

## A Study of Interrelationship Between Occurrence of Cataract in Patients Attending Ophthalmology Out Patient Department in Burdwan Medical College & Hospital with Their Liver Function Profile And Serum Sialic Acid Levels

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**Objective:** Cataract is the leading cause of preventable blindness in India, the most common form of which is senile/ age related cataract. Various studies have suggested the probability of involvement of a complex athenogenic mechanism but exact etiopathogenesis is not established yet. This study is done for investigation of effects of various pathologies which cause liver dysfunction, or membrane damage or osmotic damage in body, in general, and their correlation with the development of senile cataract.

**Materials & Methods:**As a hospital based cross-sectional study the serum levels of bilirubin, alkaline phosphatase, gamma glutamyl transferase, sialic acid, sodium and potassium are measured in 50 patients of diagnosed cataract and 50 age and sex matched individuals with no cataract but some other diagnosed ocular diseases.

**Results:** Mann Whitney test done by SPSS Software has shown significant difference of serum bilirubin, alkaline phosphatase, gamma glutamyl transpeptidase and serum sialic acid levels between the case and control groups with cases having higher values ( $p= 0.000$ ). The difference in serum electrolyte levels between the two groups was also highly significant with cases having higher serum sodium level ( $p= 0.000$ ) and along with lower serum potassium level ( $p= 0.000$ ) in comparison to controls.

**Conclusion:** The study shows that development of senile cataract is multi-dimensional in etiology with liver dysfunction, electrolyte imbalance and increased sialic acid levels (indicative of long standing inflammation and/or increased oxidative stress) aiding in cataractogenesis.

### I. Introduction

Any opacity in the lens or its capsule, whether developmental or acquired, is called a cataract. Cataract is the leading cause of preventable blindness in India. According to development, cataract is of two main types, congenital and acquired. The most common form of cataract is the second one, i.e acquired cataract and is again subdivided into four major classes according to pathogenesis. These are –

- Age related cataract
- Cataract in systemic diseases, e.g- diabetes, myotonic dystrophy, atopic dermatitis, neurofibromatosis type-2 etc.
- Secondary cataract- develop as a result of some other primary ocular diseases, e.g- chronic anterior uveitis, angle closure glaucoma, high myopia, hereditary fundus dystrophies.
- Traumatic cataract.

Among all these, the most common form is senile/ age related cataract. For this type of cataract, various studies have suggested the probability of involvement of a complex pathogenic mechanism which include age, lifestyle, socio economic status etc, but exact etiopathogenesis is not established yet. Moreover, some studies have shown significant correlation between elevated levels of various serum markers and senile cataract.

Liver diseases, especially, in its subclinical form is a common entity in developing countries like ours. Liver function tests are useful in detection and diagnosis of liver diseases as well as liver dysfunctions and also for the monitoring of therapy, diagnosis of severity and evaluation of prognosis of certain diseases. The array of tests, useful for these purposes include the measurement of –

- Total and direct bilirubin
- Total protein
- Concentration of albumin

- Activity of certain enzymes such as-
- Aspartate amino transferase (AST)
- Alanine amino transferase (ALT)
- Alkaline phosphatase (ALP)
- Lactate dehydrogenase (LD)
- Gamma glutamyl transpeptidase (GGT)

These tests not only can diagnose liver dysfunction but also can categorise the disease. Among these liver function tests, some markers are chosen for the present study. These specific parameters are not chosen randomly but are chosen on the basis of previous studies in this field for further establishment of their correlation with cataract. The liver function markers , which are chosen for this study are –

- Total bilirubin
- Alkaline phosphatase (ALP)
- Gamma glutamyl transpeptidase (GGT).

Bilirubin is a yellow pigment. It has two forms, the unconjugated and conjugated form. The unconjugated bilirubin (designated indirect acting by nature of Vanden Bergh reaction )is a non polar and lipid soluble form and is an end product of heme protein catabolism from a series of enzymatic reactions by heme oxygenase and biliverdin reductase and nonenzymatic reducing agents in the reticuloendothelial cells. Some part of this unconjugated bilirubin that has undergone conjugation in the liver cell microsome by the enzyme uridine diphosphate glucuronic acid(UDP)-glucuronyl transferase form the polar, water soluble glucuronide of bilirubin (direct reacting). Although bilirubin may have a physiological role as an antioxidant.

Alkaline phosphatase (ALP) is an enzyme marker, the activity of which is present in most organs of the body and is especially, associated with membrane and cell surface of various organs. These organs associated with ALP activity are liver, bone, placenta, mucosa of small intestine, proximal convoluted tubule of kidney etc. However, elevation of serum ALP activity, commonly originate from the pathologies of liver and bone. Consequently, serum ALP measurement is of particular interest in the investigation of hepatobiliary diseases.

Elevated plasma levels of AST and ALT are common in many disorders and not very specific for liver dysfunction. On the other hand ALP activity may also be increased in bone diseases as well as liver diseases. So, to prove the association of elevated ALP with liver dysfunction, it is necessary to prove that the raised ALP is of hepatic origin. This can be done either by iso-enzyme fractionation of ALP or by side by side measurement of another canalicular enzyme GGT. The activity of GGT tends to parallel the activity of ALP in cholestasis.

Electrolyte concentration of plasma and serum are commonly analyzed in an electrolyte profile, because, these provide the most relevant information regarding osmotic, hydration and acid base status of the body. The normal electrolyte and water composition in plasma is as follows. –

Component	Amount in Plasma
Water	3.5 litre
Na <sup>+</sup>	142 meq/lit
K <sup>+</sup>	4 meq/lit
Ca <sup>2+</sup>	2.4 meq/lit
Mg <sup>2+</sup>	2 meq/lit
Trace elements	1 meq/lit
Total cations	155 meq/lit
Cl <sup>-</sup>	103 meq/lit
HCO <sub>3</sub> <sup>-</sup>	27 meq/lit
Protein <sup>-</sup>	16 meq/lit
Organic acids <sup>-</sup>	5 meq/lit
HPO <sub>4</sub> <sup>2-</sup>	2 meq/lit
SO <sub>4</sub> <sup>2-</sup>	1 meq/lit
Total Anions	154 meq/lit

Among the various blood components mentioned above, cataract is found to be associated with elevated levels of Na<sup>+</sup> and K<sup>+</sup> in some research papers.

Sialic acid is a generic term for a family of acetylated derivatives of neuraminic acid (mainly, n-acetyl neuraminic acid). Sialic acid, being an essential component of glycoprotein and glycolipids becomes an essential component of the cell membrane also. Thus, an elevated level of sialic acid in blood indicates cell membrane damages, especially, that of small vessels. So, quite naturally, elevated sialic acid level is found in damage of small end arteries, which in turn indicates the damage of supplying organs such as, eyes, kidneys and

brain.

This study is done for a further investigation of effects of various pathologies which cause liver dysfunction, or membrane damage or osmotic damage in body, in general, and their correlation with the development of senile cataract.

## **II. Materials & Method**

**Study area :** burdwan medical college & hospital.

**Study population:**

Cases- Diagnosed cataract patients attending the out -patient department in the Department of Ophthalmology of Burdwan Medical College & Hospital. Controls- Patients diagnosed with some ocular diseases other than cataract attending the out -patient department in the Department of Ophthalmology of BURDWAN MEDICAL college & Hospital.

**Group 1-:** Patients suffering from diagnosed cataract (cases).

**Group 2-:** Patients suffering from ocular disease other than cataract (controls).

**Inclusion and exclusion criteria:**

**Cases**The inclusion criteria are :

- The patients must have diagnosed cataract.
- Age above 45 years.

**The exclusion criteria are :**

- Presence of serious physical illness (as assessed through physical examination and routine laboratory screening).
- Presence of hypertension (using antihypertensive medication or BP >140/90 mm Hg at the time of presentation).
- Presence of diabetes (using anti diabetic drugs/ life style modification for diabetes or random blood sugar >140).
- The mere presence of frank liver disease.
- Presence of any ocular trauma.
- History of long term steroid use.

**Controls –**

**The inclusion criteria are :**

- The patients must have some ocular disease other than cataract.
- Age above 45 years.

**The exclusion criteria are :**

- Presence of diagnosed cataract.
- Presence of serious physical illness (as assessed through physical examination and routine laboratory screening).
- Presence of hypertension (using antihypertensive medication or BP >140/90mm Hg at the time of presentation).
- Presence of diabetes (using anti diabetic drugs/ life style modification for diabetes or random blood sugar >140).
- The mere presence of frank liver disease.
- Presence of any ocular trauma.
- History of long term steroid use.

**Sample size:**

Cases - 50

Controls - 50

**Study period:**

February 2014 to July 2015.

**Sample design:**

A hospital-based cross-sectional case control study on patients attending Ophthalmology OPD with

diagnosed cataract and without cataract but some other diagnosed ocular diseases.

**Parameters to be studied:**

- **Serum bilirubin:** By Mod. Jendrassik & Grof's method using kit and auto-analyzer.
- **Serum alkaline phosphatase (ALP):** By pNPP Kinetic method using kit and auto-analyzer.
- **Serum gamma glutamyl transpeptidase (GGT):** By standard method using kit and auto-analyzer.
- **Serum sialic acid:** By modified Melinoff's Method- a colorimetric method based on periodide oxidation→serum mixed with TBA→formation of hexose→Eventual formation of beta formyl pyruvic acid from free sialic acid→reacts with TBA→absorption maximum at 549 nm.
- **Serum sodium & potassium:** directly by ion selective electrode machine.
- **Plasma glucose:** By glucose oxidase-peroxidase method.

**Study tools:**

- Centrifuge machine REMI T8.
- Autoanalyzer XL-600.
- ISE machine.
- Semi auto analyzer.
- Reagents for glucose oxidase-peroxidase method.
- Bilirubin, ALP & GGT kit.
- TBA (Thio Barbituric acid).
- Routine laboratory glass wares and PVC wares.

**Plan For Analysis**

Data entry was done right after collection of relevant data (oral questionnaires and blood samples) for a given patient is complete. Data analysis has been done after completion of data collection from all patients. Data thus generated was analysed for any significant variation between the parameters among the study groups. Data were analyzed for any significant correlation between the parameters and the disease by appropriate statistical methods. SPSS software, for Windows used for data analysis.

**Ethical considerations:**

The study proposal along with other relevant documents would be submitted with the Institutional Review Board for review and approval. The study will commence after such approval is obtained in writing.

**Collection of sample:**

5 ml venous blood was collected from both the cases and the controls with proper aseptic technique. Blood was collected in container having no anticoagulant and also in container having citrate and fluoride for the measurement of plasma glucose. The collected clotted blood was then centrifuged at 1500 rpm speed for 3-5 minutes for separation of serum. All the tests were done with serum obtained from clotted blood except the test for estimation of plasma glucose which was done in citrated and fluoridated sample. The status of hypertension was determined by measurement of blood pressure using sphygmomanometer and oral questionnaire done for history of drug and alcohol intake.

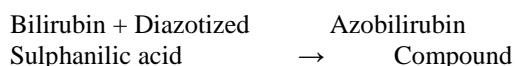
**Study technique:**

**Bilirubin Kit**

(Mod. Jendrassik & Grof's method For the determination of Direct & Total Bilirubin in serum.  
(For Invitro Diagnostic Use Only)

**Principle**

Bilirubin reacts with diazotised sulphanilic acid to form a coloured azobilirubin compound. The unconjugated bilirubin couples with the sulphanilic acid in the presence of a caffeine - benzoate accelerator. The intensity of the colour formed is directly proportional to the amount of bilirubin present in the sample.



**Normal reference values**

Serum (Direct) : upto 0.2 mg/dl  
(Total) : upto 1.0 mg/dl

It is recommended that each laboratory establish its own normal range representing its patient population.

<b>Contents</b>		30 tests	75 tests
<b>L1:</b> Direct Bilirubin Reagent		75 ml	150 ml
<b>L2:</b> Direct Nitrite Reagent	4 ml	4 ml	
<b>L1:</b> Total Bilirubin Reagent		75 ml	150 ml
<b>L2:</b> Total Nitrite Reagent	4 ml	4 ml	
<b>S:</b> Artificial Standard (10 mg/dl)	10 ml	10 ml	

### **Storage / Stability**

"All reagents are stable at Room Temperature, till the expiry mentioned on the label."

### **Reagent Preparation**

Reagents are ready to use. Do not pipette with mouth.

### **Sample material**

Serum. Bilirubin is reported to be stable in the sample for 4 days at 2-8°C protected from light as it is photosensitive.

### **Procedure**

Wavelength / filter : 546 nm / Yellow - Green  
Temperature : R.T.  
Light path : 1 cm

### **Total Bilirubin Assay**

"Pipette into clean dry test tubes labelled as Blank (B), and Test (T):"

Addition Sequence	B	T	
	(ml)	(ml)	
Total Bilirubin Reagent (L1)		1.0	1.0
Total Nitrite Reagent (L2)		--	0.05
Sample	0.1	0.1	

Mix well and incubate at R.T for 10 min. Measure the absorbance of the Test Samples (Abs.T) immediately against their respective Blanks.

### **Calculations**

Total or Direct Bilirubin in mg/dl = Abs.T x 13

### **Linearity**

"This procedure is linear upto 20 mg/dl. If values exceed this limit, dilute the sample with distilled water and repeat the assay. Calculate the value using the proper dilution factor."

### **Note**

In case the exact wavelength is not available the artificial standard (S) may be used. Measure the absorbance of the artificial standard against distilled water with the appropriate filter and keep the same for future calculations by dividing the Abs.T with the Abs. of the Std. x 10. Discard the Artificial Standard after use.

### **System Parameters**

Reaction: End Point	Interval: ---
Wavelength : 546 nm	Sample Vol.: 0.10 ml
Zero Setting : Sample Blank	Reagent Vol. : 1.05 ml
Incub. Temp : R.T.	Standard: ---
Incub. Time : 5 min/10 min	Factor : 13.00
Delay Time : ---	React.Slope: Increasing
Read Time : ---	Linearity: 20 mg/dl
No. of read : ---	Units : mg/dl

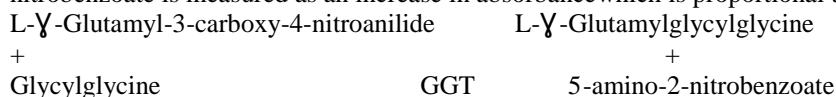
### **Alkaline Phosphatase Kit (Dea)**

(PNPP Kinetic Method)

For the determination of Alkaline phosphatase activity in serum.



GGT catalyzes the transfer of amino group between L- $\gamma$ -Glutamyl-3-carboxy-4-nitroanilide and Glycylglycine to form L- $\gamma$ -Glutamylglycylglycine and 5-amino-2-nitrobenzoate. The rate of formation of 5-amino-2-nitrobenzoate is measured as an increase in absorbance which is proportional to the GGT activity in the sample.



**Procedure:**

Wavelength / filter : 405 nm

Temperature : 37° C / 30 ° C / 25 ° C

Light path : 1 cm.

Pipette into clean dry test tube labeled as Test (T) :

<b>Addition</b>	<b>(T)</b>
<b>Sequence</b>	<b>(ml)</b>
Working reagent	1.0
Incubate at the assay temperature for 1 minute and add	
Sample	0.1

Mix well and read the initial absorbance A0 & repeat the absorbance reading after every 1, 2 & 3 minutes. Calculate the mean absorbance change per minute ( $\Delta A/\text{min.}$ ).

**Calculations:**

GGT Activity in U/L =  $\Delta A/\text{min.} \times 1158$

**Linearity:**

The procedure is linear upto 700 U/L at 37 °C. If the absorbance change ( $\Delta A/\text{min.}$ ) exceeds 0.250, use only the value of the first two minutes to calculate the result, or dilute the sample 1+9 with normal saline (NaCl 0.9%) and repeat the assay (Results  $\times 10$ ).

**Normal reference values:**

Serum (Males) : < 55 U/L at 37° C

(Females) : < 38 U/L at 37° C.

**Serum sodium & potassium:**

Serum sample, free from hemolysis is taken for estimation of serum sodium and potassium levels with the use of Ion Selective Electrode.

ISE fitted analyzers contain sodium electrodes with glass membrane and potassium electrodes with liquid ion exchange membranes that incorporates valinomycin. The principle of ISE is simple potentiometry, which determines the change in electromotive force (E, potential) in a circuit between a measurement electrode (the ISE) and a reference electrode, as the selected ion interacts with the membrane of ISE.

In application, the measuring system is calibrated by the introduction of calibrator solutions containing defined amounts of Na<sup>+</sup> and K<sup>+</sup>. The potential of the calibrator are determined and the  $\Delta E/\Delta \log$  concentration responses are stored in microprocessor memory as a comparison for calculating unknown concentration, when E of the unknown is measured. Frequent calibration, initiated by the user or by microprocessor-controlled uptake of sample from a reservoir of calibrator is a feature of most recent ISE systems.

Reference interval in adults:-

Serum sodium – 136 to 145 mmol/L.

Serum potassium- 3.5 to 5.1 mmol/L.

**Serum Sialic Acid Estimation:**

(Modified Aminoff's Method).

Colorimetric assay for sialic acid is based on periodate oxidation followed by reaction with TBA depends on formation of a hexose, 5-uronic acid product (which is a pre-chromogen) by the periodate cleavage of C<sub>6-7</sub>, C<sub>7-8</sub>, C<sub>8-9</sub> bonds in free sialic acid. There is eventual formation of  $\beta$ -formyl pyruvic acid from free sialic acid. This I turn reacts with TBA to yield a chromophore with absorption maximum at 549nm.

(Srinivasan & Sprinson 1959)

**Reagents and Materials**

Sample-2ml of clear serum free from hemolysis.

**Reagents-**

- TCA (5%)
- 0.1mol/L H<sub>2</sub>SO<sub>4</sub>
- Periodic acid (25mmol/L periodic acid in 0.125mol/L H<sub>2</sub>SO<sub>4</sub> p<sup>H</sup>- 1.2)
- Sodium Arsenite Solution (2% solution of Sodium Arsenite in 0.5 mol/L HCL)
- TBA (0.1 mol/L TBA solution in water, p<sup>H</sup>-9 adjusted with NaOH solution)
- Acid-Butanol mixture (butanol containing 5% volume/ volume in HCL)

**Instruments-**

- Centrifuge
- Water Bath
- Spectrophotometer (SYSTRONICS)

**Test Procedure-**

50 µl serum is (1:4) diluted with distilled water. 125µl of this sample is added with 5% TCA (for precipitation of serum protein) & the mixture is then centrifuged at 2000rpm for 5 minutes. Supernatant is discarded. Now sialic acid moieties hydrolyzed using 0.5ml of 0.1mol/L H<sub>2</sub>SO<sub>4</sub> at 100°C for 1 hour. 250 µl of periodic acid (25mmol/L periodic acid in 0.125mol/L H<sub>2</sub>SO<sub>4</sub> pH-1.2) is added to the above mixture & incubated in water bath at 37°C for 30 minutes.

Excess periodic acid is reduced by 0.2ml sodium arsenite solution (2% solution of Sodium Arsenite in 0.5 mol/L HCL). As soon as the yellow colour of liberated iodine started fading off, 2ml of TBA (0.1 mol/L TBA solution in water, p<sup>H</sup>-9 adjusted with NaOH solution) reagent is added drop wise with the help of a stopper. Now the mixture is incubated in water bath at 100°C for 8 minutes.

Now the tubes are cooled in ice-water & extracted with 5ml of Acid-Butanol mixture (butanol containing 5% volume/ volume in HCL)

Separation of 2 phases done by short rapid centrifugation & the color intensity of butanol phase is measured spectrophotometrically at 549nm using distilled water as blank.

**Calculation-**

Sialic acid is expressed in terms of µg/mg of serum protein.

Absorbance is converted to concentration by using Molar Extinction Coefficient of sialic acid- 70.7×10<sup>3</sup> L/mol cm.

Molecular weight of sialic acid is- 309.3.

**III. Resultanalysis**

For all six variables measured for both cases and controls, were subjected to Kolmogorov Smirnov test and the data showed skewness, i.e non parametric distribution.

**Table-1:**

Tests of Normality						
	Kolmogorov-Smirnov <sup>a</sup>			Shapiro-Wilk		
	Statistic	df	Sig.	Statistic	df	Sig.
BIL (T)	.060	50	.200*	.983	50	.687
BIL (C)	.109	50	.186	.945	50	.020
AP(T)	.100	50	.200*	.961	50	.102
AP©	.101	50	.200*	.984	50	.709
GGT(T)	.126	50	.045	.955	50	.055
GGT (C)	.104	50	.200*	.977	50	.417
NA+(T)	.122	50	.061	.850	50	.000
NA+(C)	.219	50	.000	.885	50	.000
K+(T)	.150	50	.007	.946	50	.023
K+(C)	.206	50	.000	.772	50	.000
S ACID(T)	.192	50	.000	.862	50	.000
S ACID(C)	.125	50	.049	.949	50	.032

a. Lilliefors Significance Correction  
 \*. This is a lower bound of the true significance.

So non parametric Mann-Whitney test were applied to compare data between case and control groups for each observed variable.



**Diagram – 1:**

**Diagram – 1:** showing difference in mean values between cases and controls in bilirubin, values expressed in mg/dl.

**Table-2: Mann-Whitney Test Rank for Bilirubin.**

	N	Mean (SD)	Mean Rank	Sum of Rank
Case	50	1.03 (0.39)	65.14	3257
Control	50	0.86 (0.27)	35.86	1793

**Table-3: Mann-Whitney Test Statistics for Bilirubin (Case/Control)**

Mann-Whitney U	518.00
Z	-5.047
Asym sig (2-tailed)	.000

**Diagram – 2:**

**Diagram – 3:**

**Diagram – 3:** showing difference in mean values between cases and controls in alkaline phosphatase, values expressed in IU/ml.

**Table-4: Mann-Whitney Test Rank for Alkaline Phosphatase.**

	N	Mean (SD)	Mean Rank	Sum of Rank
Case	50	138.78 (26.69)	67.30	3365
Control	50	106.60 (24.09)	33.70	1685

**Table-5: Mann-Whitney Test Statistics for Alkaline Phosphatase (Case/Control).**

Mann-Whitney U	410.000
Z	-5.79
Asym sig (2-tailed)	.000

**Diagram – 4:**

**Diagram – 5:**

**Diagram – 5:** showing difference in mean values between cases and controls in GGT ( $\gamma$ - Glutamyl Transpeptidase), values expressed in IU/ml.

**Table-6: Mann-Whitney Test Rank for GGT ( $\gamma$ - Glutamyl Transpeptidase).**

	N	Mean (SD)	Mean Rank	Sum of Rank
Case	50	49.95 (11.36)	70.06	3503
Control	50	30.43 (9.62)	30.94	1547

**Table-7: Mann-Whitney Test Statistics for GGT ( $\gamma$ - Glutamyl Transpeptidase) (Case/Control).**

Mann-Whitney U	272.000
Z	-6.74
Asym sig (2-tailed)	.000

**Diagram – 6:**

**Diagram – 7:**

**Diagram – 7:** showing difference in mean values between cases and controls in serum Sodium, values expressed in meq/L.

**Table-8: Mann-Whitney Test Rank for Sodium.**

	N	Mean (SD)	Mean Rank	Sum of Rank
Case	50	142.82(6.89)	65.74	3287
Control	50	134.56(9.22)	35.26	1763

**Table-9: Mann-Whitney Test Statistics for Sodium(Case/Control).**

Mann-Whitney U	488.000
Z	-5.26
Asym sig (2-tailed)	.000

**Diagram – 8:**

**Diagram – 9:**

**Diagram – 9:** showing difference in mean values between cases and controls in serum Potassium, values

expressed in meq/L.

**Table-10: Mann-Whitney Test Rank for Potassium.**

	N	Mean (SD)	Mean Rank	Sum of Rank
Case	50	3.46 (0.47)	29.42	1471
Control	50	4.78(0.62)	71.58	3579

**Table-11: Mann-Whitney Test Statistics for Potassium(Case/Control).**

Mann-Whitney U	196.000
Z	-7.28
Asym sig (2-tailed)	.000

**Diagram – 10:**

**Diagram – 11:**

**Diagram – 11:** showing difference in mean values between cases and controls in serum Sialic Acid, values expressed in µg%.

**Table-12: Mann-Whitney Test Rank for Sialic Acid.**

	N	Mean (SD)	Mean Rank	Sum of Rank
Case	50	3.27(0.39)	73.98	3699
Control	50	2.12 (0.23)	27.02	1351

**Table-13: Mann-Whitney Test Statistics for Sialic Acid(Case/Control).**

Mann-Whitney U	76.000
Z	-8.09
Asym sig (2-tailed)	.000

Diagram – 12:

#### IV. Discussion

Now it is well known that a number of risk factors are associated with the development of age related cataract. Significantly changed concentrations in a number of various plasma constituents have been reported to be associated with an increased relative risk of Cataract.

In the first Edinburgh based study, scientist have found significantly raised levels of bilirubin, alkaline phosphatase and GGT in cases as compared to that of controls.

In my study I have found a very significant increase in the levels of serum bilirubin, alkaline phosphatase and GGT in cataract patients as compared to controls. The rise of these levels mentioned above, in cataract group, suggest that subclinical liver dysfunction may be a causative etiology in the development of age related cataract. Moreover, significant rise of these three hepatic markers together indicate the presence of mild intra hepatic cholestasis in cataract patients. This cholestasis may be due to the presence of subclinical liver dysfunction or may be due to intake of drugs. However the exact cause of cholestasis cannot be delineated from the present data. But it is obvious that patients presenting with age related cataract bear high possibility of subclinical liver dysfunction, which may progress further to clinical one if not taken care of.

In many studies the concentrations of sodium, potassium and chloride have been studied in a group of carefully selected human senile cataractous lenses and in clear lenses from individuals of the same age group. In the course of senile cortical cataractogenesis, sodium accumulation within the lens represents a very early phenomenon, a decrease of potassium and is followed, at later stages of the disease, by an uptake of chloride.

The lens excretes sodium and concentrates potassium. Previous studies suggest the following conclusions regarding the metabolic pathways which provide the energy for such transport. (1) Under aerobic conditions in the absence of added metabolite, endogenous substrates supply sufficient energy to partially support transport. (2) The tricarboxylic acid cycle does not contribute. (3) Anaerobic glycolysis completely supports transport. (4) Of the enzyme inhibitors tested, the cation transport is most sensitive to iodoacetate and ouabain. (5) ATP is probably involved in cation transport. A possible role of Na-K ATPase has been suggested.

The major site of active transport is at the capsule. Study of exchange of <sup>22</sup>Na suggests a barrier or barriers within the lens which limit sodium exchange. These are influenced by steroids under conditions which may or may not alter the cation content of the lens.

Failure of active transport or increase in passive transport will eventually result in a shift in cations and hydration of the lens. The hydration can be accounted for by the increase in total base (sodium plus potassium) and attendant shift of anions. Localized accumulation of fluid in the lens is considered to reflect some imbalance of activities at the capsular and intralenticular barriers.

Studies have shown that changes in serum electrolytes levels can induce changes in aqueous electrolytes levels and effect on lens metabolism and probably cataract formation. The study conducted by M. Mirsamadi et.al showed Serum Na<sup>+</sup> level in senile cataract patients was higher than normal individuals. This

result might suggest that diets with high Na<sup>+</sup> content are a risk factor for age-related cataract formation, as high Na<sup>+</sup> content of the diet leads to high level of serum Na<sup>+</sup>, which in turn contributes to formation of age-related cataract. But this study did not find any significant change in serum potassium level in cataract patients.

In my study I have found a significant increase in the serum sodium level and a decrease in serum potassium level in cataract patients as compared to controls. This finding may be explained in the light that the altered serum electrolytes level may lead to alteration in the aqueous humor electrolyte level and thus may contribute to the formation of cataract. On the other hand it may be concluded that in patients presenting with age related cataract subclinical alteration of serum electrolytes level can be expected. Although, as the present study is a cross sectional study, it cannot provide definite evidence of long term electrolyte imbalance in cataract patients.

Sialic acid is basically released from terminal oligosaccharide chain of some glycoproteins and glycolipids of the acute phase. Increased level of sialic acid is favoured by the process desialylation, a process by which terminal Sialic acid has been removed by the action of sialidases from endogenous glycoprotein. Several studies showed that removal of terminal sialic acid moiety occurred due to increased oxidative stress. Thus inflammation and oxidative stress both plays pivotal role in increased serum sialic acid level.

It is already reported from the studies that presence of cataract is associated with the increase in serum sialic acid level.

It may be due to the cause that long standing inflammation leads to increased oxidative stress which in turn leads to formation of cataract. But as the level of serum sialic acid may be associated with increased oxidative stress alone, presence of long standing inflammation as a causative etiology of cataract formation cannot be established.

In my study I have found a very significant increase in serum sialic acid level in patients with cataract in comparison to age and sex matched control population. It may be due to presence of long standing inflammation due to various causes leading to increased oxidative stress, or, may be due to presence of oxidative damage alone. The exact cause cannot be assumed as this is a cross sectional study.

But the result of studies of all the parameters described, help us to reach in the conclusion that cataract is a manifestation of a number of subclinical systemic ailments, which need proper care in long term to avoid possible complications.

## **V. Conclusion**

The present study was started with the aim to find out, if there is any relationship between the occurrence of cataract and serum liver function, electrolytes and inflammatory marker sialic acid level individually. For this, blood was collected from patients suffering from cataract and non cataractous matched controls having other ocular diseases after fulfilling the inclusion and exclusion criteria. All analytes were measured after separation of serum and the data obtained were analyzed by SPSS software to find out the significance of differences between the cases and controls.

After result analysis following observations was obtained:-

- Significant difference in serum bilirubin level is present between the case and control groups with cases having higher serum bilirubin.
- Case group has significant high value of serum alkaline phosphatase in comparison to that of controls.
- Case group has significant high value serum gamma glutamyl transpeptidase in comparison to controls.
- Significant difference in serum electrolyte levels present between the case and control groups with cases having higher serum sodium level and along with lower serum potassium level in comparison to controls.
- Case population also possesses significant higher value of serum sialic acid levels in comparison to controls.

To conclude, the study supports the multi dimensional etiology of development of senile cataract. It is because that change in liver function markers and serum electrolytes are related to oxidative stress and enhance the process of lens aging. Sialic acid, being an inflammatory marker leads to the conclusion that, long standing inflammation also aids in cataractogenesis.

Although the multifactorial etiology of senile cataract was established by various studies done before, my study leads to the conclusion that otherwise healthy individuals presenting with cataract only, should undergo various other laboratory testing to check their liver function and serum electrolyte levels and the possible presence of long standing inflammation. Only extraction of cataract without these may in future lead to serious complications and morbidity in these patients who have only subclinical alteration in the mentioned serum markers at the time of presentation with cataract. But to establish the necessity of routine liver function tests and serum electrolyte levels in cataract patients and the cost effectiveness of this, a study involving much larger population and long term follow up is needed.

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#### **General Objectives-**

- [73]. To compare serum levels of liver function markers between cataract patients with that of control subjects.
- [74]. To compare serum electrolytes levels between cataract patients with that of control subjects.
- [75]. To compare serum levels of sialic acid between cataract patients with that of control subjects.
- [76]. To search for any correlation between the occurrence of cataract with the studied parameters.

#### **Specific Objectives-**

- [77]. To measure three liver function parameters namely serum total bilirubin, ALP and GGT level in patients and control subjects and to compare the values between two groups.
- [78]. To measure major serum electrolytes namely Na<sup>+</sup> and K<sup>+</sup> in patients and control subjects and to compare the values between the two groups.
- [79]. To measure serum sialic acid level as a cell membrane damage marker in patients and control subjects and to compare the values between the two groups.
- [80]. To search for any relation between elevated and or depressed levels of the mentioned parameters and occurrence of cataract. Thus to postulate the possible etiology behind the generation of senile cataract.