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Evaluation of Lenticular Reduced Glutathione, Ascorbic Acid and Lipid Peroxidation Levels among Cataractous Men

Sreeja S., Jeyanthi G.P. and Gayathri Devi S.
Department of Biochemistry, Biotechnology and Bioinformatics, Avinashilingam Institute for Home Science and Higher Education for Women, Coimbatore-641 043, Tamil Nadu, INDIA

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Abstract

Reactive oxygen species which are produced through plethora of reactions during oxidative stress in the human lens may play an important role in the formation of cataract. Lipid peroxidation may cause exacerbation of lens tissue. Glutathione and ascorbic acid are among the vital lens antioxidants that are involved in scavenging free radicals. The rationale of the present research was to assess the levels of reduced glutathione, ascorbic acid and lipid peroxidation in the eye lens among cataractous subjects. The lens samples were obtained from the ophthalmic centres in and around Coimbatore. They were categorised into apparently normal cataract men (n=15), diabetic cataract men (n=15) and hypertensive cataract men (n=15). All the parameters were analysed spectrophotometrically. The data interpretation was performed by Kruskal Wallis test. The outcome of the study showed a significant difference ($p<0.05$) in the levels of ascorbic acid and lipid peroxidation among apparently cataract men and diabetic cataract men. However, these groups did not show significant difference albeit they showed variations in the levels of glutathione. The levels of glutathione, ascorbic acid and lipid peroxidation showed no significant difference among diabetic cataract men and hypertensive cataract men. The levels of ascorbic acid were found to have significant difference ($p<0.05$) among apparently normal cataract men and hypertensive cataract men whereas similar trend was not observed in the levels of glutathione and lipid peroxidation.

Keywords: Cataract, eye lens, glutathione, ascorbic acid and lipid peroxidation.

Introduction

Eyes are the window through which we are able to enjoy the vision of outdoor world. Transparency and accommodation of eye lens facilitates to perceive a clear image of the surrounding. Any alteration in these properties of the eye lens may result in its opacification and cloudiness. It is an imperative component of the eye which is affluent in antioxidants namely reduced glutathione and ascorbic acid. When there is deterioration in the levels of these non enzymatic antioxidants, it may result in the progression of oxidative stress, ultimately leading to cataract development. It is one of the most common ophthalmic disorders prevailing in world today especially in the developing countries[1]. In this ocular disease, the lens becomes opaque as its transparency is lost. There are various causes behind the development of cataract among which oxidative stress is one major cause. Cataracts cause encumbrance to routine works such as reading or driving[2]. Globally, one among the major treatment for cataract is its surgery[3,4] which is expensive and not devoid of risk[5]. Cataract development is influenced by age, diet, heredity, medicines, exposure to sunlight and clinical complications namely hypertension, diabetes, renal diseases or any direct trauma to the eye[6,7].

There are many factors to which our eyes are continuously exposed viz sunlight, artificial lighting and pollutants which may cause oxidative stress that might lead to cataract development by damaging the ocular tissues[8]. Oxidative stress may be one of the most important reasons for cataract development[9,10]. The incessant exposure of ocular lens to oxidative tension through Reactive Oxygen Species (ROS) may alter the biochemistry and organisation of the eye lens fibres. This may cause scattering of light and loss of transparency, eventually leading to cataract[11]. According to Sacca *et al*, cataract was most frequently observed among those who were exposed to sunlight and ultra violet radiations[12]. Non enzymatic antioxidants namely ascorbic acid and glutathione are abundantly present in the ocular cells to combat the oxidative stress. Lipid peroxidation may alter the arrangement and worsen the interactions among lipid molecules and also between lipid and protein molecules[13].

With these perceptions, objective of the present study was framed to determine the levels of non enzymatic antioxidants and lipid peroxidation in cataract men with and without clinical complications namely, diabetes mellitus and hypertension.

Methodology

Human cataractous eye lens samples for the present investigation were collected from the ophthalmic centres in and around Coimbatore. This research work was approved by Avinashilingam Institutional Human Ethical Committee (HEC.2011.26) and informed consent was obtained from each

subject. The investigation was performed on 45 cataractous eye lens samples from men. They were categorised into apparently normal cataract men (ACM), diabetic cataract men (DCM) and hypertensive cataract men (HCM) groups. Men who had only cataract without any other clinical complications were included in the group of apparently normal cataract men (n=15). Diabetic cataract men group (n=15) comprised of subjects who suffered from only diabetes mellitus and cataract whereas the group of hypertensive cataract men (n=15) included those men with only hypertension and cataract. All the subjects included in the study were above 50 years of age and their cataract lenses were removed by conventional surgery. The removal of cataract eye lens through laser surgery was excluded as they would be in the fragmented form which is not suitable for the present research.

The cataractous lenses obtained from the selected subjects for the study were homogenised in 0.1 M phosphate buffer. The homogenate was centrifuged and the supernatant was taken for the estimation of non enzymatic antioxidants. The levels of reduced glutathione in cataractous lenses of the selected subjects were estimated by Moron *et al*[14] and ascorbic acid levels by Omaye *et al*[15]. Status of lipid peroxidation in cataractous lenses of the selected subjects was estimated by Niehaus and Samuelsson[16].

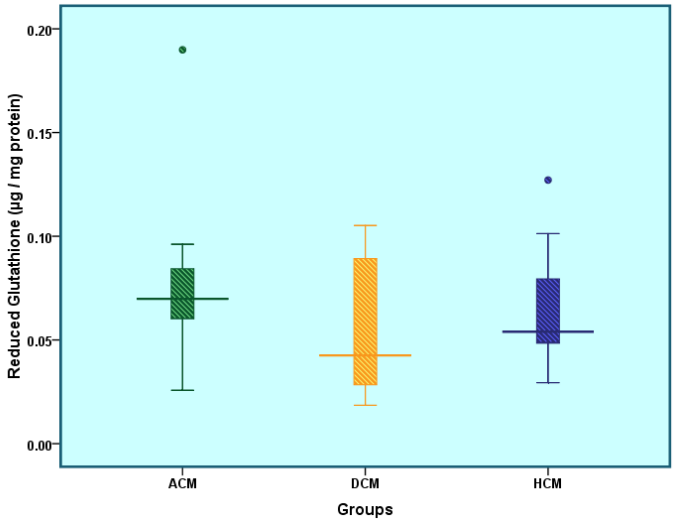
Statistical analysis for the data obtained was performed using SPSS 16 version package. The data were first and foremost subjected to normality test (Shapiro Wilk test). Since the sample size was small and the data were not normally distributed, they were interpreted by a non parametric test (Kruskal Wallis test) with the significance level at $p < 0.05$.

Results and Discussion

The present investigation was performed to compare the levels of non enzymatic antioxidants and lipid peroxidation in apparently normal cataract men and clinically complicated cataractous men which were represented by box plot graphs. These graphs link the box, median line and the error bar width.

Figure-1 depicts the levels of reduced glutathione in the eye lens among cataractous men. However, there was no significant difference in the levels of reduced glutathione in the cataractous eye lens of DCM group albeit they showed a decrease in their levels than the ACM group. The cataractous lens extracted from men belonging to diabetic and hypertensive group showed no significant difference among them in the levels of reduced glutathione. During cataractogensis it has been reported that there is a decrease in the amount of glutathione in the eye lens[17]. When there is a decline in the levels of glutathione inside the lens nucleus, it may lead to the accumulation of ascorbic acid degradation products that might alter the functions of crystalline proteins[18].

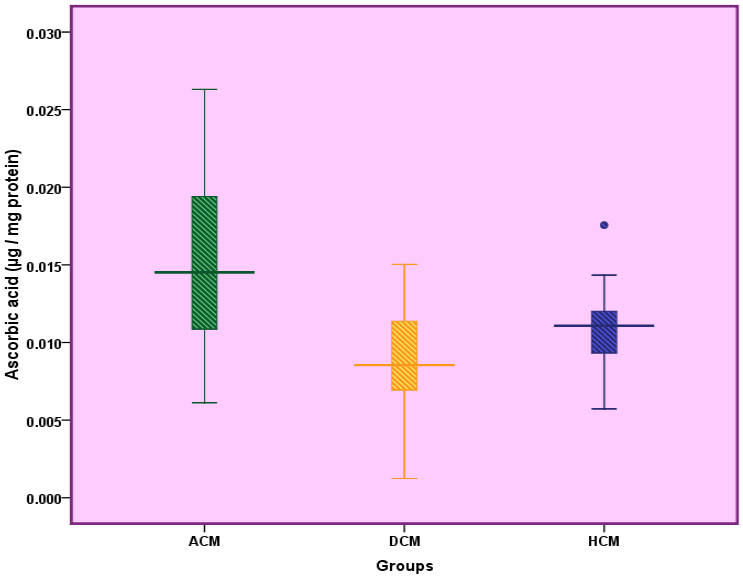
The levels of ascorbic acid in the eye lens among cataractous men are presented in figure-2. A significant decrease ($p < 0.05$) was observed in the cataractous eye lens extracted from diabetic cataract men (DCM) and hypertensive cataract men (HCM) when compared to apparently normal cataract men (ACM). But there was no significant difference between the diabetic cataract men and hypertensive cataract men. According to Tarwadi *et al* there may be a drop in the ability of eye lens to fight the oxidative tension due to the deficiency of β-carotene, ascorbic acid and polyphenols in their diet[19]. Ascorbic acid in the ocular lens precludes the damage to cation pump and decreases the photoperoxidation of membranes[20,21]. They also help in the regulation of expression of genes that are involved in apoptosis and DNA repair mechanism[22]. Cataractous lenses may have decreased levels of ascorbic acid as it may be more prone to oxidation in cataractous condition[23].



ACM: Apparently Normal Cataract Men (n=15), DCM: Diabetic Cataract Men (n=15), HCM: Hypertensive Cataract Men (n=15)

Figure-1
Levels of Reduced Glutathione in Eye Lens among Cataractous Men

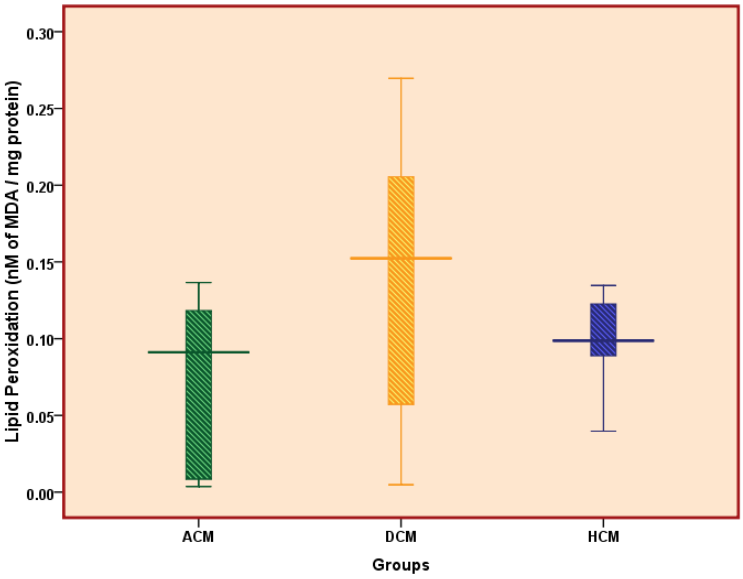
Figure-3 shows the level of lipid peroxidation in the eye lens among cataractous men. Lipid peroxidation status was found to be significantly increased ($p < 0.05$) in the cataractous eye lens extracted from DCM group when compared to ACM group whereas there was no significant difference observed between clinically complicated groups. The increased levels of reactive oxygen species due to oxidative stress may lead to cataract formation and damage the mitochondria[24]. When there is elevated oxygen content in the eye lens, there may be an increase in its growth which in turn may decline its potential to combat oxidative stress and lead to lens opacification[25].



ACM: Apparently Normal Cataract Men (n = 15), DCM: Diabetic Cataract Men (n = 15), HCM: Hypertensive Cataract Men (n = 15)

Figure-2

Levels of Ascorbic Acid in Eye Lens among Cataractous Men



ACM: Apparently Normal Cataract Men (n = 15), DCM: Diabetic Cataract Men (n = 15), HCM: Hypertensive Cataract Men (n = 15)

Figure-3

Level of Lipid Peroxidation in Eye Lens among Cataractous Men

Conclusion

The variations in the levels of non enzymatic antioxidants and lipid peroxidation in the cataractous eye lens among apparently cataract men and clinically complicated cataract men indicated that the cataractous lenses are more liable to oxidative stress. From the present investigation, it was found that the groups with clinical manifestation are more susceptible to undergo oxidation in their cataractous eye lens than the apparently normal cataract men.

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