

# Effect ageing stages on serum antioxidant enzymes the domestic rabbits *Oryctolagus cuniculus* (Linnaeus, 1758)

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**Abstract:** This study examined the effects the serum antioxidant enzymes on age stages of *Orycolagus cuniculus* (Linnaeus, 1758), using 42 rabbits and divided at different ages stages as following: - Group A: (Premature < 6 months), Group B: (Mature > 6 months < 4 years) and Group C: (Old > 4 years) of age14 rabbits from each age group 7 males and 7 females, the result reveled that, during mature stage the assayed serum catalase, Glutathione Peroxidase and Superoxide Dismutase levels were markedly increased, and were decreased at old stage and Premature stage at both sexes.

Key Word: Age stages; antioxidant enzymes; the domestic rabbits; Oryctolagus cuniculus

## **INTRODUCTION**

Aging is a complex phenomenon that depends on the interaction of numerous genes, cellular pathways and environmental risk factors. It leads to a progressive functional decline, or a gradual deterioration of physiological function, including impairment of vision loss and degeneration of some organs (Harman, 1981).

Aging defined by many scientists and researchers as a complex phenomenon that depends on the interaction of numerous genes, cellular pathways and environmental risk factors, it leads to a progressive functional decline, or a gradual deterioration of physiological function, aging is one of the major aspects of human life and has both positive and negative effects on functional abilities of the human being as well as animals (Mohammed, 2014).

Aging is the natural phenomenon, which is the process of growing old and is usually defined as the gradual biological impairment of normal function which has direct impact on the functional ability of organs and on the biological systems. Phenomenon of aging leads to changes in the brain size, vasculature, and cognition, therefore as age increases the brain shrinks and changes occur from the level of molecules to morphology, so some authors were defined aging as a process that presents various alterations in behavioral, physiological, and neurochemical processes (Hedden *et al.*, 2004; Stevens and Lowe, 2005 and Peters, 2006).

Elmansi, et al., (2011) studied the amino acids and some hormones contents in blood serum of Uromastyxaegyptius and Falco tinnunculus and he suggested that, the amino acids and hormones contents gradually decrease with the progress of aging, however in Rattusnovergicus, the amino acid contents and some hormones were markedly increased in the mid-age and then decreased with the advance age.

## **MATERIAL AND METHODS**

This study was conducted using 42 rabbits taken from Veterinary Medicine farm, Wrdama farm and Al-Bayda Aljdida farm, and divided at different ages stages as following: - Group A: Premature (before 6 months), Group B: Mature (after 6 months to 4 years) and Group C: Old (after 4 years of age). 14 rabbits from each age group 7 males and 7 females. Feed and fresh water were given ad libitum., pull up blood and the blood was collected and centrifuge at 3000 g for 5 minutes and serum collected and kept in refrigerator at -20°C. At the same time, and stored at - 20<sup>C</sup>.

## Experimental animal:

Domestic rabbits are classified as follows; Phylum: chordate, Class: Mammalia, Order Lagomorpha, Family: Leporidea, Genus: oryctolagus, Species: *Oryctolaguscuniculus* (Linnaeus, 1758).

O. cuniculus, also called a European an old world or a domestic rabbit, is the only species in its genus, the last Ice age confined the species to the Iberian Peninsula and small area of France and northwest Africa, but due to human action and adaptability of this species, European rabbit today exist in the wild on every continent except Asia and Antarctica, Domesticated O. cuniculus may be found worldwide. occurs Sweden, Norway, Poland, Germany, Czech Republic, Belgium, Ireland, Greece, Herzegovina, Bulgaria, Austria, and some Western Asia, through the Mediterranean basin to Morocco, Algeria, Tunisia and Libya. (Parker, 1990, Wilson and Reeder, 1993).

## Determine Antioxidants enzyme activity:

Superoxide dismutase (SOD) activity: Superoxide dismutase determined by Using enzymatic preparations Ready (Superoxide dismutase Assay Kit, 706002-96 well. USA) and use the spectrophotometer device at a wavelength of 450 nanometers (Kakkar*et al.*, 1978; Sun *et al.*, 1988).

**Glutathione Peroxidase**: Glutathione Peroxidase determined by Using enzymatic preparations Ready (Glutathione peroxidase Assay kit, 703102-96 Well, USA) and use the spectrophotometer device at a wavelength of 340 nm. (Rotruck *et al.*, 1973).

**Catalase**: Catalase determined by Using The enzyme activity in serum estimate colorimetric method Using enzymatic preparations Ready, and using Plat reader,

\*Corresponding Author: Prof. Yousef K. A. Abdalhafid, Faculty Science, Omar Al-Mukhtar University, Al Bayda, Libya. DIGNOSTIC PASTEUR LP 400, FRANCE at a wavelength of 540nm and at a temperature of 37°c. (Sihia, 1972).

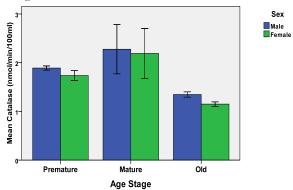
#### Statistical analysis:

Data were presented as means  $\pm$  standard error (SE). The statistical analysis was performed with multivariant analysis of variance (MANOVA) using SPSS (version 13) software package for Windows comparing the multi variations between the groups. F-test was calculated and considered statistically significant at p < 0.05.

#### Catalase:

# RESULTS

Figure (1) illustrates the catalase serum levels of both sexes of *O. cuniculus* at different age stages, during mature stage the assayed catalase of both sexes were markedly increased, on the other hand, the levels of catalase of both sexes were decreased at old stage more than the premature stage. There were no wide variations of the catalase between male and females at premature stage and old stages.



**Figure 1:** Effect aging stages on the Catalase (nmol/min/100ml) of both sexes of *O. cuniculus* at different age stages.

## Glutathione Peroxidase:

Figure (2) illustrates the Glutathione Peroxidase serum levels of both sexes of *O. cuniculus* at different age stages, during mature stage, the assayed Glutathione Peroxidase of both sexes were markedly increased. However, the levels of Glutathione Peroxidase of both sexes were decreased at old more than premature stage.

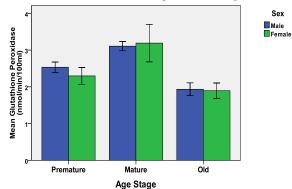


Figure 2: Effect aging stages on the Glutathione Peroxidase (nmol/min/100ml) of both sexes of *O. Cuniculus* at different age stages.

#### Superoxide Dismutase:

Figure (3) illustrates the Superoxide Dismutase serum levels of both sexes of *O. cuniculus* at different age stages. During mature stage and premature stage, the assayed Superoxide Dismutase of both sexes were markedly increased. However, the levels of Glutathione Peroxidase of both sexes were decreased at old stage, and almost stable between males and females at all age stages.

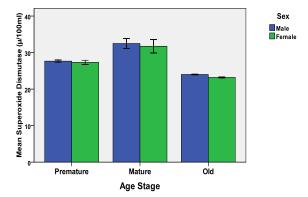


Figure 3: Effect aging stages on the Superoxide Dismutase  $(\mu/100ml)$  of both sexes of *O. cuniculus* at different age stages.

## DISCUSSION

Aging is the time related deterioration of the physiological functions, leading to the cell's inability to withstand external and internal stress. The aging process is slow in the early stages of life but rapidly increases in later stages due to the exponential nature of the process. The causative factors for the time dependant deleterious process of aging are yet not well defined and no single adequate molecular explanation for aging is currently available (Harman 1992, Timiras 1994).

The observed findings agree with, Raoet al. (1990) Cand and Verdetti, (1989). There are at least two reports, which demonstrated a decline in the Glutathione Peroxidase activity with age in wistar rats of both sexes, Rikanset al., (1992) reported that Glutathione Peroxidase activities decreased with age in male a female rats.

The variation patterns that happens in the levels of Superoxide Dismutase in this study agrees to data of literature (Sahoo and Chainy, 1997), where it possess high activity in the early ages and decreases with old stages.

Sohalet al. (1995) suggested that, Catalase in the brain of males, contrary to the literature, decreased at an intermediate age, while other references show regularity in their levels for gerbils. The observed findings of aging were closely similar to Elmansi, (2011) and Mohamed, (2012) Catalase, Superoxide Dismutase and Glutathione Peroxidase activities were gradually increased in the mid-age and gradually depleted in the old ones in eye of some vertebrates. Age-related correlation between lipid peroxidation and antioxidant enzyme activity decreased levels of carotenoids are associated with aging (Castorinaet al., 1992; Beatty et al., 2001). Finally, the author concluded that the biological change of body associated to some age stages, this study revealed that, the body be on full activity in early and midage stages, comparing with progressed age stages.

## **R**EFERENCES

- Beatty S, Murray IJ, Henson DB, Carden D, Koh H and Boulton ME. Macular pigment and risk for age-related macular degeneration in subjects from a Northern European population. Invest. Ophthalmol. Vis. Sci., 42, (2001) 439–446.
- Cand F and Verdetti J. Superoxide dismutase, glutathione peroxidase, catalase and lipid peroxidation in the major organs of aging rats. Free Radical Biology and Medicine 7, (1989) 59– 63.
- Castorina C, Campisi A, Di Giacomo C, Sorrenti V, Russo A and Vanella A. Lipid peroxidation and antioxidant enzymatic systems in rat retina as a function of age. Neurochem. Res., 17, (1992) 599–604.
- Elmansi AA. Comparative studies on the effect of aging on the eye of some vertebrates. Ph.D. Thesis, Mansoura University (2011).
- Harman D. Free radicals, aging and degenerative diseases. In: I Merit, B Chance (Eds.): Free Radicals and Aging. Basel: Birkhauser, (1992). 234.
- Harman D. The aging process. Proc. Natl. Acad. Sci. USA, 78: (1981) 7124–7128.
- Hedden T and Gabrieli JD. Nat Rev Neurosci., 5:87– 96. Peters, R. (2006). Postgard. Med. J., 82 (964): (2004) 84-88.
- Kakkar P, Das B and Vismanathan P. A modified spectrophometric assay of superoxide dismutase. Indian J. Biochem. Biophysi. 21, (1978) 130–132.
- 9. Mohamed M. Comparative studies on the effect of aging on the eye of Albino rats. Ph.D. Thesis, Mansoura University (2012).
- 10. Parker S. (1990) Grzimek s Encyclopedia of mammals. New York, NY: McGraw-Hill, Inc.

- 11. Peters R. Postgard. Med. J., 82 (964), (2006) 84-88.
- Rao G, Xia E, Richardson A. Effect of age on the expression of antioxidant enzymes in male Fischer F344 rats. Mechanisms of Ageing and Development 53 (1), (1990) 49– 60.
- Rikans LE, Snowden CD, Moore DR. Effect of aging on enzymatic antioxidant defenses in rat liver mitochondria. Gerontology 38 (3), (1992) 133–138.
- Rotruck JT, Pope AL and Ganther HE. Selenium biochemical role as a component of gluthione peroxidase purification and assay. Sci. (1973) 588–593.
- Sahoo A, Chainy GBN. Alterations in the activities of cerebral antioxidant enzymes of rat are related to aging. International Journal of Developmental Neurosciences 15, (1997) 939–948.
- Sinha AK. Colorimetric assay of catalase. Anal. Biochem. 19, (1972) 389–394.
- Sohal RS, Agarwal S, Sohal BH. Oxidative stress and aging in the Mongolian gerbil (Merionesunguiculatus). Mechanisms of Ageing and Development 81, (1995) 15–25.
- Stevens A and Lowe JS. Human Histology, 3rd ed., Elsevier Mosby. Philadelphia, Edinburgh, London, New York, Sydney. (2005). 101-106.
- Sun Y, Oberley L and Li Y. A simple method for clinical assay of superoxide dismutase. Clin. Chem. 34, (1988) 497–500.
- 20. Timiras PS. Physiological Basis of aging and Geriatrics. Boca Raton: CRC, Press, (1994) 23-25.
- Wilson D and Reeder D (1993). Mammal species of the World: A Taxonmic and Geographic refere Washington, D.C: The Smithsonian Institution.

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