Research Article

iMedPub Journals
www.imedpub.com

Annals of Clinical and Laboratory Research
ISSN 2386-5180

2018
Vol.6 No.1:217

DOI: 10.21767/2386-5180.1000217

Estimation of Serum Ascorbic Acid (Vitamin C) in the Age Related (Senile) Cataract: A Case Control Study

Saseekala Angirekula[1], Lalitsiri Atti[1] and Srihari Atti[2*]

[1]Sri Venkateswara Medical Government College, Tirupati, Andhra Pradesh, India

[2]ACSR Government Medical College, Government General Hospital, Nellore, SPSR Nellore, Andhra Pradesh, India

[*]**Corresponding author:** Dr. Srihari Atti, ACSR Government Medical College, Government General Hospital, Nellore, SPSR Nellore, Andhra Pradesh, India, Tel: +919440333221; E-mail: srihariatti@gmail.com

Received: January 01, 2018; **Accepted:** January 29, 2018; **Published:** February 08, 2018

Citation: Angirekula S, Atti L, Atti S (2018) Estimation of Serum Ascorbic Acid (Vitamin C) in the Age Related (Senile) Cataract: A Case Control Study. Ann Clin Lab Res Vol.6: No.1: 217.

Abstract

Aim: The aim was to estimate the serum Ascorbic Acid (Vitamin C) level, in the development of Age Related (Senile) Cataract, a common cause of curable blindness globally, especially in India.

Materials and methods: This was a case control study in the age group of 45-70 years with 100 patients of age related cataract and 100 age matched normal patients as a control group with estimation of the level of vitamin C in the serum of the study group and the control group.

Results: Serum ascorbic acid values were 0.54 ± 0.23 mg/dL (30.68 ± 13.07 umol/L) in senile cataract and 0.83 ± 0.35 mg/dl (47.16 ± 19.89 umol/L) in controls with a statistical difference between the cases and controls showing a significant decrease in the level of vitamin C in the serum of age related cataract patients compared with the control group.

Conclusion: The present study with the low levels of Vitamin C in serum of age related cataract than in controls shows that the reduced antioxidant status defence mechanism due to the reduced serum Ascorbic acid (Vitamin C) may play a role in the development of the senile cataract.

Keywords: Age related (Senile cataract); Ascorbic acid (Vitamin C); Serum; Blindness

Introduction

According to WHO, an estimated 180 million people worldwide are visually disabled, in which about 45 million are blind and 135 million are with visual impairment [1-4]. There are about 20 million blind people in India with 80.0% of this blindness due to preventable causes. Cataract in India is the most important cause of preventable blindness accounting to 63.7% and there is a cataract backlog of about 12 million with an increase at an estimated rate of 3.8 million annually [5]. India accounts for about 20% of the global blindness, with cataracts being the main cause [4].

Cataract is the major and leading cause of curable blindness in the world today. A cataract occurs when the normally clear and transparent lens in the eye becomes cloudy and so, Cataract describes any lens opacity from the smallest dot to complete opacification. It is classified etiologically as congenital and acquired types. Age related (Senile) cataract is the commonest clinical presentation in the acquired cataract. The prevalence of senile cataract increases with age, with 65% in the age of 50 to 59 years and in all the people aged over 80 years. Most cataracts, which develop slowly with normal aging, also are related to genetic diseases, medical conditions such as diabetes and other factors such as poor nutrition, radiation, steroids, smoking, alcohol, eye trauma or other eye surgeries [2,3].

The ocular lens with continuous exposure to UV light and ambient oxygen is at a higher risk of photo-oxidative damage due to the free oxygen radicals, generated in the course of normal routine metabolic activities of the lens. So, the free radicals playing an important role in the oxidation of the lens proteins lead to lens opacification resulting in cataract. Body defense mechanism with the antioxidants like Glutathione S-Transferase (GST), Ascorbic acid (Vitamin C), Vitamin E etc. which disposes, scavenges and suppresses the formation of free radicals or opposes the actions of the free radicals to minimize this photo oxidative damage to the ocular lens also play an important role. Thus modifiable risk factors of cataract are to be identified to establish the preventive measures [6].

Human studies demonstrated the protective role of antioxidant vitamins with their increased intake or increased serum concentrations against the degeneration of the ageing lens after oxidative damage [7-9]. An increase in ocular lens protein modification due to chronic oxidative stress with the lack of optimum intake of antioxidant micronutrients by the lens and surrounding fluids is an important factor in cataract development [10-12].

Vitamin C acts as an effective physiological antioxidant at lower partial pressures of oxygen to protect against diseases

and degenerative processes caused by oxidative stress [13]. Vitamin C has better scavenging activities in vivo than the other antioxidant enzymes due to its presence intracellularly and extracellularly [14]. Plasma ascorbic acid acts an endogenous antioxidant to protect the lipids from detectable perioxidative damage induced by aqueous peroxyl radicals [15].

There is a strong biological importance of vitamin C in the lens, as it is found at concentrations in lens or aqueous of about 20 to 30 fold than the plasma [16] and even higher in the vitreous [17]. Early studies in India (Indian Studies) reported that vitamin C concentrations were higher in the normal lenses compared with the mature cataracts [18] and noted that vitamin C levels in the Indian population were considerably lower than in western populations. Vitamin C is a powerful reducing agent protecting the lens from oxidative stress and acts synergistically with vitamin E to maintain the antioxidant activity of glutathione [19].

There was a significant association between vitamin C and age-related cataract risk in the American studies, but European studies from Spain [20,21], Sweden [22,23] and Italy [24], showed no significant association. Further cohort studies are required to confirm this association between serum ascorbate with the risk of age-related cataract. So, the aim of our study was to assess the status of the Ascorbic Acid (Vitamin C) as an antioxidant in the development of the cataract.

Aim

The aim was to evaluate the serum ascorbic acid (vitamin c) level in the development of age related (senile) cataract, a common cause of curable blindness globally, especially in India.

Materials and Methods

This was a case control study conducted in the departments of Biochemistry and Ophthalmology, SV Medical College (Govt.) and SVRR govt. General Hospital, Tirupati, AP, India, in the age group of 45-75 years with 100 patients with a clinical diagnosis of age related (senile) cataract and 100 age matched normal patients without any lens opacities. Informed consent was taken from all the patients of senile cataract and age matched normal patients. A provisional clinical diagnosis of age related cataract was assigned based on the clinical ophthalmic examination. The cases and the controls with a history of Diabetes Mellitus, Hypertension, systemic diseases, trauma etc. were excluded. The study was approved by the institute ethical committee.

Blood samples collection

10 mL of fasting blood sample drawn from a peripheral vein especially from the antecubital vein was taken from 100 patients of age related cataract and 100 age matched normal patients without cataract. For the separation of the serum, 5 ml of blood was taken into a plain vial first and then allowed to clot. Then this clotted blood was centrifuged at 3000 rpm for 5 minutes. This separated serum was used to estimate vitamin C on the same day. 1 mL of blood was taken into a fluoride vial to estimate blood glucose to exclude diabetes mellitus.

Assay of ascorbic acid: S.T.O May J.o. Turnball, 1979. Determination after devitalization with 2,4 dinitrophenylhydrazine

Principle: Ascorbic acid is oxidized by copper to form dihydroascorbic acid and diketoguloric acid. These products are treated with 2,4 DNP to form a derivative of bis 2,4 dinitrophenylhydrazine. This compound in conc. H_2SO_4 undergoes a rearrangement to form a product with an absorption band that is measured at 520 nm. This reaction is seen in the presence of Thiourea to provide an unduly reducing reaction which helps to produce a inter substance from non-ascorbic acid chromogens.

Reagents: 1) TCA (Trichloroacetic Acid) 5% and 10% in distilled water

2) 2,4 dinitrophenylhydrazine/Thiourea/Copper solution. (add 0.4 g Thiourea, 0.05 g $CuSo_4$ $5H_2O$ and 30 g 2, 4 dinitrophenylhydrazine and bring to a total volume of 100 mL with 9 NH_2SO_4.

3) 65% H_2SO_4

Procedure: 1 mL of plasma and 1 mL of Ice cold 10% TCA were mixed well and centrifuged for 20 minutes at 3500 rpm. 0.5 ml of supernatant and 1 mL of DTC were taken and incubated for 3 hrs at 37 degrees centigrade. To convert this into a interarrange product, 0.75 mL of ice cold 65% H_2SO_4 was added. Then mixed well and cooled. The final product was read with 520 nm. Instruments used were Bio-systems-BTS 320 Photometer.

Results

The study results in the 100 cases of senile cataract and 100 age matched controls in the age group of 45-70 years were evaluated using simple statistical methods. Ascorbic acid was measured in mg/dl (umol/L).

Serum ascorbic acid values were 0.54 ± 0.23 mg/dL (30.68 ± 13.07 umol/L) in senile cataract and 0.83 ± 0.35 mg/dL (47.16 ±19.89 umol/L) in controls with a statistical difference between the cases and controls showing a significant decrease in the level of vitamin C in the serum of age related cataract patients compared with the control group **(Tables 1 to 3)**.

Discussion

India accounts for about 20% of the global blindness, with cataract as the principal cause [25]. Indian Population-based studies reported high prevalence rates of cataract [26-28] compared to the western populations. Environmental, nutritional, and genetic factors may be important factors for these high rates, but with limited information on these in the Indian setting, particularly on antioxidants (especially vitamin C), which is considered to play a key role in protecting the lens

from oxidative stress [29]. Inverse association between cataract and plasma Vitamin C levels and other antioxidants was reported [30].

Cataract is an important visual problem of old people with a substantial health care cost in many countries. Age Related (Senile) Cataract is not only the most common type of Acquired Cataract, but also the most common cause of curable blindness in the developing nations. The etiology of age-related cataract is complex and multifactorial with its patho-physiology generated by the combined effects of genetic and environmental factors.

Table 1 Serum vitamin C mg/dL in cases.

S. No.	Range		Mean		SD	
	mg/dL	umol/L	mg/dL	umol/L	mg/dL	umol/L
1	0.2 -1.1	11.36-62.5	0.54	30.68	± 0.23	± 13.07

Table 2 Serum vitamin C mg/dL in controls.

S. No.	Range		Mean		SD	
	mg/dL	umol/L	mg/dL	umol/L	mg/dL	umol/L
1	0.3-1.4	17.05-79.55	0.83	47.16	± 0.35	±19.89

Table 3 Comparison of serum vitamin C mg/dL in cases and controls.

S. No.	Range		Mean		SD	
	mg/dL	umol/L	mg/dL	umol/L	mg/dL	umol/L
1.Cases	0.2 -1.1	11.36-62.5	0.54	30.68	± 0.23	± 13.07
2.Controls	0.3 -1.4	17.05-79.55	0.83	47.16	± 0.35	± 19.89

Most of the studies investigating the risk factors for cataract were conducted in the United States, and there is less information on the possible role of these risk factors in European and Asian populations. Many epidemiologic studies were published exploring the relationship of vitamin C and the risk of cataract, but, the results were inconsistent. For serum ascorbate level with the risk of senile cataract, 10 studies (three prospective studies, six cross-sectional studies and one case-control study) were conducted involving a total of 7305 cataract cases with seven in United States, two in India and one in Spain, but inverse association of serum ascorbate levels with risk of cataract was reported in only four studies.

The main difficulty in the studies to evaluate the antioxidant status in the development of Senile cataract was the inability to measure the antioxidants directly in ocular lens in vivo [6,31] and so, many investigators used the plasma or serum RBC's of the patients to evaluate the status of the antioxidants in the senile cataract [31-33].

In our case-control study of 100 cases of age related (Senile) cataract and 100 age matched controls in the age group of 45 to 70 years, Serum Ascorbic acid values were 0.54 ± 0.23 mg/dL (30.68 ± 13.07 umol/L) in senile cataract and 0.83 ± 0.35 mg/dL (47.16 ± 19.89 umol/L) in controls with a statistical difference between the cases and controls showing a significant decrease in the level of vitamin C in the serum of age related cataract patients compared with the control group.

In the study of Dherani M et al. [30], with a cross sectional study in India of a total of 1443 patients aged ≥ 50 years, 94% were interviewed, 87% attended an eye examination, and 78% gave a blood sample; 1112 (77%) were included and Compared with the levels in Western populations, Vitamin C was inversely associated with cataract with the serum Ascorbate of 0.64 (0.48 -0.85) (the highest (≥ 15 micromole/L) compared with the lowest (≤ 6.3 micromole/L). In the study of Ravilla Ravindran et al. [34], a case control study in India of a total of 5638 patients aged ≥ 60 years; Plasma vitamin C which was measured using an enzyme-based assay in plasma stabilized with metaphosphoric acid, was inversely associated with cataract with the value of 0.61 mg/dL (0.51-0.74). In the study of Kamath et al. [35], a study in INDIA of a total of 131 participants in each group, the patients with denser cataracts had a significantly lower level of serum Vitamin C. (0.91 ± 0.40 mg/dL in Group 1 with cataract versus 1.16 + 0.50 mg/dL in Group 2 without minimal cataract.

In the study of Maria Pastor Valero et al. [20], a case control study in Spain of 343 patients in the age group of 55-74 years, the serum ascorbate level was 0.30 (0.18-0.51) and serum vitamin C level above 49 micromole/L were associated with a 64% reduced risk for cataract, showing the evidence vitamin C protective role on the aging lens. In the study of Simon et al. [36], a cross sectional study in US of 416 patients in the aged group of 60-74 years, the serum ascorbate level was 0.74 (0.56-0.97). In the study of Ferringo et al. [37], a cross sectional study in US of 710 patients in the aged group of

55-75 years, the serum Ascorbate level was 0.74 (0.42-1.15). In the study of Wei et al. [38], highest serum ascorbate levels versus lowest levels were significantly associated with the risk of cataract 0.704 (0.564-0.879). In the study of Van Der Pols et al. many experimental studies showed a protective effect of vitamin C in age-related cataract and suggested that maintenance of sufficient plasma vitamin C may prevent oxidative damage in the lens with a need for more research to confirm the importance of the vitamin C in the eye lens.

Conclusion

In summary, our study and other studies showing the inverse association of serum ascorbate level with the risk of senile cataract suggested that the serum ascorbate may reduce the risk of senile cataract.

The present study with the low levels of Vitamin C in serum of age related cataract than in controls, shows an increased demand for antioxidants vitamins to meet the oxidative stress and damage and so, it may be justified to use antioxidants to raise their serum level thereby increasing its aqueous humour level to prevent senile cataract.

References

1. Park K (2011) Epidemiology of chronic non-communicable diseases and conditions- blindness In: K Park (eds). Park's Textbook of Preventive and Social Medicine (21st edn). Bhanot Publishers. India. pp. 370-372.
2. Sihota R, Tandon R (2007) The causes of and prevention of blindness. In: Ramanjit Sihota, Radhika Tandon (eds) Parsons' diseases of the eye. (20th edn) New Delhi: Elsevier. India. pp. 523-529.
3. Khurana AK, Khurana B (2012) Community ophthalmology. In: AK Khurana (eds) Comprehensive Ophthalmology. (5th edn). New Age International Publisher. India. pp. 474-484.
4. Foster A, Resnikoff S (2005) The impact of vision 2020 on global blindness. Eye (Lond) 19: 1133-1135.
5. Government of India (2002) National survey on blindness and visual outcome after cataract surgery, 2001-2002 Vol 77. National Programme for Control of Blindness, Ministry of Health, Government of India, New Delhi, India.
6. Berman ER (1991) Biochemistry of the eye. Plenum Publishing Corporation, New York, USA.
7. Jacques PF, Chylack LT Jr, McGandy RB, Hartz SC (1988) Antioxidant status in persons with and without senile cataract. Arch Ophthalmol 106: 337-340.
8. Jacques PF, Chylack LT Jr, Hankinson SE (2001) Long-term nutrient intake and early age-related nuclear lens opacities. Arch Ophthalmol 119: 1009-1019.
9. Van der Pols JC (1999) A possible role for vitamin C in age-related cataract. Proc Nutr Soc. 58: 295-301.
10. Varma SD (1991) Scientific basis for medical therapy of cataracts by antioxidants. Am J Clin Nutr 53: 335S-345S.
11. Taylor A, Jacques PF, Epstein EM (1995) Relations among aging, antioxidant status, and cataract. Am J Clin Nutr 62: 1439S-1447S.
12. Taylor A, Jacques PF, Chylack LT Jr (2002) Long-term intake of vitamins and carotenoids and odds of early age-related cortical and posterior subcapsular lens opacities. Am J Clin Nutr 75: 540-549.
13. Burton GW, Ingold KU (1984) Beta-carotene: An unusual type of lipid antioxidant. Science 224: 569-573.
14. Karthikeyan J, Rani P (2003) Enzymatic and non-enzymatic antioxidants in selected piper species. Ind Exp Biol 41: 135-140.
15. Frei B (1994) Reactive oxygen species and antioxidant vitamins: Mechanisms of action. Am J Med 97: 5S-13S.
16. Taylor A, Jacques PF, Nowell T (1997) Vitamin C in human and guinea pig aqueous, lens and plasma in relation to intake. Curr Eye Res 16: 857-864.
17. Shui YB, Holekamp NM, Kramer BC (2009) The gel state of the vitreous and ascorbate-dependent oxygen consumption: Relationship to the etiology of nuclear cataracts. Arch Ophthalmol 127: 475-482.
18. Consul BN, Mathur GB, Mehrotra AS (1968) Aqueous humor ascorbic acid in normal, cataractous and aphakic Indian subjects. J All India Ophthalmol Soc. 16: 105-108.
19. Shang F, Lu M, Dudek E (2003) Vitamin C and vitamin E restore the resistance of GSH-depleted lens cells to H2O2. Free Radic Biol Med 34: 521-530.
20. Valero MP, Fletcher AE, De Stavola BL, Vioque J, Alepuz AC (2002) Vitamin C is associated with reduced risk of cataract in a Mediterranean population. J Nutr. 132: 1299-1306.
21. Rodriguez-Rodriguez E, Ortega RM, Lopez-Sobaler AM (2006) The relationship between antioxidant nutrient intake and cataracts in older people. Int J Vitam Nutr Res 76: 359-366.
22. Rautiainen S, Lindblad BE, Morgenstern R, Wolk A (2010) Vitamin C supplements and the risk of age-related cataract: A population-based prospective cohort study in women. Am J Clin Nutr 91: 487-493.
23. Zheng Selin J, Rautiainen S, Lindblad BE, Morgenstern R, Wolk A (2013) High-dose supplements of vitamins C and E, low-dose multivitamins, and the risk of age-related cataract: A population-based prospective cohort study of men. Am J Epidemiol 177: 548-555.
24. Tavani A, Negri E, La Vecchia C (1996) Food and nutrient intake and risk of cataract. Ann Epidemiol 6: 41-46.
25. Resnikoff S, Pascolini D, Etya'ale D (2004) Global data on visual impairment in the year 2002. Bull World Health Organ.82: 844-851.
26. Krishnaiah S, Vilas K, Shamanna BR (2005) Smoking and its association with cataract: Results of the Andhra Pradesh eye disease study from India. Invest Ophthalmol Vis Sci. 46: 58-65.
27. Nirmalan PK, Krishnadas R, Ramakrishnan R (2003) Lens opacities in a rural population of southern India: the Aravind Comprehensive Eye Study. Invest Ophthalmol Vis Sci 44: 4639-4643.
28. Vashist P, Talwar B, Gogoi M (2011) Prevalence of cataract in an older population in India: the india study of age-related eye disease. Ophthalmol 118: 272-278.
29. Linetsky M, Shipova E, Cheng R, Ortwerth BJ (2008) Glycation by ascorbic acid oxidation products leads to the aggregation of lens proteins. Biochim Biophys Acta. 1782: 22-34.

30. Dherani M, Murthy GV, Gupta SK, Young IS, Maraini G, et al. (2008) Blood levels of vitamin C, carotenoids and retinol are inversely associated with cataract in a North Indian population. Invest ophthalmol Vis Sci 49:3328-3335.

31. Mohan M, Sperduto RD, Angra SK, Milton CR, Mathur RL, et al. (1989) The India-US case control study group. India-US case-control study of age related cataracts. Arc Ophthalmol 107: 670-676.

32. Garg R, Varma M, Mathur SP, Murthy PS (1996) Blood Lipid peroxidation products and antioxidants in senile cataract. Ind J Biochem 11: 182-186.

33. Knekt P, Heliovaara M, Rissanen A, Aromaa A, Aaran RK, et al. (1992) Serum antioxidant vitamins and risk of cataract. Br J Med 305: 1392-1394.

34. Ravilla DR, Vashist P, Gupta S, Young IS, Maraini G, et al. (2011) Inverse association of vitamin C with cataract in older people in India. Ophthalmol 118: 1958-1965.

35. Kamath Y, Bhat S, Iqbal S, Rao LS (2017) The association of age-related cataract and serum Vitamin C. Indian J Clin Exp Ophthalmol 3: 287-290.

36. Simon JA, Hudes ES (1999) Serum ascorbic acid and other correlates of self-reported cataract among older Americans. J Clin Epidemiol 52: 1207-1211.

37. Ferrigno L, Aldigeri R, Rosmini F (2005) Associations between plasma levels of vitamins and cataract in the Italian-American Clinical Trial of Nutritional Supplements and Age-Related Cataract (CTNS): CTNS report #2. Ophthalmic Epidemiol 12: 71-80.

38. Wei L, Liang G, Cai C, Jin LV (2015) Association of vitamin C with the risk of age-related cataract: a meta-analysis. Acta Ophthalmologica 94: e170-e176.