A STUDY OF ASCORBIC ACID CONCENTRATION IN THE AQUEOUS HUMOR AND INTRA OPERATIVE CHALLENGES DURING CATARACT SURGERY IN PATIENTS WITH PSEUDO EXFOLIATION AS COMPARED TO NORMAL
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ABSTRACT: PURPOSE: To compare the concentration of ascorbic acid in the aqueous humor and serum of cases of pseudoexfoliation and age and sex matched controls and to observe the intra operative challenges faced during cataract surgery in cases of pseudo exfoliation and controls. MATERIALS AND METHODS: It was a case control study with 40 cases of pseudo exfoliation and 40 age and sex matched controls with similar grade of cataract. Procedure involved collection of 0.15ml of aqueous humor in a tuberculin syringe prior to the side port incision, from cases and controls which was and immediately sent for biochemical analysis by spectro photometry. Intra operative difficulties were observed and compared. Serum samples of 5ml each from cases and controls were also analyzed for as corbate concentration and compared. The statistical analysis was done using standard t- test and p value estimation. RESULTS: The ascorbic acid concentration in aqueous humor of cases is significantly lower than in controls and the intra operative challenges during cataract surgery were more in cases than in controls. CONCLUSION: The lowering of ascorbic acid concentration in the aqueous humor of pseudo exfoliation patients emphasizes the role of oxidative damage in the etiology of pseudo exfoliation. The study throws light on the challenges encountered during cataract surgery in pseudo exfoliation. KEYWORDS: Pseudo exfoliation, Ascorbic acid, Spectrophotometry.

INTRODUCTION: Pseudo exfoliation is an age related generalized disorder of the extracellular matrix, characterized by the production and progressive accumulation of a fibrillar extracellular material in many ocular tissues.(1) It is characterized by the presence of flaky pseudo exfoliation material at the pupillary border, anterior lens capsule, zonules, iris and cornea. (1)
It plays an etiological role in open angle glaucoma, angle closure glaucoma, cataract and retinal vein occlusion. It is associated with an increase in serious complications at the time of cataract extraction such as zonular dialysis, capsular rupture and vitreous loss.\(^{(2)}\)

Etiology of pseudo exfoliation is unknown. There is speculation that it may be caused by oxidative damage and the presence of free radicals, although the mechanism of damage is still under study.\(^{(3)}\)

Ascorbic acid is one of the major anti-oxidants in human body and the estimation of it's concentration in aqueous humor and serum of patients with pseudo exfoliation correlates with the aetiological role of oxidative damage. Cataract surgery complication rate is also proven to be higher in pseudo exfoliation patients in several studies. Considering that pseudo exfoliation is a commonly encountered clinical finding in Kerala population, this study becomes significant.

**AIM OF THE STUDY:** This Study Aimed at:
1. Comparing the ascorbic acid levels in aqueous humor and serum of 40 patients with and without pseudo exfoliation, who underwent cataract surgery.
2. Also, the intra operative challenges encountered during cataract surgery in cases and controls were noted and compared.

**METHODS:**

**Design of the study:** Case control study which included consecutive patients with and without pseudo exfoliation who underwent cataract surgery, over a period of one year.

**Sample size:** 40 cases, 40 controls.

**Inclusion Criteria of Cases:** Cases of pseudo exfoliation who were diagnosed by slit lamp examination (Indicated by the presence of powdery pseudoexfoliative material at pupillary border, anterior lens capsule, zonules), and in the angle by gonioscopy, who were posted for cataract surgery.

**Exclusion Criteria:**
1. Patients with pseudo exfoliation with IOP> 30mm Hg,
2. Diabetes mellitus.
3. Other ocular co-morbidities such as uveitis, vascular occlusion.

**Selection of Controls:** Age and sex matched persons with a similar grade of cataract (graded using LOCS-3 classification) as that of cases and operated by the same surgeon.
**Procedure:** (0.1mL) of aqueous humor was aspirated from cases and controls, using a tuberculin syringe at the beginning of small incision cataract surgery (before the step of side port incision), and immediately sent for ascorbic acid assay by spectrophotometry. Simultaneously serum samples of 5mL each were also sent for ascorbate assay by spectrophotometry. The intraoperative challenges encountered in cases and controls were observed and compared. Statistical analysis was done by unpaired t-test using SPSS version 16.

**OBSERVATIONS AND RESULTS: AGE DISTRIBUTION:** Majority of the cases (50.0%) belonged to the age group 61-70 years. 37.50% of the cases were in the age group 71-80 yrs, 7.50% in the age group of 51-60 yrs, and 5.0% in the age group of more than 80 yrs. Controls were age matched for the cases.

**SEX DISTRIBUTION:** Of the 40 cases, 22 were males (55.0%) and 18 were females (45.0%). Controls were sex matched for cases.
TYPE OF CATARACT: Of the 40 cases, 27 had nuclear cataract (67.50%), which was followed in frequency by cortical cataract (12%).

ASCORBIC ACID CONCENTRATION IN AQUEOUS HUMOR: The range of ascorbic acid values obtained in the total study population was (1.08-12.65g/dL).

The mean ascorbic acid concentration in the aqueous humor of cases was 3.005(SD 1.20)mg/dL and that of controls was 6.9482(SD 1.89)mg/dL. The decrease in ascorbic acid in cases was found to be statistically significant when compared to that of controls with a p value of <0.01.

ASCORBIC ACID CONCENTRATION IN SERUM: The ascorbic acid values obtained in the serum of the total study population was in the range (0.24-3.45mg/dL). The mean concentration in cases was 0.937(SD 0.54)mg/dL, and that in controls was 1.08 (SD. 607 mg/dL. Although the mean value in cases was lower than that of controls, the difference was not statistically significant (p>0.05)

INTRAOPERATIVE CHALLENGES DURING CATARACT SURGERY: Of the 40 cases, intra operative challenges were observed in 16. Surgeries in controls were uneventful. Of the observed intra operative challenges, poor pupillary dilatation and zonular dialysis together (ppd+zd) were seen in 44%, poor pupillary dilatation (ppd) in 31%, zonular dialysis (zd) in 18.8%, and hyphaema in 6.2%.
DISCUSSION: The role of oxidative stress in the etiology of pseudo exfoliation is debatable. The concentration of ascorbic acid as a measure of oxidative stress had been found to be lowered in previous studies by Koliakos et al.(4) There is no available literature on such studies in the south Indian population. This study agrees to the findings of the before mentioned study.

| STUDY                          | ASCORBATE CONCENTRATION IN AQUEOUS HUMOR OF PEX CASES(mM/L) | ASCORBATE CONCENTRATION IN AQUEOUS HUMOR OF CONTROLS(mM/L) |
|-------------------------------|-------------------------------------------------------------|----------------------------------------------------------|
| KOLIAKOS AND KONSTAS et al(2002)(4) | 0.86                                                        | 1.15                                                     |
| KOLIAKOS et al(2003)          | 0.75                                                        | 1.19                                                     |
| CURRENT STUDY                 | 0.17                                                        | 0.39                                                     |

Table 1: comparison of ascorbate concentration in aqueous humor of PEX cases in different studies

The vitamin C concentrations in the study population as a whole is far lower than that of western population as assessed by Koliakos et al.(4) But the values are consistent with that of previous Indian studies by GB Mathur, BN Consul and AS Mehrotra(5) and also in a different study by Chatterjee and Ghosh,(6) and Nema and Srivastava.(7)
| NAME OF THE WORKER               | TYPE OF CASES | ASCORBATE IN mg percent |
|---------------------------------|---------------|-------------------------|
| KRONFIELD                       | NORMAL        | 17.20                   |
| PURCELL ET AL                   | NORMAL        | 18.0                    |
|                                 | CATARACT      | 12.10                   |
| BERARDINIS ET AL                | NORMAL        | 17.10                   |
|                                 | CATARACT      | 17.34                   |
| CHATTERJEE AND GHOSH(6)         | CATARACT      | 5.07                    |
| NEMA AND SHRIVASTAVA(7)         | NORMAL        | 5.07                    |
|                                 | CATARACT      | 4.72                    |
| CONSUL, MATHUR AND MALHOTRA(5)  | NORMAL        | 5.2                     |
|                                 | CATARACT      | 4.8                     |

Table 2: Comparison of ascorbate concentration in aqueous humor of cataract patients in various Indian and western studies

In this study, the mean concentration of ascorbate in the serum of PEX cases is 0.937mg/dL and that in the age and sex matched controls is 1.08mg/dL. The values in cases are lower than that of controls, but are not statistically significant.

This is consistent with the study by Zoric et al, done in 2006, the vitamin C concentration in sera was estimated colorimetrically using 2,4-dinitrophenylchloralazine, and results were expressed in micro M/L. The mean serum vitamin C concentration in PEX cases was found to be 52.21±13.42microM/L and that in controls was 50.44±13.26 micro M/L. Although the values in PEX cases is lower than that of controls, the difference is not statistically significant.\(^{(8)}\)

In contrast, according to the study by Yilmaz et al, the serum vitamin C concentrations were significantly lower in cases of XFS when compared with the control group.\(^{(9)}\)

In the current study, the intra operative challenges observed were also higher in cases as compared to controls, the most common being poor papillary dilatation and zonular dialysis. Scorolli et al\((1998)\) studied the rate of intraoperative complications induced by pseudoexfoliation syndrome (PSX) in cataract surgery in 1052 patients and concluded that PSX was associated with a statistically significant increase in intraoperative complications during cataract surgery \(p<0.0001\).\(^{(10)}\) Vickie lee, Anthony Maloof et al\((2002)\) did a study regarding “cataract surgery in Pseudo exfoliation syndrome.” They concluded that cataract patients with pseudo exfoliation have higher rates of complications during and after cataract surgery compared to the patients without pseudo exfoliation.\(^{(10)}\) A study by Abid Naseem et al\((2007)\) showed that cataract surgery in eyes with pseudoexfoliation has higher incidence of operative complications like posterior capsular rupture, zonular dialysis, vitreous loss and intraocular bleeding.\(^{(11)}\) Bayramlar H et al study \((2007)\) showed that in manual small-incision cataract surgery, pseudoexfoliation syndrome has an increased intraoperative posterior capsule complication rate that increases as the level of cataract maturity increases.\(^{(12)}\)
CONCLUSION: The role of oxidative stress in the etiology of pseudo exfoliation in south Indian population has been ascertained by this study. Supplementation of anti-oxidants can be attempted in preventing the occurrence and delaying the progression of pseudo exfoliation. This study can be considered as a fore runner to such interventional supplementation studies.

Also, the study confirms the increased risk of intra operative challenges during cataract surgery in cases when compared to controls. Hence, a meticulous pre-operative evaluation is mandatory in patients with pseudo exfoliation undergoing cataract surgery.

Limitations:
1. Small sample size. The current study includes only 40 cases and 40 controls.
2. The method used for biochemical analysis in the current study is spectrophotometry which is less sensitive when compared to microplate assay, used in previous studies.
3. The current study measures only ascorbic acid concentration, and ignores the other anti-oxidants. Hence it cannot precisely comment on the role of oxidative stress in the etiogenesis PEX.

REFERENCES:
1. Ritch R, Schlötzer-Schrehardt U. Exfoliation syndrome. Surv Ophthalmol 2001; 45: 265–315.
2. Naumann G, Schlötzer-Schrehardt U, Küchle M. Pseudoexfoliation syndrome for the comprehensive ophthalmologist. Intraocular and systemic manifestations. Ophthalmol 1998; 105: 951–968.
3. Schlötzer-Schrehardt U, Koca MR, Naumann G. Pseudoexfoliation syndrome. Ocular manifestation of a systemic disorder? Arch Ophthalmol 1992; 110: 1752–6.
4. Koliakos G, Konstas A, Schlötzer-Scherardt U, Bufidis N, Ringvold A. Ascorbic acid concentration is reduced in the aqueous humor of patients with exfoliation syndrome. Am J Ophthalmol 2002; 134: 879–883
5. BN Consul, GB Mathur, AS Mehrotra; Aqueous humor ascorbic acid in normal, cataractous and aphakic Indian subjects.
6. Chatterjee.B and Ghosh, B .P: Total ascorbic acid in aqueous humor and serum in Indian patients with and without cataract.
7. Nema,H.V. and Shrivastava,S.P.: Ascorbic acid in aqueous humor and serum in normal and mature cataract on Indian subjects.
8. Zoric L. D, Miric, S. Milenkovic, P. Jovanovic, and G. Trajkovic. Pseudoexfoliation syndrome and its antioxidative protection deficiency as risk factors for age-related cataract.European journal of ophthalmology 16, no. 2. 2006 March-April;16(2) 268-73.
9. Yilmaz, Ayça, et al. Serum oxidant/antioxidant balance in exfoliation syndrome. Clinical & experimental ophthalmology.2005 February; 33.1: 63-66.
10. Scorolli L, Scorolli L, Campos EC, Bassein L, Meduri RA. Pseudoexfoliation syndrome: a cohort study on intraoperative complications in cataract surgery. Ophthalmologica.1998; 212: 278-280.
11. Lee V, Maloof A. Cataract surgery in pseudoexfoliation syndrome. Comp. Ophthalm. Update. 2002 Jan; 3(1): 169-171.
12. Bayramlar, Hüseyin, Ibrahim F. Hepsen, and Harun Yilmaz. Mature cataracts increase risk of capsular complications in manual small-incision cataract surgery of pseudoexfoliative eyes. Canadian Journal of Ophthalmology/Journal Canadien d'Ophtalmologie. 2007; 42.1: 46-50.

13. Naseem, Abid, et al. Cataract surgery in patients with pseudoexfoliation. Pak J Ophthalmol. 2003; 23.3.