

Original Article**Association of Plasma and Aqueous Humor Ascorbic Acid Level with Morphology Pattern of Age-Related Cataract**

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Abstract**Background**

Age-related cataract is one of the most important causes of preventable blindness among elderly population in the world. Among various causes of cataract, the level of ascorbic acid in the plasma and aqueous humor is an important parameter that can prevent cataract formation. The level of ascorbic acid in the plasma and aqueous humor is also an indicator of various morphological pattern of cataract.

Materials and Methods

A hospital-based cross-sectional study was conducted among 136 patients with immature age-related cataract (≥ 50 years) who underwent Manual Small Incision Cataract Surgery in the Ophthalmology Department of BPKIHS for the duration of 1 year. Plasma and aqueous humor ascorbic acid levels were determined by standard competitive ELISA test.


Results

Ascorbic acid concentrations in aqueous humor as well as plasma were statistically significant with the nuclear type of cataract ($p < 0.001$). There was a positive correlation between plasma and aqueous ascorbic acid level with $r = 0.532$ and $p < 0.001$.

Conclusion

The finding of the study conclude that nuclear type of cataract has a strong association with both aqueous humor and plasma ascorbic acid level.

Keywords: Aqueous humor, Ascorbic acid, Cataract, Plasma

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Introduction

The prevalence of Blindness (presenting BCVA 3/60 in the better eye) in Nepal was 2.5% in people aged ≥ 50 years where cataract accounts 62.2% [1]. Exact etiology of senile cataract is not known, there are various factor causing it, which includes environmental, genetic, nutritional. The role of ascorbic acid is an important antioxidant that prevents cataract formation [2]. The ascorbic acid is present in both aqueous and serum, where the concentration of aqueous ascorbic acid is 20-30 times more than the plasma ascorbic acid. The patient with senile cataract has reduced concentration of aqueous ascorbic acid [3] as well as in eyes of animals with experimental cataract [4]. High concentration of ascorbic acid in the aqueous humor represents a filter that prevents Ultraviolet light penetration in the lens and thus protects tissue from oxidative damage. The cation pumps damage caused by UV radiation as well as photoperoxidation in the membranes is reduced by ascorbic acid in the lens [5].

As the global life expectancy is in the rise so the burden of cataract is increasing too. Only available treatment of cataract till today is surgery. Knowing the modifiable risk factors can delay the formation of cataract which can reduce the surgical cost as well as improve the quality of life. Because the incidence of cataract in the developing world will be more than the cataract surgical rate as a result of which cataract blindness will continue to increase in coming decades. In rural China, 35.7% of the cataract patient underwent cataract surgery [6]. It is estimated that if the onset of cataract is delayed by 10 years than the need for cataract extractions would be diminished by one-half [7]. For the above mentioned reasons it is necessary to research for the pharmacological therapy for the cataract management. In this research we analyzed the association of plasma and aqueous ascorbic acid level and grade different degrees of nuclear, cortical, and subcapsular cataract according to the Lens Opacities Classification System, version II (LOCS III) [8].

This study was designed with the objective of assaying the levels of plasma and aqueous humor ascorbic acid in subjects with the various morphology of cataract.

Materials and Methods

This study was a hospital based cross-sectional study conducted in the Department of Ophthalmology & Department of Biochemistry, B.P. Koirala Institute of Health Sciences, Dharan, Nepal from February, 2016 to January, 2017. The study was initiated after acquiring approval from

the Institutional Ethical Review Board, B.P.Koirala Institute of Health Sciences. A verbal consent was acquired after the patient or patient party was explained about the study including the details about the procedure, its advantages and disadvantages. Patient with the diagnosis of age-related immature cataract undergoing Manual Small Incision Cataract Surgery were included in the study. The study considered 95% CI and 90% power with pretested value of correlation between aqueous and plasma Ascorbic acid was 0.289 which estimated the sample size of 122 but the study used consecutive sampling technique to recruit sample units for one year. Therefore, the sample size of the study following consecutive sampling technique was 136. A complete ophthalmological examination was done, which include: distance visual acuity testing using the Snellen method, near visual acuity using Jaeger's tables, intraocular pressure measurement with applanation tonometry, slit lamp examination of anterior and posterior segment of the eye with artificial mydriasis. The Lens Opacity Classification System III (LOCS III) [8] was used by an ophthalmologist to classify the various morphological pattern of age-related cataract. In this study only age-related immature cataract were enrolled after exclusion of complicated cataracts, post traumatic cataracts, glaucoma, intraocular inflammation, diabetes mellitus, systemic hypertension, leprosy, tuberculosis, smokers, pure vegetarians and those receiving systemic vitamin supplementations. Aqueous humor samples were obtained before creation of the scleral tunnel. A small amount (0.2 mL) of aqueous humor was aspirated through *ab externa* (outside- in) limbal paracentesis with a 27 gauge needle on a tuberculin syringe. The extracted quantity of aqueous humor was replaced with the same amount of isotonic Ringer lactate solution, and the operating procedure Manual Small Incision Cataract Surgery was resumed in regular fashion. At the same time, 3 ml of blood was collected from an antecubital vein and was transferred EDTA vial and was centrifuged at 10g for 10 minutes and plasma was separated. The plasma was stored at -20°C till the biochemical analysis (Enzyme Linked Immunosorbent Assay) was done in Immunoassay laboratory of the B.P.Koirala Institute of Health Sciences, Dharan.

After collection of the data, a coding was done and then entered the data at Microsoft excel 2007 and converted it into SPSS 11.5 for statistical analysis. Normality of the data was tested by Kolmogorov Smirnov test. After normality test Values of the level of ascorbic acid in the aqueous



humour and plasma were expressed as mean \pm SD present in various morphological pattern of age-related immature cataract. P-values were calculated by using independent t-test, Mann-Whitney test, Pearson's correlation coefficients to find out the significant differences at 95% confidence interval.

Results

The total number of patients in our study was 136. The mean age in years was found 68.61 with SD 8.8. The frequency of the male was found more than the female. Among the morphology of the cataract the nuclear, Cortical and posterior subcapsular had present in 118 (86.7%), 51 (37.5%), 33 (33%) case respectively. Mean aqueous ascorbic acid concentration was 26.48 ± 12.67 mg/dl, ranging from 4mg/dl to 56 mg/dl. Similarly, mean plasma concentration of ascorbic acid in patients was 1.18 ± 0.56 (mg/dl) ranging from 0.4 mg/dl to 4.3 mg/dl.

Table 1: Baseline characteristics of patients.

Characteristics	Category	Mean \pm SD/n	Percentage
Mean Age (yrs)	Male	68.61 \pm 8.8	
	Female		
Gender	Male	92	67.6%
	Female	44	32.7%
Morphology of Cataract			
Nuclear	Present	118	86.7%
	Absent	18	13.3%
Cortical	Present	51	37.5%
	Absent	85	62.5%
Posterior Subcapsular	Present	33	24.26%
	Absent	103	75.74%
Aqueous Ascorbic Acid level (mg/dl)		26.48 \pm 12.6	
Plasma Ascorbic Acid Level (mg/dl)		1.18 \pm 0.56	

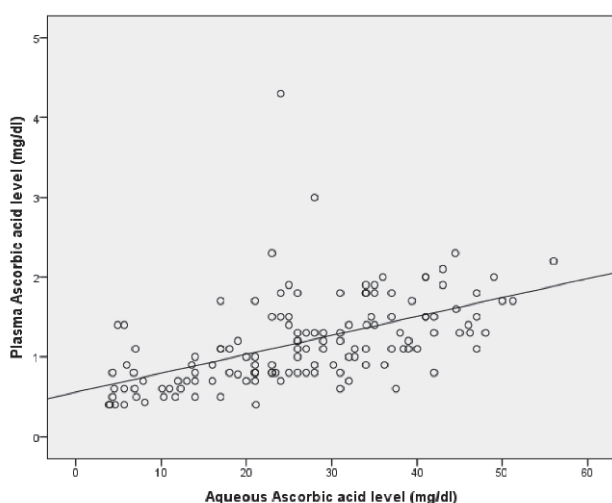


Figure 1: Correlation between aqueous ascorbic acid and plasma ascorbic acid.

Figure 1 shows the positive correlation between the plasma and aqueous ascorbic acid level which is statistically significant at $p < 0.001$ with Pearson correlation coefficient $r = 0.532$

Table 2: Relationship of aqueous Ascorbic Acid with various morphology of cataract

Type of Cataract	Result	Mean \pm SD	p-value	remarks
Nuclear	Present	29.02 \pm 11.24	<0.001*	Significant
	Absent	9.90 \pm 8.47		
Cortical	Present	29.21 \pm 10.94	0.087*	Not significant
	Absent	24.85 \pm 13.41		
Posterior Subcapsular	Present	26.09 \pm 9.29	0.681*	Not significant
	Absent	26.61 \pm 13.62		

*Mann-Whitney test

Relationship between various morphology of cataract with the aqueous ascorbic acid level is shown in table 2. There was a significant difference in the level of aqueous ascorbic acid level for nuclear Cataract (M=29.02, SD 11.24) and no nuclear cataract (M=9.90,SD=8.47). Among the morphology of cataract only nuclear cataract was found to be significant at $p < 0.001$ with ascorbic acid level in the aqueous humour but cortical and posterior subcapsular cataract were found to be insignificant. These results suggest that aqueous ascorbic acid level really does have an effect on nuclear morphology of cataract.

Table 3: Relationship of plasma Ascorbic acid with various morphology of cataract

Type of Cataract	Result	Mean \pm SD	p-value	remarks
Nuclear	Present	1.22 \pm 0.48	<0.001*	Significant
	Absent	0.92 \pm 0.90		
Cortical	Present	1.20 \pm 0.49	0.579*	Not significant
	Absent	1.17 \pm 0.60		
Posterior Subcapsular	Present	1.22 \pm 0.66	0.652**	Not significant
	Absent	1.17 \pm 0.53		

*Mann-Whitney test

**Independent t test

Relationship between various morphology of cataract with the plasma ascorbic acid level is shown in table 3. There was a significant difference in the level of plasma ascorbic acid level for nuclear Cataract (M=1.22, SD 0.48) and no nuclear cataract (M=0.92,SD=0.90). Among the morphology of cataract only nuclear cataract was found to be significant at $p < 0.001$ with ascorbic acid level in the plasma but cortical and posterior subcapsular cataract were found to be insignificant. These results suggest that plasma ascorbic acid level really does have an effect on nuclear morphology of cataract.

Discussion

Ascorbic acid is one of the most important antioxidant in the aqueous humor and important for the maintenance of the lens transparency. Human aqueous humor Ascorbic acid concentration is more than 20-fold higher than in the plasma



[9]. Study conducted in Eastern Nepal where plasma and aqueous ascorbic acid were measured, the mean plasma ascorbic acid levels of subjects from the mountainous region was 0.65 mg/dl and that of the patients from sub-tropical lowlands was 0.85mg/dl and mean aqueous ascorbic acid levels of the subjects from the mountainous region was 17.5 mg/dl and that from sub-tropical lowlands was 23.47 mg/dl [10]. In our study, mean plasma concentration of ascorbic acid in patients was 1.18 ± 0.56 (mg/dl) ranging from 0.4 mg/dl to 4.3 mg/dl, which is wider in range compare to other study. Similarly, aqueous ascorbic acid concentration was 26.48 ± 12.67 (mg/dl), ranging from 4mg/dl to 56 mg/dl which is higher in level compare with similar study done by Badhu et. al. in Nepal. Study conducted in India, showed aqueous ascorbic acid of 5.07 mg/dl in normal lens and 4.72 mg/dl in cataracts lens [11]. Ascorbic acid in the aqueous is 20-30 folds that of plasma [12], whereas in our study it is approximately 22 folds.

Role of ascorbic acid as an antioxidant and free radical scavenger is quite important to prevent or delay the development of senile cataracts [13],[2]. Higher plasma levels of ascorbic acid are associated with reduced prevalence of nuclear and posterior sub-capsular cataracts [14].

Strong inverse associations remained for ascorbic acid with nuclear, cortical, and Posterior SubCapsular cataract [12]. Tessier et. al. showed that as ascorbic acid level decreases lens turbidity increases [15]. Study conducted by Miratashi did not showed any significant association between the level of aqueous and plasma ascorbic acid with all kinds of cataract [9].

Different study done in the various part of the world regarding the role of both aqueous and plasma ascorbic acid with different morphology of cataract shows various results, whereas in our study compare with different morphology of cataract, there was only significant association between nuclear cataract with the level of both aqueous and plasma ascorbic acid.

Conclusion

The finding of the study conclude that nuclear type of cataract has a strong association with both aqueous humor and plasma ascorbic acid level. Therefore, ascorbic acid level in aqueous and plasma is considered as a predictor of nuclear type of cataract. The study can be established that supplementation of ascorbic acid (Vitamin C) can reduce the occurrence of nuclear type of age-

related immature cataract. The study recommends a trial of supplementation of ascorbic acid (Vitamin C) to reduce age related immature cataract with nuclear morphology.

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Conflicts of interests: None

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