clindamycin along with Vit E as compared to clindamycin alone.

Super oxide dismutase (U/g of Hb)

The Mean $\pm$ S.E. values of SOD before and after treatment in the control and different treatment groups across different days are presented in Table 2 A significantly lower SOD values were observed in group T₁ (1009.5 $\pm$ 67.31^{dA}) and group T₂ (959.17 $\pm$ 54.96^{aA}) compared with the healthy control group (T₀) on day 0. These findings are consistent with Mehar *et al.* (2018) [11] and Sivel *et al.* (2022) [19].

Table 2: Levels of SOD (U/g of Hb) in the different treatments and control groups

Parameters	Groups (n = 06)	Mean $\pm$ SE			
		0 DAY	7 th DAY	14 th DAY	21 st DAY
SOD (U/g of Hb)	T ₀	1333.18 $\pm$ 101.14 ^{eA}	1338.72 $\pm$ 82.67 ^{eB}	1335.8 $\pm$ 76.99 ^{eB}	1352.47 $\pm$ 34.58 ^{eB}
	T ₁	1009.5 $\pm$ 67.31 ^{dA}	1025.5 $\pm$ 73.58 ^{dA}	1038.67 $\pm$ 60.2 ^{cC}	1041.67 $\pm$ 89.15 ^{cD}
	T ₂	959.17 $\pm$ 54.96 ^{aA}	1047.83 $\pm$ 66.86 ^{dB}	1173.5 $\pm$ 62.57 ^{dC}	1176.5 $\pm$ 69.38 ^{dD}

Means with different superscript in capital letter in a row and small letter in a column differ significantly ($P \leq 0.05$)

Treatment groups (T₁-T₂) exhibited progressive increases in SOD levels from Day 7 onwards ($P \leq 0.05$), attaining values approaching the control group by Day 21, which were significant when compared to Day 0 values. Group T₂ showed the most rapid and substantial elevation, becoming nearly comparable to controls by Day 14 and stabilizing. In contrast, T₁ showed moderate progressive improvements but remained somewhat below control levels. Similar Improvements were also recorded by Verma (2023) [23] after regimens like clindamycin with vitamin E.

Catalase (U/g of Hb)

The Mean $\pm$ S.E. values of Catalase before and after treatment in the control and different treatment groups across different days are presented in Table 3 catalase values were significantly lower in group T₁ (936.5 $\pm$ 25.34^{aA}) and group T₂ (931.5 $\pm$ 32.56^{aA}) compared with the healthy control group (T₀) on day 0 These findings were in correspondence with the reports of Mehar *et al.* (2018) [11], Sivel *et al.* (2022) [19]

Table 3: Levels of Catalase (U/g of Hb) in the different treatments and control groups

Parameters	Groups (n = 06)	Mean $\pm$ SE			
		0 DAY	7 th DAY	14 th DAY	21 st DAY
CATALASE (U/gHb)	T ₀	1029.2 $\pm$ 58.61 ^{bC}	1028.7 $\pm$ 98.51 ^{eB}	1028.52 $\pm$ 99.03 ^{dA}	1038.52 $\pm$ 102.89 ^{dA}
	T ₁	936.5 $\pm$ 25.34 ^{aA}	984.5 $\pm$ 56.89 ^{bC}	1004.5 $\pm$ 53.8 ^{cC}	1014.5 $\pm$ 22.74 ^{cD}
	T ₂	931.5 $\pm$ 32.56 ^{aA}	990.33 $\pm$ 34.23 ^{dB}	1117.5 $\pm$ 61.31 ^{eC}	1126.5 $\pm$ 80.82 ^{eC}

Means with different superscript in capital letter in a row and small letter in a column differ significantly ($P \leq 0.05$)

Treatment groups (T₁-T₂) exhibited progressive increases in SOD levels from Day 7 onwards ($P \leq 0.05$), attaining values approaching the control group by Day 21, which were significant when compared to Day 0 values. Group T₂ showed the most rapid and substantial elevation, surpassing other treatment and nearing control levels by Day 14. In

contrast, T₁ showed moderate progressive improvements but remained below control levels. Similarly, Verma (2023) [23] also recorded improvements in catalase values in pyoderma dogs when treated with clindamycin alone or along with Vit. E.



Plate 1: (A) Multiple pustular lesions on the medial aspect of the thigh (B) Extensive alopecia on the head and thorax (C) Superficial pyoderma with red inflamed patches (D) Interdigital lesions in dog with pyoderma (E) Multiple raised skin lesions on the shoulder, chest and forelimb (F) Multifactorial alopecia with crusted papules and erosions distributed over the dorsal trunk and limbs (G) Moth eaten alopecia affecting the dorsal region (H) There are deep, irregular ulcers on the tail and perineal region (deep pyoderma)

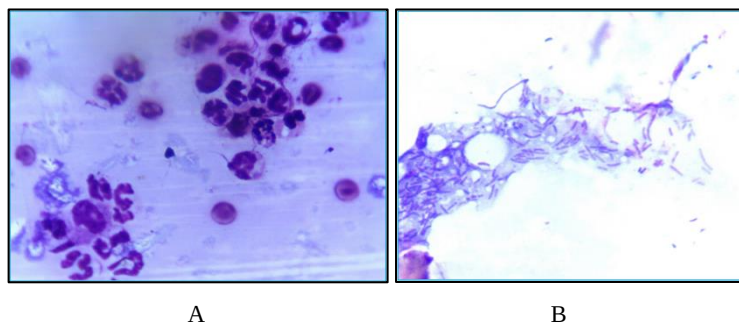


Plate 2: A. Impression smear showing degenerated neutrophils (100x) B. Impression smear showing Rods Bacilli (100x)

Summary and Conclusion

Dogs with pyoderma exhibited elevated lipid peroxidation and reduced antioxidant enzyme activities (SOD, Catalase) at baseline, confirming oxidative stress involvement. Both therapeutic regimens—Marbofloxacin alone (T₁) and Marbofloxacin with Vitamin E (T₂)—produced progressive improvements; however, T₂ achieved faster normalization of oxidative stress parameters and superior clinical recovery. Adjunctive Vitamin E enhanced antibiotic efficacy, reduced oxidative damage, and accelerated healing, supporting its role as a valuable antioxidant in canine pyoderma management.

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References

1. Bajwa J. Canine superficial pyoderma and therapeutic considerations. *Can Vet J.* 2016;57:204.
2. Beco L, Guaguere E, Mendez CL, Noli C, Nuttall T, Vroom M. Suggested guidelines for using systemic antimicrobials in bacterial skin infections: part 1—diagnosis based on clinical presentation, cytology and culture. *Vet Rec.* 2013;172(3):72.
3. Bloom PB, Rosser EJ. Efficacy of once daily cefovecin sodium hydrochloride in the treatment of superficial bacterial pyoderma in dogs. *Journal of the American Animal Hospital Association.* 2001;37:537-542.
4. Bourguignon E, Guimarães LD, Ferreira TS, Favarato ES. Dermatology in dogs and cats. In: Carreira RP, editor. *Insights from Veterinary Medicine.* 2013;1:3-4.
5. Brannan TS, Maker HS, Raes TP. Regional distribution of catalase in the adult rat brain. *Journal of Neurochemistry.* 1981;36(1):307-309.
6. Devriese LA, Vancanneyt M, Baele M, *et al.* *Staphylococcus pseudintermedius* sp. nov., a coagulase-positive species from animals. *Int J Syst Evol Microbiol.* 2005;55(4):1569-1573.
7. Hillier A, Alcorn JR, Cole LK, Kowalski JJ. Pyoderma caused by *Pseudomonas aeruginosa* infection in dogs: 20 cases. *European Society of Veterinary Dermatology.* 2006;17(6):432-439.
8. Hillier A, Griffin CE. The ACVD task force on canine atopic dermatitis (I): incidence and prevalence. *Vet Immunol Immunopathol.* 2001;81(3-4):147-151.
9. Kapun AP, Salobir J, Levart A, Kalcher GT, Svete AN, Kotnik T. Vitamin E supplementation in canine atopic dermatitis: improvement of clinical signs and oxidative stress. *Vet Rec.* 2014;175(22):560.
10. Madesh M, Balasubramanian KA. Microtiter plate assay for superoxide dismutase using MTT reduction by superoxide. *Indian Journal of Biochemistry and Biophysics.* 1998;35(3):184-188.
11. Meher S, Gupta AR, Dalai HK, Nayak SM, Samal P, Sethy K, *et al.* Evaluation of altered erythrocytic oxidative stress indices in association with canine pyoderma. *Pharma Innov J.* 2018;7(7):532-534.
12. Mendelsohn C, Rosenkrantz W, Griffin CE. Practical cytology for inflammatory skin diseases. *Clinical*

- Techniques in Small Animal Practice. 2006;21(3):117-121.
13. Miller WH, Griffin CE, Campbell KL. Muller and Kirk's small animal dermatology. 7th ed. St Louis: Elsevier Mosby; 2013. p. 71-78.
 14. Paulsen CE, Carroll KS. Cysteine-mediated redox signaling: chemistry, biology, and tools for discovery. Chem Rev. 2013;113(7):4633-4679.
 15. Puppel K, Kapusta A, Kuczyńska B. The etiology of oxidative stress in the various species of animals: a review. J Sci Food Agric. 2015;95(11):2179-2184.
 16. Rehman SU. Lead induced regional lipid peroxidation in brain. Toxicol Lett. 1984;21:333-337.
 17. Santoro D, Marsella R, Pucheu-Haston CM, Eisenschenk MN, Nuttall T, Bizikova P. Pathogenesis of canine atopic dermatitis: skin barrier and host-micro-organism interaction. Vet Dermatol. 2015;26(2):84-e25.
 18. Sasaki T, Kikuchi K, Tanaka Y, Takahashi N, Kamata S, Hiramatsu K. Reclassification of phenotypically identified *Staphylococcus intermedius* strains. J Clin Microbiol. 2007;45(9):2770-2778.
 19. Sivel GN, Yagcı BB. Evaluation of oxidative stress on dogs with demodicosis. Turkish Journal of Veterinary Research. 2022;7(1):7-13.
 20. Snedecor GW, Cochran WG. Statistical methods. 8th ed. Ames (IA): Iowa State University Press; 1994. p. 217-235.
 21. Trouba KJ, Hamadeh HK, Amin RP, Germolec DR. Oxidative stress and its role in skin disease. Antioxid Redox Signal. 2002;4:665-673.
 22. Veeranki MB. Clinico-diagnostic and therapeutic studies on superficial pyoderma in dogs [M.V.Sc. thesis]. Tirupati: Sri Venkateswara Veterinary University; 2015.
 23. Verma S. Prevalence and therapeutic evaluation of clindamycin vis-à-vis antioxidant supplementation against pyoderma in dogs [M.V.Sc. thesis]. Jabalpur: Nanaji Deshmukh Veterinary Science University; 2023.
 24. Wolf R, Wolf D, Ruocco V. Vitamin E: the radical protector. J Eur Acad Dermatol Venereol. 1998;10:103-117.