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Short Communication

CHANGES IN LIPID PEROXIDATION AND ANTI-OXIDANT ENZYME STATUS WITH AGEING IN DOGS

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SUMMARY

The present study was conducted to evaluate the alterations in oxidative stress parameters viz., malondialdehyde (MDA) and superoxide dismutase (SOD) in geriatric dogs. The study was performed in 12 apparently healthy adult (2-4 years age) and 12 apparently healthy geriatric (≥8 years age) dogs of different breeds. The oxidative stress profile revealed significant (P<0.05) difference in values. The value of MDA (3.53±0.43 nmol MDA/mg of Hb) was significantly higher whereas, SOD (35.99±1.96 U/mg of Hb) was significantly lower in apparently healthy geriatric dogs as compared to the apparently healthy adult dogs (1.11±0.26 nmol MDA/mg of Hb and 51.68±3.49 U/mg of Hb, respectively).

Key words: Ageing, Antioxidant, Dogs, Geriatric, Lipid Peroxidation, Oxidative Stress.

Ageing is the time related deterioration of the physiological functions, leading to the cell's inability to withstand external and internal stress. The causative factors for the time dependant deleterious process of ageing are yet not well defined and no single adequate molecular explanation for ageing is currently available (Singh *et al.,* 2009). Increased oxidative stress and its definitive role in the age-related diseases is well documented fact in human beings (Pandey and Rizvi, 2010). One of the most popular explanations of how ageing occurs at the molecular level is the oxidative stress hypothesis. Free radicals are highly reactive molecules that are produced during normal metabolism in the body or after exposure to environmental pro-oxidants. Excess free radicals cause a dangerous chain reaction that can destroy nucleic acid, proteins, lipids and other cellular compounds (Haliwell and Gutteridge, 1999).

Body counteracts against ill effects of free radicals via antioxidant defense system that comprises of antioxidant enzymes like superoxide dismutase (SOD), catalase, glutathione peroxidase and glutathione reductase. Oxidative stress supervenes when generated free radicals exceed the capacity of antioxidant defense of the body. Estimation of lipid peroxidation and antioxidant enzyme activity in blood are indirect but reliable methods for assessment of free radical activity and oxidative stress (Fang *et al*., 2002). The present study was conducted to determine the variation in oxidant– antioxidant status with ageing in dogs.

The study was conducted at Small Animal OPD unit of Teaching Veterinary Clinical Complex, Faculty of Veterinary Science and Animal Husbandry, Sher-e-Kashmir University of Agricultural Sciences and Technology, R.S. Pura, Jammu. Two groups, I consisting of 12 apparently healthy adult dogs (2-4 years age) and II with 12 apparently healthy geriatric dogs (≥8 years age) of different breeds were the subject of the study. Heparinised whole blood samples were used for the estimation of lipid peroxidation and superoxide dismutase. One per cent RBC lysate and 33 per cent RBC lysate were used for the estimation of superoxide-dismutase, and lipid peroxidation, respectively.

The lipid peroxidation activity in erythrocytes was determined according to method described by Shafiq-ur-Rahman (1984) and was expressed as nmol MDA formed/ml packed cells. The molar extinction coefficient (EC) of MDA-TBA complex at 535 nm is 1.56 X 10^8/M/cm and calculation was done using formula:

= (OD/EC) x (Total volume of reaction mixture/Amount of sample taken) x 10^9 x 2 (incubation time)

The activity of superoxide dismutase (SOD) in erythrocyte lysate was determined by the method given by Marklund and Marklund (1974) and expressed as SOD Units/mg of Hb. Calculation was done using formula:

SOD activity (Units/g of Hb) = $(\Delta E_0 - \Delta E)/ \Delta E_0$ x ½ x 1/ g of haemoglobin in 0.02 ml

ΔE_0 = change of absorbance of pyrogallol

ΔE = change of absorbance of sample

Statistical analysis was done by independent t-test to determine the differences between the groups. The results are summarized in Table 1.

The mean value of MDA in apparently healthy geriatric dogs was found significantly (P<0.05) higher (3.53±0.43 nmol MDA/mg of Hb) than that of adult dogs (1.11±0.26 nmol MDA/mg of Hb) (Table 1). The results of present study are in accordance with those of Todorova *et al*. (2005); Thaiklang *et al.* (2005); Hwang *et al*. (2008); Suresh *et al*. (2010), Bhar *et al*. (2011) and Jain *et al.* (2013); who also reported increase in MDA levels with ageing.

MDA is an excellent marker of lipid peroxidation and oxidative stress (Kasapoglu and Ozben, 2001). Increased oxidative damage with ageing as reflected by elevated MDA levels in geriatric dogs could be a result of cumulative effect of repeated exposure to ionizing

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Table 1
Oxidative stress parameters of apparently healthy adult and geriatric dogs

Name of parameters	Adult dogs (n=12)	Geriatric dogs (n=12)
MDA (nmol MDA/mg of Hb)	1.11±0.26[a]	3.53±0.43[b]
SOD (U/mg of Hb)	51.68±3.49[a]	35.99±1.96[b]

Different superscripts indicate significant difference within a row at P<0.05

radiations, mitochondrial dysfunction and reduction in antioxidant defense mechanisms due to ageing.

ROS/free radicals are produced in the body as a byproduct of aerobic metabolism damage cellular macromolecules that include lipids, proteins, DNA and mitochondrial components. Mitochondria are the major source of ROS/ free radicals in the body. The accumulation of these oxidative damages may be a major contributory factor in cellular ageing. Lipid peroxidation is an important biological consequence of oxidative cellular damage. It is a chain reaction and leads to generation of more and more free radicals which in turn cause further peroxidation of other PUFA and results in greater cell damage and dysfunction.

Cell membranes which are made up of large amounts of PUFA are highly susceptible to oxidative attack. Saxena *et al*. (2007) reported that lipid peroxidation contributes to local membrane destabilization that alters the proper trafficking of intracellular vesicles, phagocytosis, degranulation, antigen presentation, receptor mediated ligand uptake etc. leading to age related deterioration in many cellular functions.

Depletion of antioxidants is an indirect marker of oxidative stress during ageing. The activity of SOD, a superoxide radical scavenging enzyme was found to be significantly decreased in apparently healthy geriatric dogs (35.99±1.96 U/mg of Hb) than that of adult dogs (51.68±3.49 U/mg of Hb). Similar findingswere also reported by Todorova *et al*. (2005); Singh *et al.* (2009) and Jain *et al.* (2013).

Superoxide dismutase is the first line of defense against the superoxide radical, which is formed from molecular oxygen by single electron transfer. This enzyme converts the highly reactive superoxide radicals (O^{2-}) into less toxic hydrogen peroxide and decreases cell damage. Hydrogen peroxide is further detoxified either by catalase or by glutathione peroxidase to water and oxygen. The decreased levels of SOD activity with ageing reflect the cellular damage due to accumulation of O^{2-}. The diminished activity of SOD may be due to its progressive inactivation by H_2O_2, or increased glycosylation of SOD with ageing (Saxena and Lal, 2006).

The findings of this study indicate that with ageing, there is an increase in lipid peroxidation and decrease in endogenous antioxidant function in dogs.

REFERENCES

Bhar, A., Ghosh, C.K., Bandyopadhayay, S., Das, S., Sarkar, S. and Debnanth, P.K. (2011). Haematobiochemical and antioxidative changes in geriatric canines. *Indian Vet. J.* **88(3):** 70-71.

Fang, Y.Z., Yang, S. and Wu, G. (2002). Free radicals, antioxidants and nutrition. *Nutr.* **18 (10)**: 872-879.

Haliwell J. and Gutteridge, M.C. (1999). Free Radicals in Biology and Medicine. 3rd edn. Oxford University Press, Oxford.

Hwang, I.N., Yoon, Y.S., Yoo, K.Y., Choi, J.H., Kim, D.W., Yi, S.S., Seong, I., Lee, J.K.S. and Won, M.H. (2008). Differences in lipid peroxidation and Cu, Zn superoxide dismutase in the hippocampal CA1 region between adult and aged dogs. *J. Vet. Med. Sci.* **70(3):** 273–277.

Jain, S., Patel, P.R. and Ravel, S.K. (2013). Changes in haematological parameters in adult and geriatric dogs. *Vet. Pract.* **14(2):** 230-231.

Kasapoglu, M. and Ozben, T. (2001). Alterations of antioxidant enzymes and oxidative stress markers in ageing. *Exp. Gerontol.* **36(2)**:209-20.

Marklund, S. and Marklund, M. (1974). Involvement of superoxide anion radical in autoxidation of pyrogallol and a convenient assay of superoxide dismutase. *Eur. J. Biochem.* **47 (3)**: 469–474.

Pandey, K.B. and Rizvi, S.I. (2010). Markers of oxidative stress in erythrocytes and plasma during ageing in humans. *Oxid. Med. Cell. Longev.* **3(1)**: 2-12.

Saxena, R. and Lal, A.M. (2006). Effect of Ageing on antioxidant enzyme status and lipid peroxidation. *J. Indian Acad. Geriat.* **2(2)**: 53-56.

Saxena, R. and Lal, A.M. (2007). Culprit effect of altered total antioxidant status and lipid peroxidation mediated electrolyte imbalance on ageing. *J. Indian Acad. Geriat.* **3**: 137-144.

Shafiq-ur-Rahman. (1984). Lead-induced regional lipid peroxidation in brain. *Toxicol. Lett.* **21(3)**: 333-337.

Singh, K., Kaur, S., Kumari, K., Singh, G. and Kaur, A. (2009). Alteration in lipid peroxidation and certain antioxidant enzymes in different age group under physiological condition. *J. Hum. Ecol.* **27(2):** 143-147.

Suresh, D.R., Kumaran S., Annam, V. and Hamsaveena (2010). Age related changes in malondialdehyde: total antioxidant capacity ratio-a noval marker of oxidative stress. *Int. J. Pharm Bio Sci.* **1(2)**: 1-6.

Thaiklang, J., Suriyasathaporn, W., Tangphokhanon, W., Vinitkekumnuegy, U. and Chewo, T. (2005). Relationship between oxidative stress, hematology, blood chemistry in adult and senile dogs. *Chiang Mai Vet. J.* **3**: 15-20.

Todorova, I., Simeonova, G., Kyudrukova, D.D. and Gadjeva, V. (2005). Reference values of oxidative stress parameters (MDA, SOD, CAT) in dogs and cats. *Comp. Clin. Path.* **13(4):** 190-194.