

# **CLINICAL AND BIOCHEMICAL STUDIES ON SENILE CATARACT**

Dissertation submitted to  
**THE TAMIL NADU DR. M.G.R. MEDICAL UNIVERSITY  
CHENNAI, INDIA**



**M.S. DEGREE EXAMINATION  
BRANCH – III OPHTHALMOLOGY**

**APRIL – 2014**

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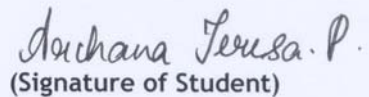
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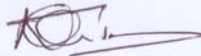


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## CERTIFICATE

This is to certify that the dissertation entitled "**CLINICAL AND BIOCHEMICAL STUDIES ON SENILE CATARACT**" is a Bonafide work done by **DR.ARCHANA TERESA P.** Postgraduate student in M.S. (Ophthalmology) during APRIL 2011 to MARCH 2014, under our direct supervision and guidance, at our Institute, in partial fulfillment for the award of M.S. Degree in Ophthalmology for the Tamilnadu Dr.M.G.R. Medical University, Chennai.

Dr.C.A.Nelson Jesudasan, M.S., D.O.M.S.,  
Professor and Director  
(Guide)

Dr. C.G.Vanila, M.S., D.N.B. (Ophthalmology)  
Professor of Ophthalmology  
Co-Guide

**Dr. C. A. NELSON JESUDASAN**  
Director, Joseph Eye Hospital  
Tiruchirappalli.

Dr.C.A.Nelson Jesudasan, M.S., D.O.M.S.,  
Professor and Director

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# **CLINICAL AND BIOCHEMICAL STUDIES ON SENILE CATARACT**

## **ABSTRACT**

### **AIM:**

To determine the frequency of occurrence of different types of senile cataract and assess the possible putative correlations with lenticular protein profiles and to compare visual outcomes and factors influencing them, in patients undergoing small incision cataract surgery (SICS) and phacoemulsification cataract surgery at a tertiary eye care hospital in India.

### **PATIENTS AND METHODS:**

520 patients-130 in the SICS group and 390 in the phaco group were enrolled in the study. The putative factors influencing the outcome of surgery, such as gender, the presurgical presence of systemic co-morbid conditions and of ocular conditions, and the degree of nuclear sclerosis of the cataractous lens, was ascertained. Differences in degree of visual improvement between the two surgical groups were sought. An attempt was made to study the human lens proteomics, by subjecting varying grades of nuclear cataract to sodium dodecyl polyacrylamide gel electrophoresis (SDS-PAGE) and 2DE analytical techniques, and the results obtained, were analysed.

**Results:**

Analysis of lenticular profiles revealed that NC3 was the most common cataract among males and in the SICS group, whereas NC4 was more common among females and in the phaco group. There was no difference in post-operative mean visual acuity between SICS and phaco in males, although such a difference existed in females with better visual acuity in the phaco group. Proteomic studies showed that the cataractous lens samples showed a statistically significant reduction in band intensity on SDS-PAGE and subsequently done two dimensional gel electrophoresis (2-DE) and MALDI-TOF analysis revealed varying levels of alpha-crystallins in the cataractous lens

**Conclusion:**

Both SICS and phacoemulsification yield satisfactorily similar visual outcomes and there are no other obvious factors such as grade of cataract, associated ocular and systemic comorbid conditions, that would favour choosing one procedure rather than the other. Additional biochemical studies on various grades of human cataract may help in identifying factors triggering cataractogenesis



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Master Chart

# *Introduction*

## INTRODUCTION

The eye is a highly specialized organ, which provides a view of the surrounding world by converting incident light into electrical signals that are interpreted by the brain. The human crystalline lens is meant to be transparent to enable it to focus visible light (400-700 nm) on the retina of the eye, providing increased sensitivity and allowing the information contained by that light to be spatially resolved.<sup>1,2</sup>

The term ‘cataract’ refers to a clouding or opaque area over the transparent ocular lens, which occurs when some of the proteins in the lens begin to aggregate, the end-result being impairment of vision<sup>3</sup>. Children may occasionally be born with the condition, or cataract may occur secondary to ocular injury or inflammation, to refractive errors such as myopia or to systemic diseases such as hypertension and diabetes mellitus. However, by far, the condition is mostly related to increasing age<sup>4</sup>. In fact, since cataract most commonly appears in elderly individuals, it is frequently referred to as “senile cataract”; if cataract develops before the age of 60, it may be called a “presenile” cataract.

It has been estimated that, globally, approximately 45 million people worldwide are blind, half of them due to cataract<sup>5,6,7</sup>. However, changing demographic structures of populations, particularly the increasing

proportion of the elderly, and enhanced life expectancy in the developing world, suggest that this number will increase dramatically in the coming years. In fact, projections by the World Health Organisation indicate that in 2020, close to 40 million people will be blind due to cataract<sup>8,9</sup>. In India, cataract has been reported to be responsible for 50-80% of bilateral blindness<sup>10,11,12</sup>. Thus, it is essential that all possible measures be taken to tackle this condition which may be responsible for a diminution in the quality of life.

At present, the most effective treatment of cataract is the surgical removal of the opacified lens; however, there continues to be a backlog of the services provided in many parts of the world. Globally, the two main techniques of cataract surgery are manual small incision cataract surgery and phacoemulsification; a third technique, microincision cataract surgery, is becoming increasingly popular<sup>13</sup>.

In phacoemulsification, first described in 1967 by Charles D. Kelman, there is ultrasonic fragmentation of the crystalline lens, and a small incision (with a standard size of around 2.75 mm); this technique permits rapid visual rehabilitation postoperatively and low- induced astigmatism. However, capital investment and recurring costs are high, and extensive surgical training is required. Manual small incision cataract surgery, first described by Blumental *et al.* (1992)<sup>14</sup>, is popular in Asia and

Africa since it is considerably less costly than phacoemulsification while still offering rapid visual recovery and reduced astigmatism. A 6 mm to 6.5 mm scleral incision allows the insertion of a 6 mm intraocular lens (IOL); the posterior capsule of the lens is left intact.

With increasing safety of ocular surgical techniques and improved visual results, it is no longer necessary to wait for the cataract to become sufficiently mature; in fact, surgery can be performed at a much earlier stage when phacoemulsification is used<sup>15</sup>. Cataract surgery may be performed depending on the patient's age and visual function demands<sup>16</sup>. Certain surgical complications can be avoided if the cataract is removed before it becomes too advanced<sup>17</sup>. Surgery should be considered when the benefits from removal of symptoms outweigh the small risks caused by modern surgery<sup>18</sup>.

With increasing volumes of cataract surgery in many parts of the world, the importance of well-performed reviews is being increasingly recognized. Publications have described outcome of cataract surgery, possible complications and improvements in final visual acuity<sup>19,20</sup>. However, additional information is needed with reference to factors that possibly influence outcomes, such as the gender of the patient, the presence of systemic co-morbid conditions, the type of surgery done and the degree of sclerosis of the cataract.

Two-dimensional gel electrophoresis (2-DE) and mass spectrometric (MS) analysis are the tools used in the emerging field of proteomics<sup>21</sup>; these hold great promise for determining putative modifications in lenticular structure that contribute to cataractogenesis. 2-DE is capable of simultaneously resolving complex mixtures of modified crystallins, which are then quantified by image analysis, and, subsequently, post-translational modifications on excised spots can be determined by MS. There are few studies on proteomic aspects of human lenses, and fewer studies on cataractous lens of varying degrees of nuclear sclerosis.

In the present study, an analysis was done of patients undergoing small incision cataract surgery (SICS) and phacoemulsification cataract surgery at a tertiary eye care hospital. Differences in degree of visual improvement between the two surgical groups were sought. In addition, putative factors influencing the outcome of surgery, such as gender, the presurgical presence of systemic co-morbid conditions and of ocular conditions, and the degree of nuclear sclerosis of the cataractous lens, were sought. In addition, cataractous lenses of varying grades of sclerosis were subjected to sodium dodecyl polyacrylamide gel electrophoresis (SDS-PAGE) and 2DE analytical techniques, and the results obtained were analysed.

# *Aim of the Study*

## **AIMS OF THE STUDY**

To compare outcomes in patients undergoing small incision cataract surgery (SICS) and outcomes in patients undergoing phacoemulsification cataract surgery at a tertiary eye care hospital in India.

### **Primary outcome measures**

1. To determine the frequency of occurrence of different types of senile cataract (based on the Lens Opacities Classification System [LOCS] Version III) in patients about to undergo cataract surgery.
2. To compare the proportions of individuals achieving good functional vision (presenting visual acuity better than or equal to 6/12 in the operated eye) following small incision cataract surgery with the proportions achieved in individuals undergoing phacoemulsification cataract surgery.
3. To identify putative factors influencing post-operative visual outcome, such as age, gender, types of senile cataract and presence of co-morbid systemic diseases (esp. diabetes mellitus & hypertension),



## **Secondary outcomes**

1. To note the occurrence of intraoperative complications such as posterior capsular rent and other complications as reported.
2. To note the occurrence of early post-operative complications.
3. To determine putative correlations between different types of senile cataract and lenticular protein profiles.

# *Review of Literature*

## REVIEW OF LITERATURE

The human lens is a cellular organ, its stability and transparency occurring as a result of tight packing of crystalline proteins into a glass-like microarchitecture. Senile (age-related cataracts), one of the commonest types of acquired cataracts, affects equally persons of either gender, usually above the age of 50 years. By the age of 70 years, more than 90% of individuals are likely to have developed a senile cataract. The condition is usually bilateral, but, for the most part, one eye is affected earlier than the other.

Anatomically speaking, the human lens can be divided into two principal regions, namely, the nucleus and the cortex. While the nucleus is present at birth, the cortex tends to be formed by differentiation of the lenticular epithelial cells throughout life<sup>22</sup>. Concomitant with the maturation of lenticular fibres, intracellular organelles, such as the mitochondria, tend to degenerate<sup>23</sup>. Thus, mature lenticular fibres tend to be devoid of cytoplasmic organelles, so that the bulk of the lens is made up of lipid membranes packed with lenticular proteins. This high concentration of structural proteins (lenticular crystallins) leads to an increase in the refractive index of the lens<sup>24</sup>. Any aberration in the structure or quantum of specific lenticular crystallins can lead to cataract<sup>25</sup>.

Senile cataractogenesis is essentially related to the ageing process. Although its precise etiology is unknown, various factors are implicated, such as heredity, ultraviolet radiation, dietary factors, childhood dehydrational crises and smoking<sup>22</sup>. These factors tend to influence the age of onset, type and maturation of senile cataract. Heredity plays an important role in the age of onset, incidence and maturation of senile cataract in different families<sup>26,27,28,29,30,31</sup>. Ultraviolet radiation has been implicated in the early onset and maturation of senile cataract, based on the observations made in several epidemiological studies<sup>32,33,34,35,36</sup>. Javitt and Taylor<sup>33</sup> found that the latitude of an individual's residence correlated directly with the UV-B content of sunlight, because the incident angle of the sun determines the atmospheric penetration of ultraviolet radiation. Interestingly, although a consistent dose-dependent association between ocular UV-B exposure and two common types of cataract (cortical and posterior subcapsular) has been shown by epidemiological studies, the same studies (unlike experimental studies) did not provide sufficient evidence to link UV-B exposure to senile macular degeneration in humans<sup>32</sup>.

A diet that is deficient in some proteins, vitamins (riboflavin, vitamins C and E) or essential elements has also been blamed for early onset and maturation of senile cataract<sup>37,38</sup>.

The incidence of three different cataract types tends to increase with age: nuclear cataracts, which account for approximately 60% of age-related cataracts; cortical cataracts which account for approximately 30% and posterior subcapsular cataracts, which account for the remaining 10%<sup>39</sup>. Klein *et al.*<sup>39</sup> described the cumulative incidence of age-related cataracts and cataract surgery over a 10-year interval through a prospective epidemiologic study on persons aged 43 to 86 years who were participating in the Beaver Dam Eye Study; there were 4926 individuals at baseline and 2764 individuals for first, five-year and ten-year follow-ups.

Photographs of the lens in each patient were taken using specially modified cameras and graded per codified rules by trained graders into nuclear, cortical, and posterior subcapsular cataracts. It was found that in right eyes, incident nuclear cataract occurred in 19.4%, cortical cataract in 17.4%, posterior subcapsular cataract in 6.1%, and cataract surgery in 10.8%. The incidence of all these end-points was found to be increased with age. Women had significantly higher incidences of nuclear cataract and cataract surgery than men. Time trends seemed to be influencing the

incidence of cataract surgery. These authors concluded that age-related cataracts are common events in aging, and that age-specific cataract surgery incidence seemed to be increasing.

With cataract being the leading cause of preventable blindness worldwide, cataract surgery is one of the most frequently performed operations in the world<sup>16</sup>. Hatch *et al.*<sup>40</sup> sought to estimate cataract surgery rates for the province of Ontario (Canada's largest province [state] with a population of about 13 million), by direct standardization of age- and sex-specific cataract surgery rates, obtained from the Ontario Health Insurance Plan Database, to Ontario's population projections; they found that the need for cataract surgery in this region was likely to more than double in the ensuing 25 years (the study was done in 2012).

Advancements in phacoemulsification and intraocular lens (IOL) technology have ushered in a new era of cataract surgery. Over the years, innovations in IOL design and phacoemulsification instrumentation have potentiated improved surgical outcomes, reduced perioperative morbidity and increased likelihood of spectacle independence<sup>16</sup>. A big step forward in improved surgical techniques of cataract surgery and improved outcomes was the gradual introduction of operating microscopes during the 1970s. These operating microscopes offered better intraocular visibility

and ability to safely place multiple corneal sutures. In addition, such microscopes provide the advantages of leaving the posterior capsule intact, leading to a reduced risk of potentially blinding complications, such as vitreous loss or retinal detachment, and also permitting the implantation of an IOL in the posterior chamber.

Currently, phacoemulsification is the standard of care for cataract extraction in the western world. The major advantage of phacoemulsification is that it has reduced the morbidity from cataract surgery by reducing the incision size with subsequent faster recovery and decreased risk of complications including endophthalmitis. In addition, the learning curve for this procedure seems to be remarkably small. Meeks *et al.* (2013)<sup>41</sup> reported on a retrospective cohort evaluation of the safety and efficacy of phacoemulsification cataract extraction and manual extracapsular cataract extraction performed by beginning resident surgeons in a medical centre in Dallas, Texas, USA. A review was performed of each resident's (post-graduate student) series of initial cataract surgery procedures as a late first-year or second-year resident. Data were collected for cases performed over almost a 6-year period during which initially the first primary surgeon cases were extracapsular cataract extraction and, later, the first primary surgeon cases were phacoemulsification.

Complications (vitreous loss or dropped nucleus) occurred in six (2.5%) of 244 cases in which phacoemulsification was performed by a beginner resident primary surgeon and in seven (4.1%) of 172 cases in which extracapsular cataract extraction was performed. Posterior chamber IOLs were placed in all but two phacoemulsification cases and four patients in whom extracapsular cataract extraction was performed. Moreover, three cases in the phacoemulsification group and one in the group that had undergone extracapsular cataract extraction required a reoperation within 90 days. These authors concluded that phacoemulsification cataract extraction can be taught safely and effectively to residents with no cataract surgery experience as a primary surgeon.

Mithal *et al.* (2012)<sup>42</sup> evaluated the outcome of manual SICS under topical anesthesia with lignocaine 2% jelly in a prospective interventional case series involving 128 patients with senile cataract; intra-cameral anesthesia was not used, and there was only a single operating surgeon. More than 55% of patients did not experience pain, while more than 90% had mild to no pain. Complications occurred in only two of the surgeries, while the surgeon reported a favourable experience with regard to the cooperation of the patients, stability of the anterior chamber, relative ease of surgery and relative absence of complications. Interestingly, a study in



2004<sup>43</sup> on 120 patients surveyed just before and one month after surgery found that there was a wide gap between expectations and what was actually achieved. While 60% of patients expected to achieve a perfect Visual Function Index score, and the average expected score was 96.1, the score actually achieved was 89.8; it was found that patients harboured unrealistic expectations regarding driving at night, reading small print, and doing fine handiwork. Surprisingly, improvement in visual function did not correlate with satisfaction in vision. The author of the study felt that the results highlighted the highly unrealistic expectations harboured by patients with cataract and underscored the need for physicians to attempt to control the expectations of their patients; the author even went so far as to say that controlling patient expectations was probably more effective than improving patients' postoperative outcome in terms of maximizing patient satisfaction<sup>43</sup>.

Several studies have tried to evaluate visual outcomes of SICS versus that in phacoemulsification Gogate *et al.* (2007)<sup>44</sup> sought to compare the cost of phacoemulsification cataract surgery, using foldable lenses, with that of manual SICS in a hospital setting in India through fixed-facility and recurrent (consumables) costing for both the surgeries based on information collected at different sources using standard norms.

Prior to this, a single masked randomized controlled clinical trial had been conducted to compare the safety and efficacy of the two techniques for rehabilitation of the cataract patient. The participants included 400 patients and four surgeons. The average cost of a phacoemulsification surgery for the hospital was Indian rupees (INR) 1978.89 while that of the SICS surgery was INR 720.99, with INR 500.99 being the fixed-facility cost common to both. The cost of phacoemulsification cataract surgery was higher due to the cost of the foldable lens used. Moreover, phacoemulsification required additional expenditure to cover the financial depreciation of the instrument, the replacement of faulty or old parts, and cost of the annual maintenance contract. These authors concluded that manual SICS is far more economical than phacoemulsification, safe and yields a visual result that is comparable with that of phacoemulsification.

In a prospective randomised study in Nepal, Ruit *et al.* (2007)<sup>45</sup> sought to compare the efficacy and visual results of phacoemulsification with that of manual sutureless SICS in 108 consecutive patients with visually significant cataracts. Both surgical techniques were found to achieve excellent surgical outcomes with low complication rates. On postoperative day 1, the groups had comparable uncorrected visual acuity (UCVA) while the SICS group had significantly less corneal edema. At six

months, 89% of the SICS patients had uncorrected visual acuity of 20/60 or better and 98% had a best-corrected visual acuity of 20/60 or better versus 85% of the patients in the phacoemulsification group with uncorrected visual acuity of 20/60 or better and 98% with best corrected visual acuity of BCVA of 20/60 or better; this difference was not statistically significant. Interestingly, the surgical time for SICS was found to be significantly shorter than that taken to perform phacoemulsification. These authors concluded that both phacoemulsification and SICS achieved excellent visual outcomes with low complication rates. However, they also opined that since SICS is significantly faster, less expensive, and less dependent on high-end technology than is phacoemulsification, SICS might be the more appropriate surgical procedure for the treatment of advanced cataracts in the developing world.

Singh *et al.* (2009)<sup>46</sup> sought to compare the safety and efficacy of different types of surgical procedures (phacoemulsification versus SICS) for cataract surgery in immature cataract.

In this prospective randomized controlled trial performed on 93 and 89 patients with immature senile cataract selected for phacoemulsification and SICS, respectively, there was no significant difference between the groups in terms of gender, age and pre-operative visual acuity. More than

two- thirds of patients in the phacoemulsification group and more than three quarters of the patients in the SICS group had good visual outcome (6/6-6/18) on the first postoperative day; this difference was not statistically significant. Poor outcome (<6/60) was recorded in 6% (phacoemulsification group) and 1% (small incision cataract surgery group). Mean visual acuity was 0.43 +/- 0.27 in the phacoemulsification group and 0.47 +/- 0.24 in the SICS group. Interestingly, the mean time for surgery was statistically significantly shorter in the SICS group than in the phacoemulsification group. These authors concluded that SICS cataract surgery with implantation of a rigid polymethylmethacrylate lens is a suitable surgical technique to treat immature cataract in developing countries.

Wong<sup>47</sup>, who examined rates of cataract extraction in Singapore, found that the average rate was highest for Indians (about 396.5 per 100, 000/year), followed by Chinese (371.2 per 100, 000/year) and Malays (only 237.2 per 100, 000/year); it was also found that women had higher rates of cataract extraction than men, and this pattern was seen across the three racial groups. This investigator was of the opinion that these racial variations suggested varying predisposition to cataract development and/or the threshold for cataract surgery. Gupta *et al.* (2013)<sup>48</sup>, who sought to

determine the prevalence of cataract surgery as well as factors in association with post-surgical outcomes of vision in migrant Indians living in Singapore, conducted a population-based study in 3, 400 Indian immigrants residing in Singapore. Post-operative visual impairment was defined as best-corrected or presenting visual acuity of 20/60 or worse. The age- and gender-standardized prevalence of cataract surgery was 9.7% in Singapore resident Indians, while the post-operative visual impairment, as defined by best corrected visual acuity, was found to occur in 10.9% of the eyes studied. The main causes of this impairment were found to be diabetic retinopathy, posterior capsular opacification, and age-related macular degeneration. These authors<sup>48</sup> concluded that socioeconomic variables and migration did not significantly contribute to the prevalence of cataract surgery, and that diabetic retinopathy was a major cause of postoperative visual impairment in migrant Indians living in Singapore. A study conducted in Fukui (Japan) echoed similar findings (Morikubo *et al.* 2004)<sup>49</sup>. No significant differences were observed in any preoperative parameter between diabetic and nondiabetic individuals. However, there was a significantly higher increase in corneal thickness one month post-surgery, and significantly greater losses of corneal endothelial cells at one day and one week after surgery, in diabetics than in non-diabetics.

Lam *et al.* (2007)<sup>50</sup> sought to evaluate the visual acuity and astigmatism of individuals undergoing manual cataract extraction by local surgeons in rural China over a 4 month period.

Among 313 eligible subjects, 242 (77%) could be contacted, of whom 176 (73%) were examined. Of those examined, mean  $\pm$  SD age was 69.3 $\pm$ 10.5 years, and 66.5% were female; 35 had been operated on bilaterally at Sanrao, and 85.2% had a preoperative presenting visual acuity of 6/60 or worse. Presenting and best-corrected postoperative acuity in the eye that was operated on were 6/18 or better in 83.4% and 95.7%, respectively.

Skiadaresi *et al.* (2012)<sup>51</sup> found poor correlations between cataract morphology and visual symptoms with the exception of the correlation found between blurred vision and posterior subcapsular cataracts.

Although excellent outcomes are frequently reported for cataract surgery in urban areas of China and India, the same cannot be said for rural areas of Asia, wherein poor visual outcomes, low visual function and quality of life and poor utilization of rehabilitation services after cataract surgery are often reported. To address this issue, Congdon and associates (2007)<sup>52</sup> sought to evaluate the postoperative visual function and utilization of refraction and second-eye surgical facilities available to individuals

undergoing cataract surgery in rural China. Interestingly, although visual function was high in this group of individuals and there was substantial potential benefit if refraction and second-eye surgery services were utilized, actual uptake of services was modest because individuals did not feel the need to do so. The authors concluded that programs to improve the utilization of services needed to focus on provision of reading glasses and on strategies to reduce costs<sup>52</sup>.

Lai *et al.* (2013)<sup>53</sup> investigated the clinical outcomes of cataract surgery in 207 individuals aged 90 and older who underwent cataract surgery for primary senile cataracts.

Interestingly, 79.7% of the 207 participants (mean age  $92.0 \pm 2.1$ ), 79.7% achieved visual improvement after cataract surgery in spite of a high incidence of systemic comorbidities such as hypertension (66.2%), diabetes mellitus (25.1%), and myocardial infarction (19.8%), as well as ocular comorbidities such as age-related macular degeneration (15.9%), glaucoma (10.6%), and myopic degeneration (5.3%). The most common complications were vitreous loss (8.2%), posterior capsular rupture (7.2%), and zonular rupture (4.8%). The authors concluded that despite a high prevalence of systemic and ocular comorbidities in very elderly adults, good clinical outcomes of cataract surgery were attainable<sup>53</sup>.

Crystallins are structural proteins which possess the ability to form soluble oligomers in high concentrations; specific, short range interactions are also possible, which increase refractive power of the lens and maintain transparency<sup>54</sup>. Crystallins are divided into three major classes, termed as alpha-, beta- and gamma-crystallins<sup>55</sup>.

Alpha crystallin, a member of the small heat shock protein family of molecular chaperones, is an aggregate of two polypeptides, namely, alpha-A and alpha-B crystallins, that share 55% amino acid sequence identity<sup>56</sup>. Alpha crystallin has chaperone-like properties, being capable of binding to unfolded or denatured proteins and suppressing non-specific aggregation<sup>57</sup>. The chaperone activity of alpha crystallin helps to prevent the formation of large light-scattering aggregates, inactivation of enzymes, and, possibly, cataract formation<sup>58,59</sup>. Thus, alpha A crystallin is of much importance in development of the lens and in the development and maintenance of lenticular transparency.

In the Beta crystallin superfamily, beta-crystallins have seven primary gene products; four of these are acidic, namely beta A1, beta A2, beta A3 and beta A4), while three are basic, namely, beta B1, beta B2, and beta B3<sup>55,60,61</sup>. An important function of beta B1 is in controlling the higher



assembly of beta crystallins and the potential role of truncated versions of the protein in cataract formation have also been reported<sup>62</sup>.

Cataract-specific forms of alpha-, betaB1 and B2-crystallins have been observed to lack from 5 to 22 residues in their termini. Truncated forms of alpha- and beta- crystalline were also observed to be more abundant in the water insoluble fraction, suggesting that their truncation may contribute to the formation of insoluble protein in cataractous lenses. Loss of 11 or more C-terminal residues in alpha-crystallin dramatically decreases its chaperone activity<sup>63</sup>.

Two-dimensional electrophoresis (2-DE) and mass spectrometric analysis (MS) are the tools in the emerging field of proteomics<sup>21</sup>; these hold a great promise in determining the modifications in lenticular crystalline proteins that contribute to cataractogenesis. 2-DE is capable of simultaneously resolving complex mixtures of modified crystallins. These resolved crystallins can be then quantified by image analysis, and then, subsequently, post- translational modifications on excised spots can be determined by MS.

The review of literature emphasizes the fact that there are still various aspects of cataract and cataract surgery that need to be studied. This underlines the relevance of the current study, in which an analysis has

been made of patients undergoing small incision cataract surgery (SICS) and phacoemulsification cataract surgery at a tertiary eye care hospital. Previous studies have not reported significant differences in final visual outcome between these two types of surgery, so, in the current study, differences in degree of visual improvement between the two surgical groups were sought. Different authors have reported on various factors that may possibly influence the outcome of therapy. Hence, in the present study, putative factors influencing the outcome of surgery, such as gender, the presurgical presence of systemic co-morbid conditions and of ocular conditions, and the degree of nuclear sclerosis of the cataractous lens, were sought. Increasingly, proteomic studies of cataractogenesis are being performed in a bid to unravel the mechanisms leading to the development of cataract. There have been many studies on experimental cataracts, but comparatively fewer on human cataracts. Hence, in the present study, human lenses with varying grades of nuclear cataract were subjected to sodium dodecyl polyacrylamide gel electrophoresis (SDS-PAGE) and 2DE analytical techniques, and the results obtained were analysed.

# *Materials & Methods*

## **MATERIALS AND METHODS**

Clinical aspects of the present dissertation were studied in patients presenting with diminished vision due to lenticular opacification at the Institute of Ophthalmology, Joseph Eye Hospital, Tiruchirapalli, Tamilnadu, over a period of 11 months (December 1, 2011 to October 31, 2012). Biochemical aspects were studied concurrently. Clinical and biochemical data were analysed and interpreted subsequently (March 2013 to September 2013). The study had the approval of the Institutional Review Board and conformed to the tenets of the Declaration of Helsinki ; written informed consent was also obtained from each patient.

### **CLINICAL ASPECTS**

Over an 11-month period, 604 patients with diminished vision due to cataract were seen. Of these, 22 individuals did not provide consent to participate while 30 individuals could not be enrolled due to the presence of one, or multiple, exclusion criteria. During data analysis, it was found that records of an additional 32 patients were incomplete. Hence, analysis could be performed for the clinical aspects of the study only on 520 patients.

**Study design:** This was a prospective, comparative interventional study done on patients presenting with various grades of cataract over an 11-

month period, the available interventions being small incision cataract surgery (SICS) or phacoemulsification cataract surgery. Provided that there was no specific indication to perform one particular intervention (SICS or phacoemulsification), the selection of the intervention was based on the patient's choice.

**Sample size calculation:** A key parameter to be investigated in the current study was the visual acuity following SICS or phacoemulsification cataract surgery. Based on published data, it was hypothesized that phacoemulsification surgery would yield a marginally better post-operative mean visual acuity than that achieved by SICS, and that this difference could be shown to be statistically significant by a Student 't' test. To calculate the minimum total sample size (two-tailed hypothesis) and minimum sample size per group (two-tailed hypothesis), the parameter values defined were:

- a) Anticipated effect size (Cohen's  $d$ ): 0.5
- b) Desired statistical power level ( $\beta$ -error): 0.8 (80%)
- c) Probability level ( $\alpha$  –error): 0.05

Calculation was done using an A-priori sample size calculator for Student 't' tests at the URL: <http://www.danielsoper.com/statcalc3/calc>, which yielded the following result:

*Minimum total sample size (two-tailed hypothesis): 128*

*Minimum sample size per group (two-tailed hypothesis): 64.*

In actual fact, a total of 520 patients were enrolled in the clinical study (130 in the SICS group; 390 in the phacoemulsification group)

**Inclusion Criteria:** Patients were considered for inclusion in the study if:

- they provided written informed consent, and were 40 to 70 years of age;
- exhibited normal anterior segment morphology (other than for the presence of senile cataract in one or both eyes);
- showed normal intraocular pressure (IOP) or senile cataract with primary open angle glaucoma under medical management (with or without control of IOP), or intumescent lens with narrow angles on gonioscopy (without any peripheral anterior synechiae);
- exhibited a normal fundus or ultrasound B-scan evidence of a normal posterior segment;
- exhibited patent naso-lacrimal ducts on syringing; or
- had systemic hypertension, diabetes mellitus or hypercholesterolemia that was under control with medical management.

**Exclusion criteria:** Patients were not considered for the current study if they exhibited **any one** of the following exclusion criteria:

- declined to undergo surgery;
- were less than 45 years of age;
- suffered from uncorrected high refractive errors or amblyopia ;
- had a conjunctival swab report showing significant growth of potentially pathogenic bacteria;
- had nasolacrimal duct obstruction due to untreated acute or chronic nasolacrimal duct infection;
- suffered from nystagmus or congenital anomalies of the eye;
- suffered from external ophthalmoplegia with or without multiple cranial nerve palsies;
- suffered from corneal opacities (nebula, leucoma), dystrophies and degenerations;
- exhibited active allergic, infective or inflammatory conditions such as episcleritis, scleritis, conjunctivitis, keratitis, anterior, intermediate or posterior uveitis, optic or retrobulbar neuritis or infective foci anywhere in the body;
- suffered from traumatic cataract, complicated cataract or polar cataract;
- suffered from phacolytic glaucoma;

- suffered from intraocular, intraorbital or systemic malignancies;
- had a B-scan showing posterior segment abnormalities other than normal age-related changes;
- suffered from uncontrolled systemic hypertension, diabetes mellitus, ischemic heart disease, chronic kidney disease, asthma or tuberculosis were receiving topical or systemic steroids or immunosuppressive agents reported a recent history of a cardiovascular or cerebrovascular accident.

**Clinical examination:** All patients were subjected to visual acuity measurement objectively by auto-refractometer and subjectively by Snellen chart and the best corrected visual acuity (BCVA) was recorded after Snellen to decimal conversion.

The patient's head position and eye position were assessed before starting the ocular examination. Extraocular movements were recorded in all nine cardinal positions of gaze. This was followed by examination of each eye separately.

The appearance of the upper and lower lid margins, the position of both the lids and the interpalpebral fissure height and the lid crease were noted. This was followed by a careful examination of the bulbar and palpebral conjunctiva and fornices was done.



Slit-lamp biomicroscopic examination was done for all patients. The status of all the corneal layers, namely the epithelium, stroma and endothelium, was noted. The presence of guttae, keratic precipitates, pseudoexfoliative material and pigments on the corneal endothelium was recorded.

The central and peripheral anterior chamber depth was assessed using Shaffers system of grading of anterior chamber depth. The presence of flare or cells was also looked for in the anterior chamber.

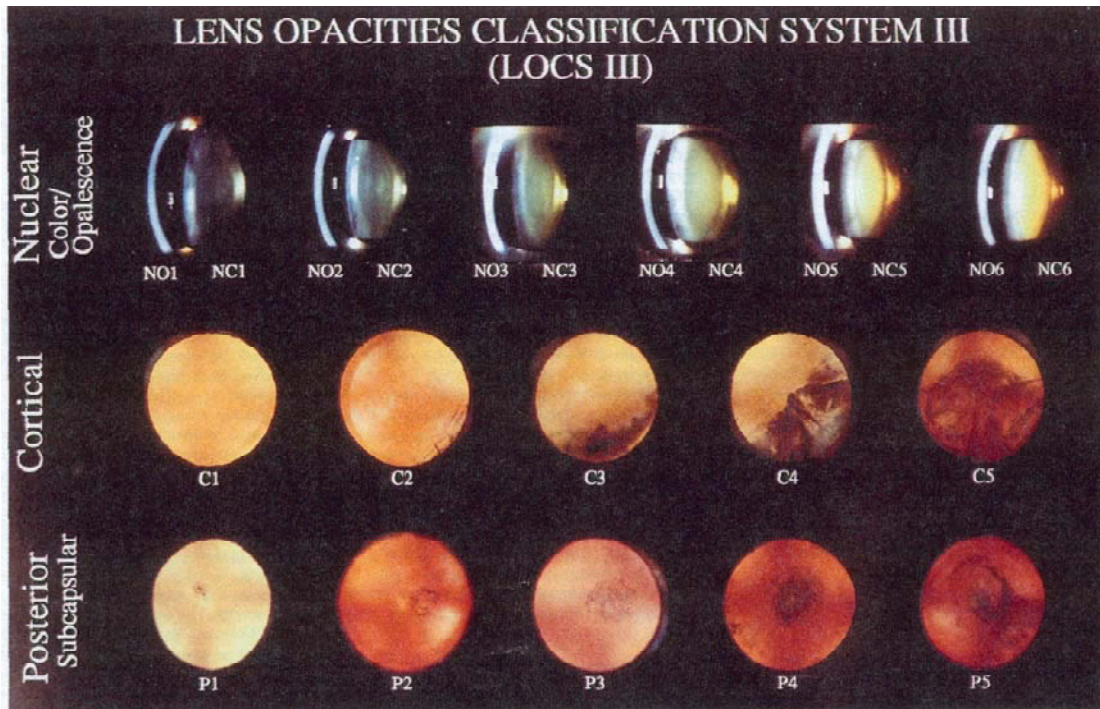
The colour and pattern of the iris, and the presence of peripheral anterior synechiae, iris atrophic patches, or pre-existing surgical or non-surgical peripheral iridotomy wounds were noted. The pupillary size, shape, regularity, presence of synechiae, reaction to direct and consensual light reflex and presence of pseudoexfoliative material along the papillary ruff were also noted.

In cases of mature cataract, the macular function was assessed by the presence of a normal pupillary reaction and the absence of any relative afferent pupillary defect (RAPD).

**Assessment of lenticular status:** The lenticular status was assessed using the Lens Opacification Classification System (LOCS III) (Chylack *et al.*,

1993)<sup>64</sup>. The LOCS III contains an expanded set of standards (Fig. 1) selected from the Longitudinal Study of Cataract slide library at the Center for Clinical Cataract Research (Boston, MA, United States of America). It consists of six slit lamp images for grading nuclear colour (NC) and five retroillumination images for grading posterior subcapsular cataract (PSCC) (Fig. 1).

Nuclear cataract was graded by comparing the colour of the lens to be graded with the standard colour photographs defined by the LOCS III as NC grades 1 to 6. While grading nuclear colour, two regions of the nucleus, namely, the entire cross-sectional view of the nucleus and the posterior subcapsular reflex, were viewed.



**Figure 1.** Slit- lamp images for grading nuclear colour and retroillumination images for grading posterior subcapsular cataract in the Lens Opacities Classification System III. From: Chylack LT Jr. *et al.* Lens Opacities Classification System III [LOCS III]. *Arch Ophthalmol* 1993; 111: 831-836. (Reproduced with permission from the American Medical Association, License No. 3286581148272 dated December 12, 2013).

This grading is different from that followed in LOCS II in that the colour of the entire nucleus is considered, in addition to the posterior reflex. This method thus avoids overestimation of the brunescence stage, which occurs occasionally when the reflex alone is used.

Posterior subcapsular cataracts were graded by using only posteriorly focused retro-illumination images. The area of the opacity in the lens being graded was compared with the standards 1 to 5. The posterior segment status was assessed using the slit-lamp and a 90D lens. In patients in whom there was no view of the posterior segment, ultrasound B-Scan was done to exclude any underlying ocular pathology.

**Pre-operative workup:** The patency of the nasolacrimal duct was assessed.

Keratometry was performed on both eyes manually with a Super KMS-6™ keratometer, (Bausch and Lomb, USA) and with an automated keratometer (KM-500 NIDEK™) Autokeratometer, and the average keratometry reading was then calculated.

Intraocular lens (IOL) power calculation was done by a non-contact method using the IOL Master 500 (Carl Zeiss Medite AG); in patients with more dense cataracts that totally obscured media clarity, a contact method,

using the immersion technique using the Ocuscan <sup>TM</sup> P/N 683-3001-505 (Alcon Surgicals, USA) was resorted to. The *SRK-II formula (A-2.5L-0.9K)* was used to calculate IOL power, taking into account the axial length (L) and average keratometry (K) reading. 'A' constant was calculated as provided by the manufacturer.

**Pre-operative preparations:** On the day of surgery, a thorough preoperative assessment of the patient was performed. This was followed by pre-operative preparation of the patient. The patients were instructed to wash their face with soap and water. Appropriate head and foot gear were provided before entry into the operation theatre complex. The skin around the eye was cleaned with povidone-iodine 5%. Following this, peribulbar block was given to the eye to be operated. This was again followed by cleaning of the conjunctival cul de sac and the skin around the eye with povidone iodine.

### **Operative techniques**

Surgery (both SICS and phacoemulsification) was performed under the *magnification* of an operating microscope (Leica Microsystem<sup>TM</sup> Model M220F12, Singapore; Carl Zeiss S88 OPMI Lumere T <sup>TM</sup> Model 302608-9020-000, Carl Zeiss Surgical GmbH, Oberkochen, Germany).

In patients undergoing **SICS**<sup>65</sup>, following conjunctival peritomy, bleeders were cauterized. The expected size and density of the nucleus determined the size of the tunnel, with the extraction of earlier stages of nuclear cataracts requiring only a small tunnel (sufficient for the IOL optic to pass through) while very big, brown nuclei required a larger tunnel size. A 3 plane sclerocorneal tunnel incision extending at least 1 to 2 mm into the clear cornea was made. Following continuous curvilinear capsulorhexis, hydrodissection was performed, followed by use of a vectis or viscoexpression. A 360 degree cortex aspiration was done using a Simcoe cannula, followed by placement of the posterior chamber IOL.

In the **phacoemulsification** technique<sup>66,67,68</sup>, the same pre-operative protocol for patient preparation was followed. After conjunctival peritomy, a 5 mm corneo-scleral tunnel incision was made. Continuous curvilinear capsulorrhexis not exceeding 5mm was also performed. The phaco probe (an ultrasonic handpiece with a titanium or steel needle) was used (Millenium™ Ref CX6200 S721305, Bausch and Lomb Inc., Rochester, NY, USA; Alcon Infiniti Ozil™. Vision System, Alcon Laboratories, USA; Megatron Gender AG™, Heidelberg, Germany). The tip of the needle vibrated at an ultrasonic frequency to sculpt and emulsify the cataract while the pump aspirated particles through the tip. A second fine

steel instrument, called a "chopper", was used from a side port to help with chopping the nucleus into smaller pieces. Either a 'stop and chop' technique or a 'divide and conquer' technique was followed to break the nucleus. Thus, the cataract was broken into two or four pieces and each piece was emulsified and aspirated out with suction. After removing all hard central lenticular nuclear pieces by phacoemulsification, the softer outer lens cortex was removed by suction only. An irrigation-aspiration probe or a bimanual system was used to aspirate out the remaining peripheral cortical matter, while leaving the posterior capsule intact.

An IOL was placed in the capsular bag. For implanting a polymethylmethacrylate IOL, the incision had to be enlarged while this was not necessary to implant a foldable IOL.

All patients were examined the day after surgery as a routine post-operative day one follow-up. The status of the cornea was noted and presence of striate keratopathy, Descemet's membrane folds, Descemet's membrane stripping, and epithelial or stromal edema was also noted. The anterior chamber was examined for presence of excessive flare or cells, hyphema, exudates and hypopyon. The central and peripheral anterior chamber depth was also noted. The iris was examined for any surgical iridectomy and iris atrophy. The positioning of the posterior chamber IOL,

including centration, was noted. The fundus was examined for vitreous clarity and presence of any underlying posterior segment pathology. IOP was recorded by non contact tonometry.

The patients were started on topical corticosteroids for hourly use and were advised to come for followup after one week. At the follow-up visit, the patient's uncorrected visual acuity was measured. The anterior segment was evaluated for presence of corneal edema, flare and cells. The posterior chamber IOL position and centration were noted.

## **BIOCHEMICAL STUDIES**

These were performed on representative cataractous lenses from patients undergoing cataract surgery, and on transparent lenses obtained during eye donation.

**Sodium-Dodecyl-Sulfate Polyacrylamide Gel Electrophoresis (SDS-PAGE) analysis of total lenticular protein (soluble and insoluble fractions) pattern :** The total soluble and insoluble protein pattern was analyzed by subjecting the sample to 4-20% gradient SDS-PAGE following the method of Laemmli (1970)<sup>69</sup>. The gradient was prepared using 60% sucrose along with 30% acrylamide, Tris buffer, ammonium persulphate (APS) and N, N, N', N'-tetramethylethylenediamine (TEMED). Two different percentages (4% and 20%) of separating gel solutions were



constituted and the gradient was prepared by mixing these two solutions using a gradient mixer. The mixed solution was poured into a sealed glass plate setup (8 cm high and 1.5 mm thick) for polymerization. After the polymerization was complete, the 3% stacking gel was poured over the separating gel and a Teflon comb was inserted to form wells. Later, the Teflon comb was removed and the wells were rinsed with distilled water. The basal strip was then removed and the glass plate with polymerized gel was fixed to the electrophoretic apparatus. When the setup was ready, the samples were mixed with equal amounts of gel-loading dye and loaded into the well. Electrophoretic separation was allowed for 180 minutes at a constant voltage of 50V for the stacking gel and 100V for the separating gel. The electrode solution used was 25 mM Tris, 0.193 M glycine buffer containing 0.1% SDS. Broad range protein markers were simultaneously run for comparison. The gel was stained using Coomassie Brilliant Blue R-250 and destained with 7% acetic acid and 40% ethanol. The bands that developed, after staining and destaining, were scanned in a gel documentation system (Bio-Rad, USA) to determine the intensity of the bands. The concentration of proteins in each band was determined from the standard graph plotted against the intensities of the bands corresponding to the known standard marker proteins. The program Quantity One SW (Bio-Rad, USA) was used for the analysis of intensity of bands in each lane of

the gels. Density profiles of each lane of the gels were used to calculate the peak intensity of the selected protein bands corresponding to soluble and insoluble proteins.

**Two dimensional (2D) gel electrophoresis analysis of the pattern of total lenticular protein (soluble and insoluble fractions):** Four lenses from each group were homogenized in 200 mL lysis solution containing protease inhibitors, followed by centrifugation, as described earlier. The supernatant containing the soluble protein was removed, and the pellet (insoluble protein) was washed twice with homogenizing buffer. The insoluble protein was then resuspended in 8M Urea, and the protein content in both the soluble and insoluble fractions were measured by the

Bradford method (1976)<sup>70</sup>, using bovine serum albumin as a standard. Both fractions of lens proteins were aliquoted into 400-mg portions and stored at -70°C.

Immobilized pH gradient (IPG) strips (linear pH 5-8, 7 cm) (Bio-Rad, USA) were rehydrated overnight with 160 µg of total protein derived from individual subjects as mentioned above and premixed with a rehydration buffer. The first-dimensional separation was performed

in Protean IEF Cell (Bio-Rad, USA) at 20°C, using stepwise mode to reach 10,000 Vh. After completion of the isoelectric focusing, the separated proteins were equilibrated with another buffer for a further 15 min. The equilibrated IPG strips were then transferred onto 12% acrylamide slab gels (8 × 9.5 cm) and the second-dimensional separation was performed in Mini-Protean Tetra Cell (Bio-Rad, USA) with the current of 200 v/gel for approximately 40 min. The resolved protein spots were then visualized using Coomassie Brilliant Blue R-250 stain.

PD Quest (Bio-Rad), was used for matching and analysis of protein spots on 2-D gels. A reference gel was created by combining into one image all of the spots that had appeared in the individual gels. The reference gel was then used for matching of corresponding protein spots among different gels. Background subtraction was performed and the intensity volume of each spot was normalized with the total intensity volume (summation of the intensity volumes obtained from all spots within the same 2-D gel). Protein spots of particular interest, based on pI and MW, were excised with a clean scalpel, and processed according to the method described by Courchesne and Patterson (1999)<sup>71</sup>. 85 Peptides were directly spotted into the matrix-assisted laser desorption/ionization (MALDI) plate with 10 mg/mL  $\alpha$ -cyano-4-hydroxycinnamic acid 50%

acetonitrile and 0.1% (v/v) TFA, and the spots were allowed to dry completely. Mass spectra of positively-charged ions were recorded on an Ultraflex MALDI TOF mass spectrometer (Bruker Daltonik) under the control of FlexControl™ 2.2 software (Bruker Daltonik GmbH).

### **Statistical Analysis**

SPSS™ statistical software version 16 (Statistical Package for the Social Sciences for MS Windows, SPSS Inc., Chicago, IL, USA) and Microsoft 2007 Office Excel™ software were used for statistical calculations. Differences in age, mean visual acuity and other continuous variables were calculated by independent sample Student 't' tests. Yates's corrected chi-square tests were used to analyse differences between categorical variables, using two by two tables, when appropriate. All statistical tests were two-sided (two-tailed), and  $P \leq 0.05$  was considered to be statistically significant.

# *Results*

## **RESULTS**

Clinical aspects of the present dissertation were studied in patients presenting with diminished vision due to lenticular opacification at the Institute of Ophthalmology, Joseph Eye Hospital, Tiruchirapalli, Tamilnadu, over a period of 11 months (December 1, 2011 to October 31, 2012). Biochemical aspects were studied concurrently. Clinical and biochemical data were analysed and interpreted subsequently (March 2013 to September 2013).

### **1. CLINICAL ASPECTS**

Over an 11-month period, 604 patients with diminished vision due to cataract were seen. Of these, 22 individuals did not provide consent to participate while 30 individuals could not be enrolled due to the presence of one, or multiple, exclusion criteria. During data analysis, it was found that records of an additional 32 patients were incomplete. Hence, analysis could be performed for the clinical aspects of the study only on 520 patients.

Using an A-priori sample size calculator for Student 't' tests, the **minimum total sample size** (two-tailed hypothesis) was calculated to be 128 while the **minimum sample size per group** (two-tailed hypothesis) was calculated to be 64 (details in Materials and Methods section). In actual fact, a total of 520 patients were enrolled in the clinical study (130 in the SICS group; 390 in the phacoemulsification group). Hence, the requirements for sample size were met in the present study.

Patients who were enrolled in the study underwent monocular cataract surgery either by small incision cataract surgery (SICS) or by phacoemulsification, as described earlier (Materials and methods). Patients who underwent SICS constituted the "SICS group" while those who underwent phacoemulsification constituted the "Phaco group". There were 130 patients in the SICS group and 390 patients in the Phaco group (Table 1).

**Table 1 : Age and gender of patients undergoing small incision cataract surgery or phacoemulsification cataract surgery at a tertiary eye care hospital**

Gender of patient	Patients undergoing small incision cataract surgery (SICS group)		Patients undergoing phacoemulsification cataract surgery (Phaco group)		Overall Total
	Number	Mean age $\pm$ SD (years)	Number	Mean age $\pm$ SD (years)	
Males	39	60.95 $\pm$ 4.44	122	61.66 $\pm$ 4.61	61
Females	91	61.97 $\pm$ 4.82	268	61.56 $\pm$ 4.33	359
Group total	130		390		520

Abbreviation: SD= standard deviation d.f.= degree of freedom

*Statistical Analysis*

a. Chi-square test (2X2 table)

b, c, d. & e. Student 't' test

a. Gender distribution in SICS group versus Phaco group

$$\chi^2 (d. f. = 1) = 0.048 ; P > 0.05$$

b. Mean age of males in SICS group versus mean age of males in Phaco group

$$(unpaired) \text{ 't' } (d. f. = 159) = 0.08 ; P= 0.93$$

c. Mean age of females in SICS group versus mean age of females in Phaco group

$$(unpaired) \text{ 't' } (d. f. = 357) = 0.05 ; P= 0.96$$

d. Mean age of males versus mean age of females in SICS group

$$(unpaired) \text{ 't' } (d. f. = 128) = 0.13 ; P= 0.9$$

e. Mean age of males versus mean age of females in Phaco group

$$(unpaired) \text{ 't' } (d. f. = 388) = 0.01 ; P= 0.99$$



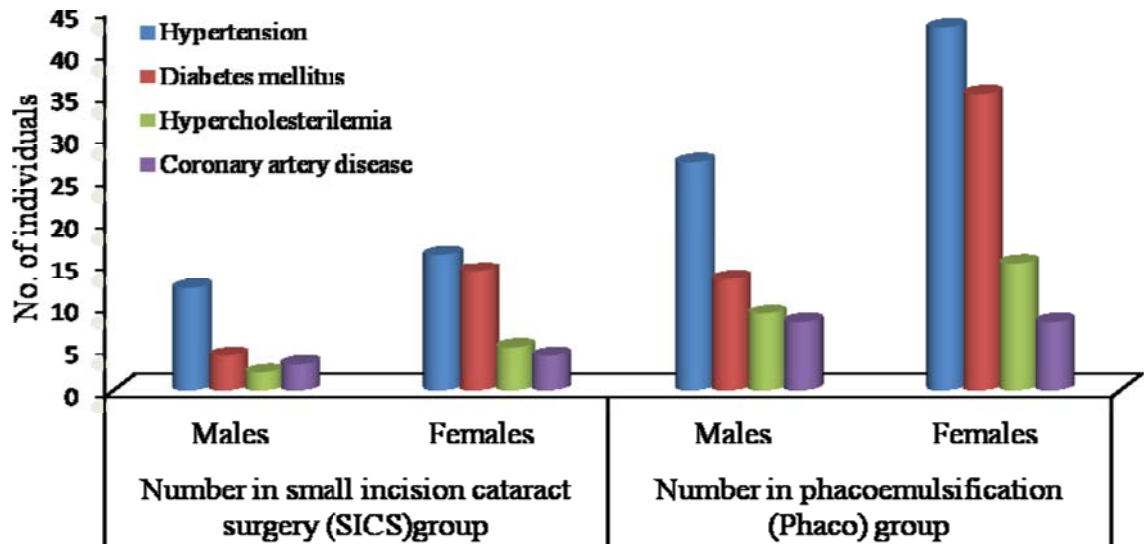
## **1.1 Age and gender distribution**

There were 39 males and 91 females in the SICS group and 122 males and 268 females in the Phaco group (Table 1); these differences were not statistically significant ( $\chi^2$  (degree of freedom [d.f]=1) = 0.05 ;  $p > 0.05$ ). The mean ages of males and females were 60.95 + 4.44 years and 61.97 + 4.82 years, respectively, in the SICS group and 61.66 + 4.61 years and 61.56 + 4.33 years, respectively, in the Phaco group (Table 1); these differences were not statistically significant (SICS group males versus females,  $t' = 0.13$  [d.f. = 128];  $p = 0.9$  ; Phaco group males versus females,  $t' = 0.01$  [d.f.=388];  $p=0.99$ ; males in SICS versus Phaco groups,  $t' = 0.08$  [d.f.=159],  $p=0.93$ ; females in SICS versus phaco groups,  $t' = 0.05$  [d.f.= 357]  $p = 0.96$ ). Thus, patients in the SICS group and phaco group were, essentially, age-and gender-matched.

## **1.2 Co-morbid systemic conditions**

Co-morbid systemic conditions noted in the patients in the two groups were hypertension, diabetes mellitus, hypercholesterolemia and coronary artery disease (Figure 2).

**Figure 2 : Presence of systemic co-morbid conditions in patients undergoing small incision cataract surgery or phacoemulsification cataract surgery at a tertiary eye care hospital**



*Abbreviations:* d.f.= degree of freedom

### *Statistical Analysis*

- Proportions of patients with systemic co-morbid conditions in SICS group ( $60/130 = 46\%$ ) versus (vs.) Phaco group ( $158/390 = 41\%$ ) :  $\chi^2$  (d. f. = 1) = 1.04 ;  $P > 0.05$
- Proportion of males (78[48%] of total 161 males) vs. females (140[39 %] of total 359 females) with systemic co-morbid conditions:  $\chi^2$  (d. f. = 1) = 4.47;  $P = 0.03$
- Proportions of males (39 [24%] of total 161 males) vs. females (59 [16.4%] of total 359 females) with hypertension:  $\chi^2$  (d. f. = 1) = 4.4;  $P=0.036$
- Proportions of males (17 [10.6%] of total 161 males) vs. females (49 [13.6%] of total 359 females) with diabetes mellitus:  $\chi^2$  (d. f. = 1) = 0.96;  $P=0.33$

### **1.2.1 In SICS group versus Phacoemulsification group**

Overall, co-morbid conditions occurred in 60 (46%) of 130 patients in the SICS group and in 158 (40.5%) of 390 patients in the Phaco group (Figure 2); this difference was not statistically significant ( $\chi^2$  [d.f.=1]=1.04;  $p > 0.05$ ). Hypertension was, by far, the most frequent co-morbid condition in males in both groups, affecting 57% in the SICS group and 47% in the phaco group (Figure 2).

### **1.2.2. In Males versus Females**

Interestingly, co-morbid conditions occurred significantly more frequently in males (78 [48.4 %] of 161) than in females (140 [39%] of 359) studied ( $\chi^2$  [d.f.=1] = 4.47 ;  $P = 0.03$ ) (Figure 2). Hypertension and diabetes mellitus were the most frequent co-morbid conditions in females in both groups, affecting 41% and 36%, respectively, in the SICs group, and 43% and 35%, respectively, in the Phaco group (Figure 2). Interestingly, hypertension was seen significantly more frequently in males (39 [24%] of 161 males) than in females (59 [16.4%] of 359 females) in the current study ( $\chi^2$  [d.f.=1] = 4.4 ;  $P=0.04$ ). Although diabetes mellitus occurred more frequently in females (49 [13.6%] of 359) than in males (17 [10.6%] of 161), this difference was not statistically significant ( $\chi^2$  [d.f.=1] = 0.96;  $P=0.33$ ).

### **1.3 Types of cataracts**

Various grades of nuclear (Figures 3, 4 and 5), posterior subcapsular (Figures 7, 8 and 9) and end-stage cortical (mature and hypermature) cataracts (Figures 10 and 11) were seen in the patients. These cataracts were graded according to the LOCS classification III (Fig.1 in Materials and Methods section); these details are listed in Table 2 and Figures 6, 12 and 13.

#### **1.3.1 In SICS group versus Phacoemulsification group**

In the SICS group (total 130 patients), three patients (2%) had NC2, 52 (40%) had NC3, 38 (29%) had NC4, eight (6%) had NC5 and 29 (23%) had mature / hypermature cataracts ; corresponding percentages in the Phaco group (total 390 patients) were 2%, 30%, 34%, 8% and 26%, respectively (Table 2). These differences were not statistically significant ( $\chi^2$  [d.f.=4]=4.8 ; P=0.31).

Sixty-six (12.7%) of 520 patients in the current study suffered from diabetes mellitus.

The percentage of diabetics in SICS group with posterior subcapsular cataracts was 50%.

**Table 2: Grades of cataracts in patients undergoing small incision cataract surgery or phacoemulsification cataract surgery at a tertiary eye care hospital**

Grades of nuclear cataract (LOCS classification)	In small incision cataract surgery (SICS) group			In phacoemulsification cataract surgery(Phaco) group			Overall Total
	Males	Females	Sub total (%)*	Males	Females	Sub total (%)*	
NC 2	0	3	3 ( 2%)	1	7	8 ( 2%)	11
NC 3	18	34	52(40%)	41	76	117(30%)	169
NC 4	10	28	38(29%)	32	100	132(34%)	170
NC 5	1	7	8 (6%)	9	24	33( 8%)	41
MC / HMC	10	19	29(23%)	39	61	100(26%)	129
Group Total	39	91	130(100%)	122	268	390(100%)	520

\* Percentage of group total

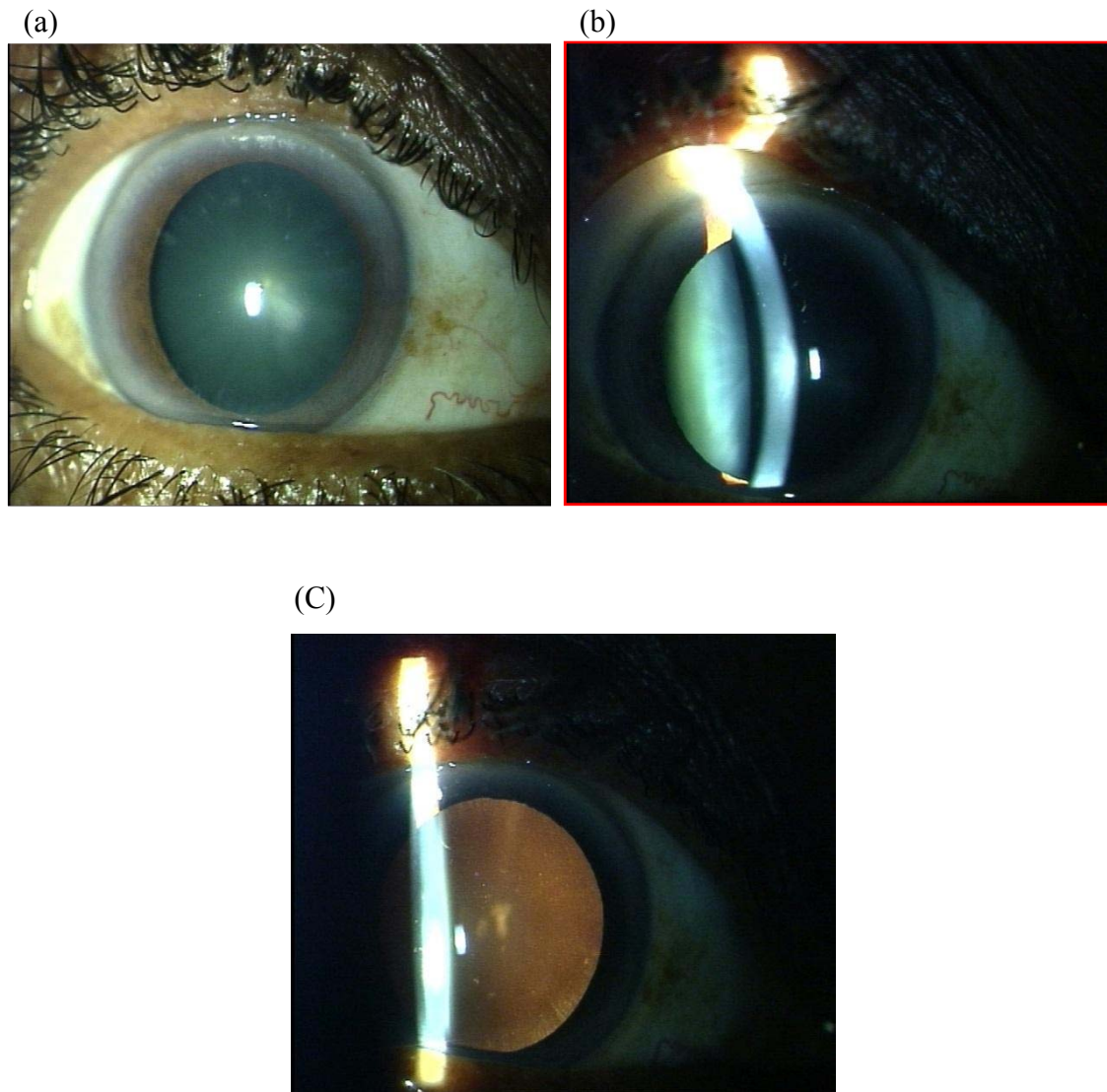
*Abbreviations:* LOCS =Lens Opacities Classification System III

NC= nuclear cataract ; MC/HMC= mature cataract/hypermature cataract

*Statistical Analysis*

- a. Proportions of patients with different grades of cataracts in SICS group versus(vs.) Phaco group:  $\chi^2$  (d. f. = 4) = 4.8 ; P=0.31
- b. **Proportions of males vs. females with different grades of cataracts:**  
 $\chi^2$  (d. f. = 4) = 10.6; P= 0.03 ; Yates' corrected  $\chi^2$  (d. f. = 4) = 8.63; P= 0.07

## NUCLEAR CATARACT GRADE 2



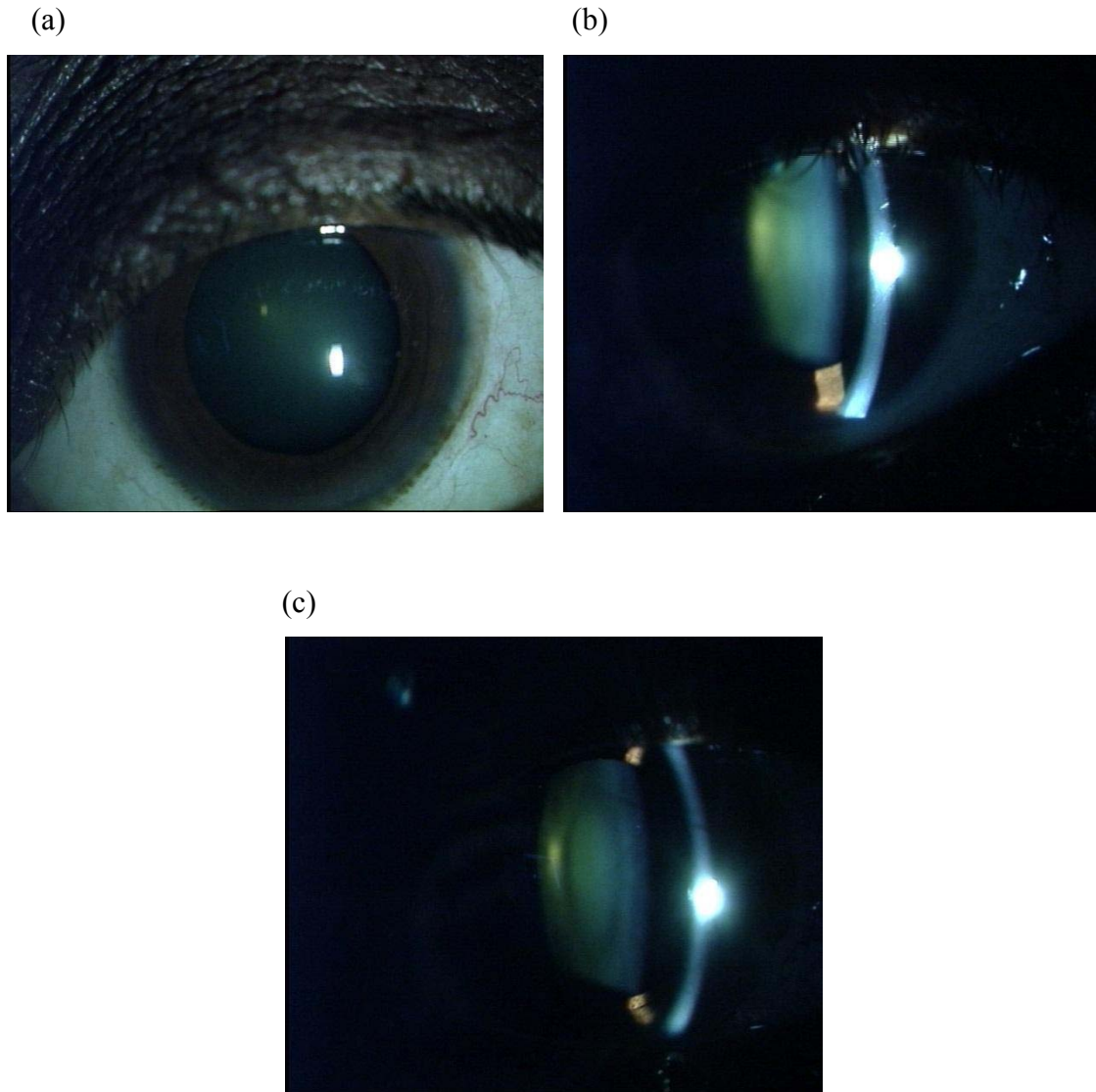
**Figure 3. Anterior segment photographs of the right eye showing Grade 2 nuclear cataract**

(a) – on diffuse illumination (8X magnification)

(b) – with stereoscopic slit beam illumination

(c) – on retroillumination with slit

### NUCLEAR CATARACT GRADE 3



**Figure 4. Anterior segment photographs of the right eye showing Grade 3 nuclear cataract**

(a) – on diffuse illumination (8X magnification)

(b) – with Stereoscopic slit beam illumination

(c) – on retroillumination

## NUCLEAR CATARACT GRADE 4

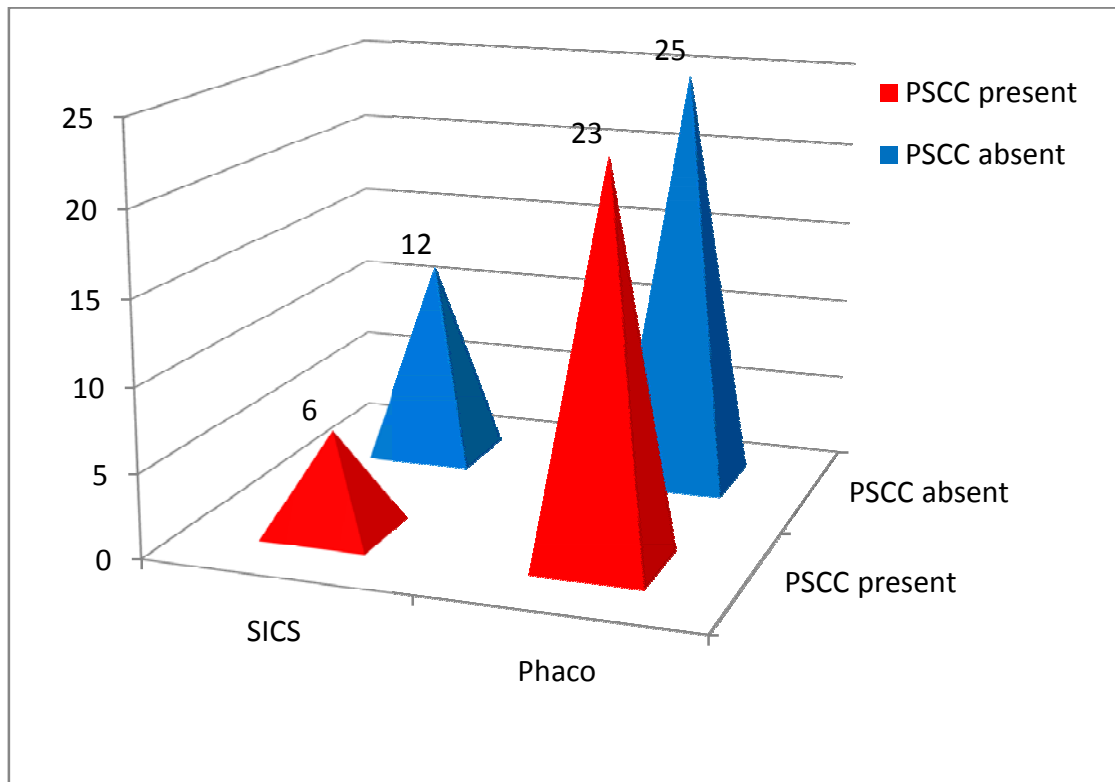


**Figure 5. Anterior segment photographs of the right eye showing Grade 4 nuclear cataract**

- (a) – on diffuse illumination (8X magnification)
- (b) – on diffuse illumination (12X magnification)
- (c) – on slit beam illumination



**Figure 6. Occurrence of posterior subcapsular cataracts in patients with diabetes mellitus undergoing small incision cataract surgery or phacoemulsification cataract surgery at a tertiary eye care hospital.**



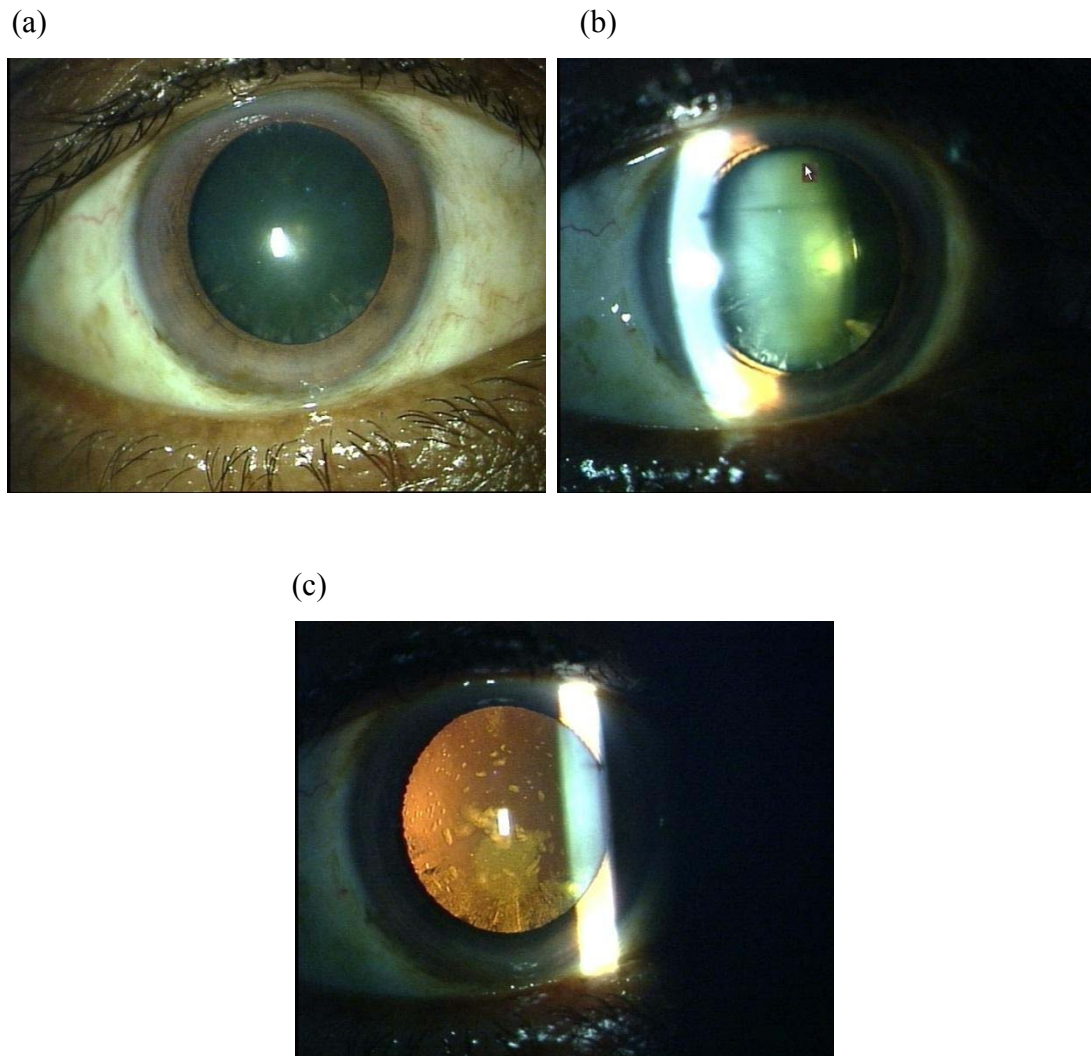
Percentage of diabetics in this study : 66 out of 520 = 12.7%

Percentage of diabetics in SICS group with posterior subcapsular cataracts= 50%

Percentage of diabetics in Phaco group with posterior subcapsular cataracts=47.9%

Posterior subcapsular cataracts occurred in 23 out of total 66 diabetics= 43.9%, in the current study

**POSTERIOR SUBCAPSULAR CATARACT GRADE 1**



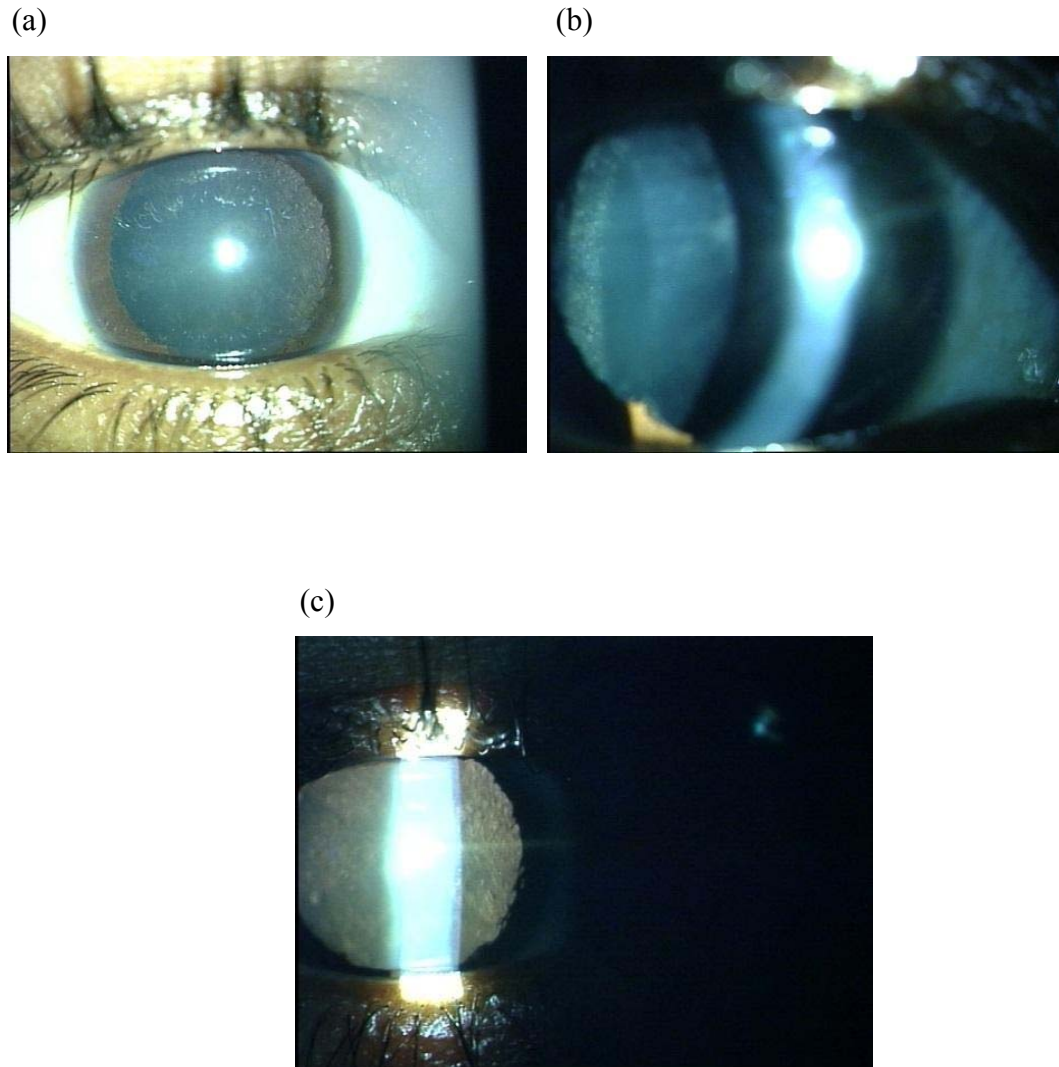
**Figure 7. Anterior segment photographs of the right eye showing posterior subcapsular cataract Grade 1 on**

(a) – Diffuse illumination (8X magnification)

(b) – Slit beam illumination

(c) – on retroillumination

## POSTERIOR SUBCAPSULAR CATARACT GRADE 2



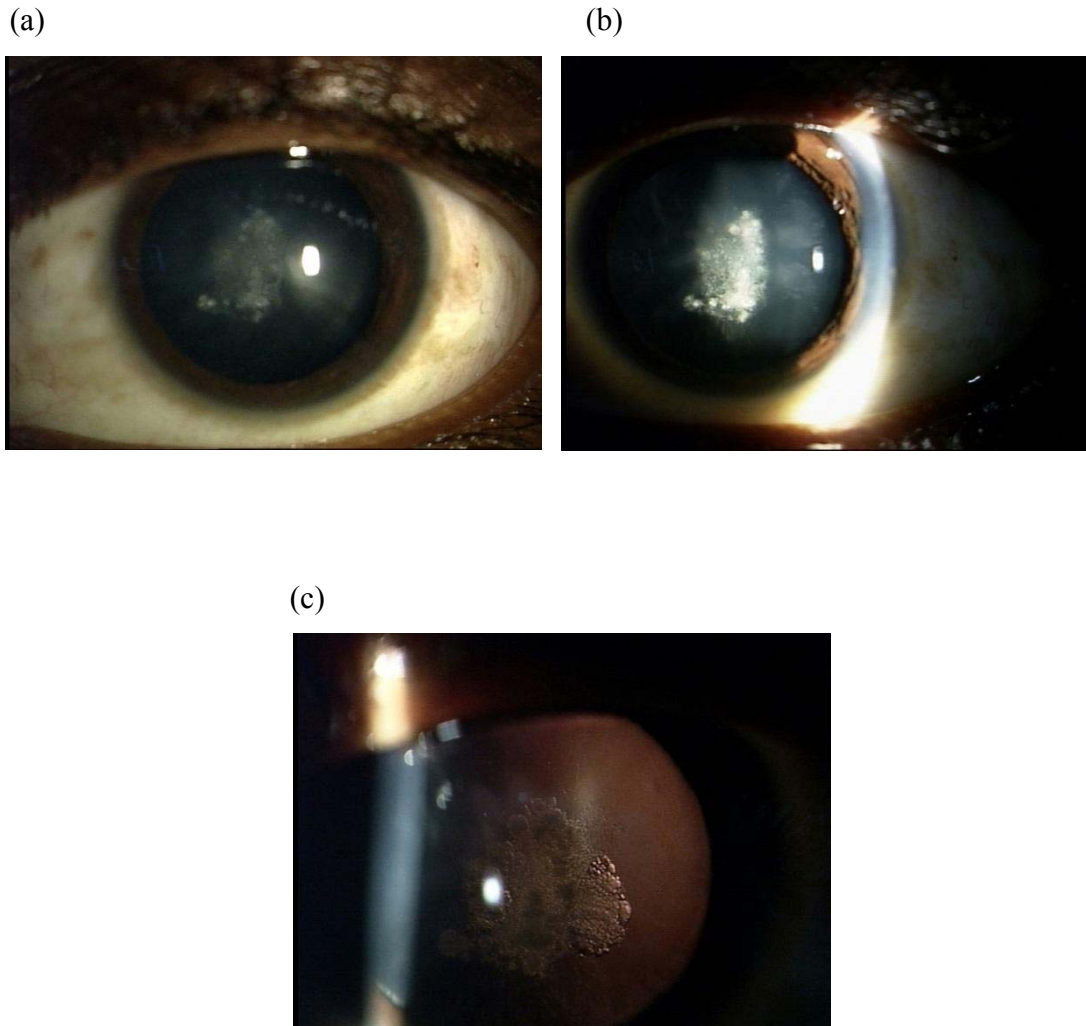
**Figure 8. Anterior segment photographs of the right eye showing posterior subcapsular cataract Grade 2 on**

(a) – Diffuse illumination (8X magnification)

(b) – on Slit beam illumination

(c) – on retroillumination

### POSTERIOR SUBCAPSULAR CATARACT GRADE 3



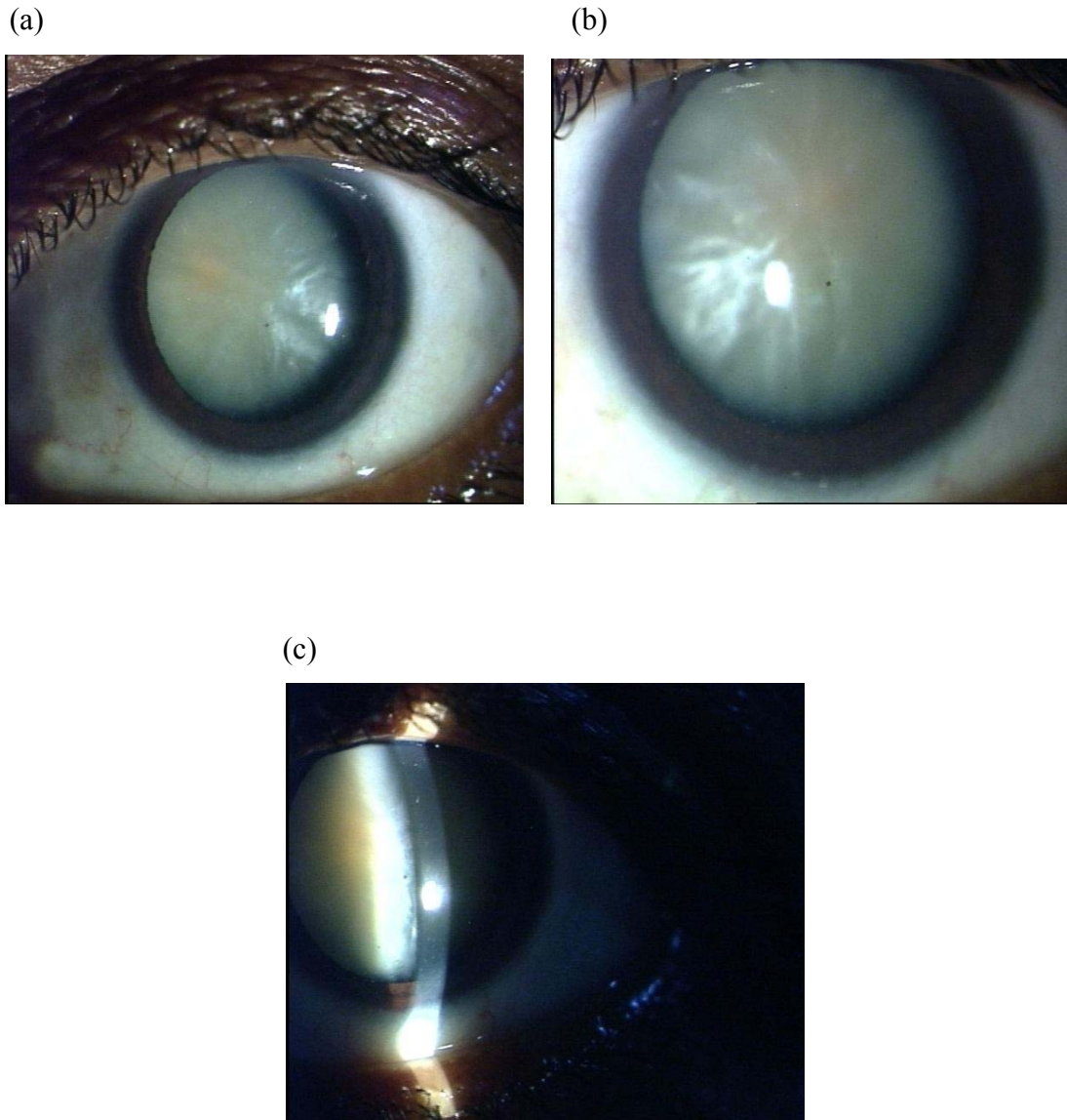
**Figure 9. Anterior segment photographs of the right eye showing posterior subcapsular cataract Grade 3 on**

(a) – Diffuse illumination (8X magnification)

(b) – on Slit beam illumination

(c) – on retroillumination

## MATURE CATARACT

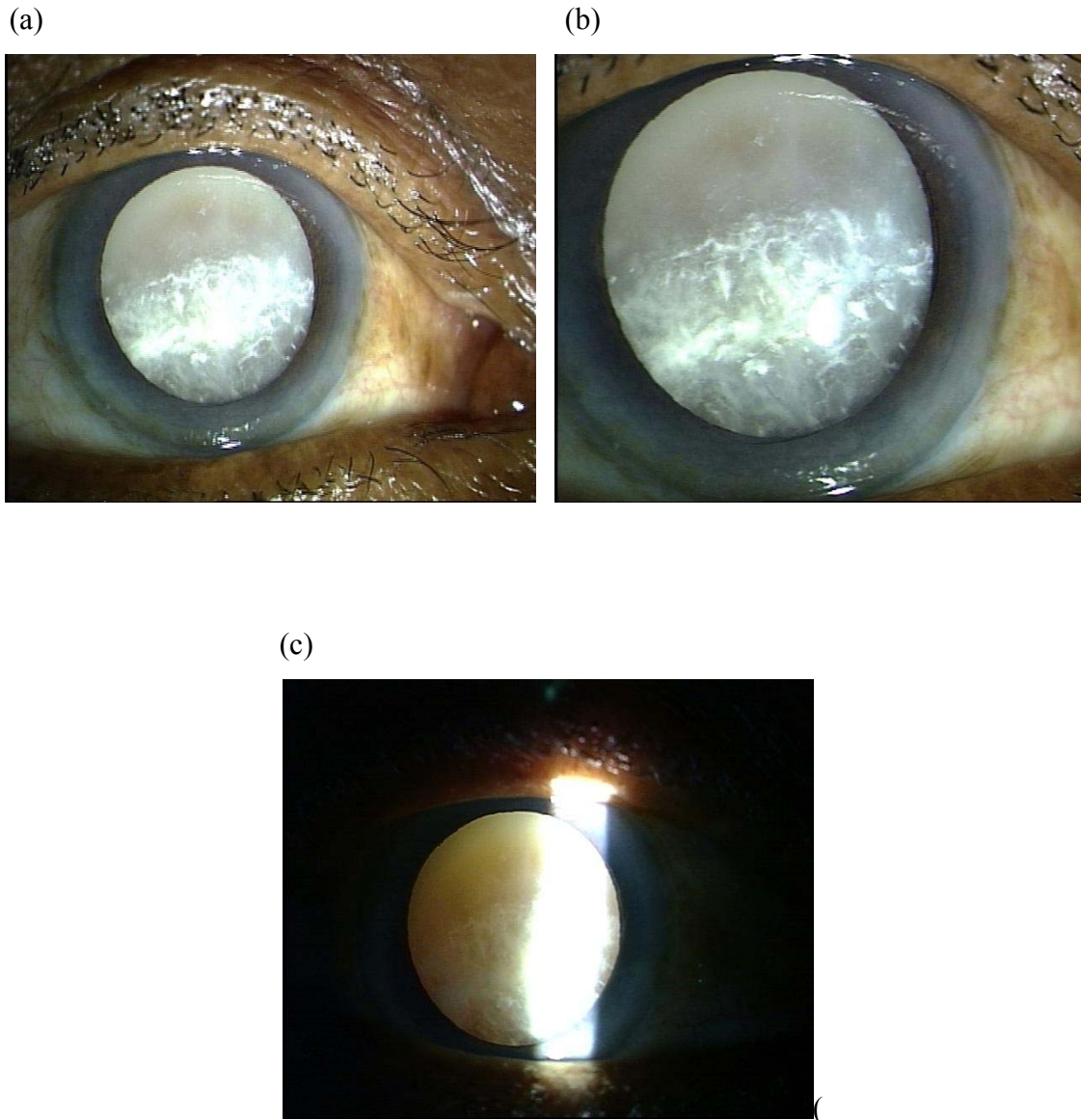


**Figure 10. Anterior segment photographs of the left eye showing mature cataract on**

- (a) – Diffuse illumination (8X magnification)
- (b) – Diffuse illumination (12X magnification)
- (c) – on Slit beam illumination



## HYPERMATURE CATARACT



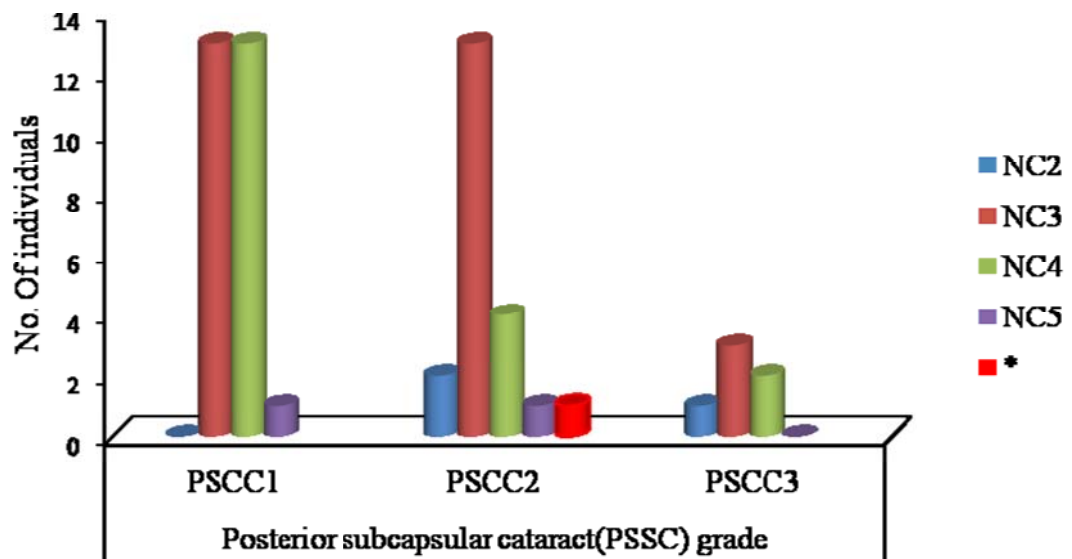
**Figure 11. Anterior segment photographs of the right eye showing hypermature cataract (sclerotic type) on**

(a) – Diffuse illumination (8X magnification)

(b) – Diffuse illumination (12X magnification)

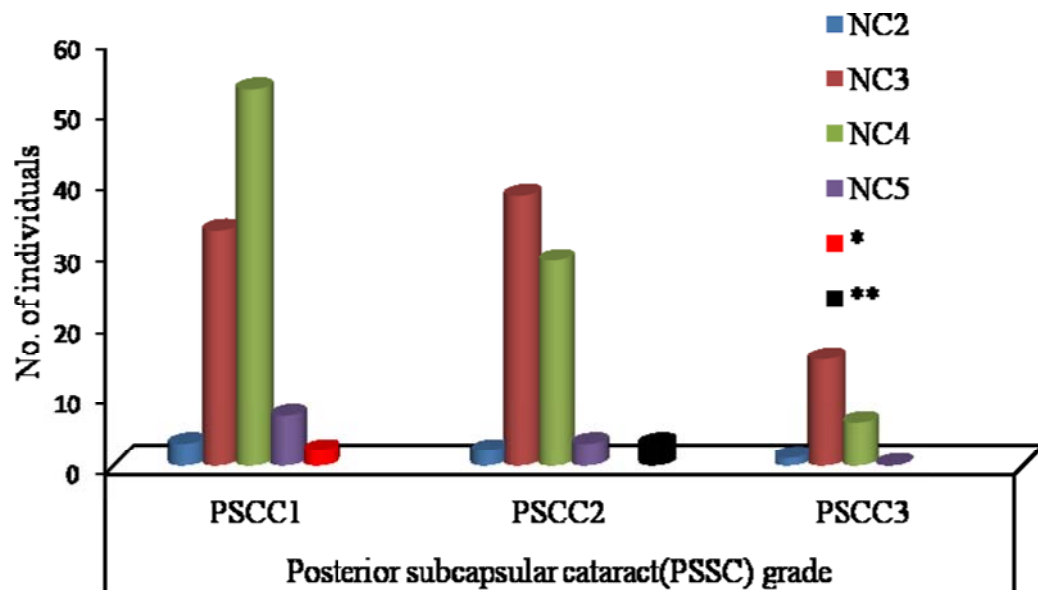
(c) – Slit beam illumination

**Figure 12 :** Association of posterior subcapsular cataracts with various grades of nuclear cataracts in patients undergoing small incision cataract surgery at a tertiary eye hospital



\* Posterior subcapsular cataract (grade 2) not associated with nuclear cataract

**Figure 13 :** Association of posterior subcapsular cataracts with various grades of nuclear cataracts in patients undergoing phacoemulsification cataract surgery at a tertiary eye hospital



\* Posterior subcapsular cataract (grade 1) not associated with nuclear cataract

\*\* Posterior subcapsular cataract (grade 2) not associated with nuclear cataract



The percentage of diabetics in the Phaco group with posterior subcapsular cataracts was 47.9%. Posterior subcapsular cataracts occurred in 23 out of total 66 diabetics= 43.9%, in the current study (Fig. 6).

In the SICS group (total 130 patients), 54 patients (41.5 %) had different grades of posterior subcapsular cataract (PSSC) in association with nuclear cataract (Fig.11); 29 had PSSC1, 21 had PSSC 2 and six had PSSC 3. In the Phaco group (total 390 patients), 195 (50%) had PSSC cataracts in association with nuclear cataracts (Fig. 12); 95 had PSSC 1, 78 had PSSC 2 and 21 had PSSC 3 (Fig. 12). These differences were not statistically significant ( $\chi^2$  with Yates' correction [d.f.=1]=2.5 ; P=0.11).

### **1.3. 2 In Males versus Females**

In males, NC2 cataracts were noted in 0.4 %, NC3 cataracts in 37%, NC4 cataracts in 26%, NC5 cataracts in 6.2% and mature / hypermature cataracts in 30.4% ; in female patients, the percentages were 3%, 31%, 35%, 9% and 22%, respectively. Interestingly, these gender differences approached statistical significance ( $\chi^2$  [d.f.=4] = 10.6, P = 0.03 : with Yates' correction,  $\chi^2 = 8.63$ , P = 0.07) (Table 2).

PSSC cataracts were noted (in association with nuclear cataracts) in 71 males and 179 females (Figures 11 and 12). In males, PSSC 1 cataracts were seen in 33 (46.5% of 71 males), PSSC 2 was seen in 30 (42.3 %) and

PSSC 3 in eight (11.2% of 71 males); in females the corresponding numbers (percentages) were 89 (49.7% of 179 females), 69 (38.5%) and 19 (10.6% of 179 females (Fig. 11 and 12). These differences were not statistically significant ( $\chi^2$  with Yates' correction [d.f.=1] = 1.26, P = 0.26).

#### **1.4 Pre-operative and post-operative visual acuity**

In the 130 patients who had undergone SICS, the post-operative mean visual acuity (decimals),  $0.58 \pm 0.02$ , was significantly better than the pre-operative mean visual acuity of  $0.07 \pm 0.01$  (unpaired 't' [d.f.=258] = 21.6 ; P < 0.0001) (Table 3). This significant improvement was seen in both males and females (Table 3).

Similarly, in the 390 patients who underwent phacoemulsification surgery, the post-operative mean visual acuity ( $0.64 \pm 0.01$ ) was significantly better than the pre-operative mean visual acuity of  $0.07 \pm 0.01$  (unpaired 't' [d.f.= 776] = 38.7 ; P < 0.0001) (Table 4). This significant improvement was seen in both males and females (Table 4).

A comparison was made of the visual acuity results obtained in the SICS and Phaco groups (Tables 3 & 4). With reference to **mean pre-operative visual acuities** (decimal units), no significant differences between the groups were observed when total patients (SICS [ $0.07 \pm 0.01$ ]

versus Phaco [0.07 + 0.01] ;  $t = 0.28$  [d.f.=519,  $P = 0.78$ ), males (SICS [0.08 + 0.01] versus Phaco [0.06 + 0.01];  $t = 1.25$  [d.f. = 159],  $P=0.21$ ) or females (SICS [0.07 + 0.01] versus Phaco [0.07 + 0.01]) were compared. However, when **post-operative** mean visual acuities (decimal units) were analysed and compared, the following interesting observations emerged (Tables 3 & 4):

- a) there was no significant difference between the post-operative mean visual acuity in the SICS and Phaco groups in males (SICS [0.67 + 0.04] versus Phaco [0.63 + 0.03];  $t = 0.88$  (d.f.=159),  $P = 0.38$ );
- b) in females, the post-operative mean visual acuity in the Phaco group [0.64 + 0.02] was *significantly higher* (better) than that in the SICS group [ 0.55 + 0.03] ( $t = 2.79$  [d.f.=357] ;  $P = 0.0055$ ).
- c) When all patients were considered, the post-operative mean visual acuity in the Phaco group (0.64 + 0.01) was higher (better) than that in the SICS group [0.58 + 0.02], and this difference approached statistical significance ( $t=1.86$  [d.f.=518] ;  $P = 0.06$ ).

**Table 3 : Mean pre-operative and mean post-operative visual acuity in patients undergoing small incision cataract surgery at a tertiary eye care hospital**

Gender of patients	Number of patients	Mean (+ SEM) visual acuity (decimals)		Statistical Analysis (Student 't'[paired])
		Pre-operative	Post-operative	
Male	39	0.08 + 0.01	0.67 + 0.04	t = 12.6 (d. f.=38); P < 0.0001
Female	91	0.07 + 0.01	0.55 + 0.03	t= 17.8 (d. f.=90); P < 0.0001
All	130	0.07 + 0.01	0.58 + 0.02	t = 21.6 (d. f.=129); P < 0.0001
Statistical Analysis(Student 't' [unpaired]) Males versus Females		t=0.82 (d.f.=128) P=0.42	t=2.52 (d.f.=128) P=0.01	

Abbreviation SEM = Standard error of mean, d.f.= degree of freedom

**Table 4 : Mean pre-operative and mean post-operative visual acuity in patients undergoing phacoemulsification cataract surgery at a tertiary eye care hospital**

Gender of patients	Number of patients	Mean + SEM visual acuity (decimals)		Statistical Analysis (Student 't'[paired])
		Pre-operative	Post-operative	
Males	122	0.06 + 0.01	0.63 + 0.03	t = 20.7 (d. f.=121); P < 0.001
Females	268	0.07 + 0.01	0.64 + 0.02	t= 33.1 (d. f.=267); P < 0.0001
All	390	0.07 + 0.01	0.64 + 0.01	t = 38.74 (d. f.=389); P < 0.0001
Statistical Analysis (Student 't'[unpaired]) Males versus Females		t=1.03 (d.f.=388) P=0.30	t=0.41 (d.f.=388) P=0.68	

Abbreviation SEM = Standard error of mean, d.f.= degree of freedom

### **1.5 Surgical and post-operative complications, pre-existing pathology and combined surgery**

The frequency of surgical and post-operative complications, pre-existing pathology and combined surgery was compared between the SICS and Phaco groups (Table 5). Complications were encountered in five (4%) of 130 patients in the SICS group and 17 (4.3%) of 390 patients in the Phaco group, this difference was not statistically significant ( $\chi^2$  [d.f.=1] = 0.06 ; P = 0.8) (Table 5). Pre-existing pathology was present in six (4.6%) of 130 SICS group patients and nine (2.3%) of 390 Phaco group patients (Table 5). This difference was not statistically significant ( $\chi^2$  [d.f.=1] = 1.9; P = 0.17). Combined surgery was needed for 13 (10.0 %) of 130 SICS group patients and 17 (4.4 %) of 390 phaco group patients (Table 5); this difference was statistically significant ( $\chi^2$  with Yates' correction [d.f.=1] = 4.72 ; P=0.03).

When differences between males and females in frequency of occurrence of these parameters were compared, no statistically significant differences were observed, except within the Phaco group, where pre-existing pathology was present in six (5%) of 122 males and three (1.1%) of 268 females (Yates'  $\chi^2$  (d.f.=1) = 3.81 ; P = 0.05).

**Table 5: Complications, pre-existing pathology and combined surgery in small incision or phacoemulsification cataract surgery at a tertiary eye care hospital**

Parameter	In small incision cataract surgery (SICS) group (total n = 130)			In phacoemulsification cataract surgery (Phaco)group(total n= 390)			Overall Total
	Males	Females	Sub total	Males	Females	Sub total	
Surgical complications	2	3	5	7	10	17	22
Pre-existing pathology	2	4	6	6	3	9	15
Combined surgery	4	9	13	5	12	17	30
Group total	8	16	24	18	25	43	67

**Statistical Analysis (Chi-square with Yates' correction)**

- a) Combined surgery needed for 13 (10.0 %) of 130 SICS group patients and 17 (4.4 %) of 390 phaco group patients;  $\chi^2$  with Yates' correction [d.f.=1] = 4.72 ; P=0.03.
- b) Phacogroup ; Pre-existing pathology in 6 of 122 males and 3 of 268 females  $\chi^2$  (d. f. = 1) = 3.81 ; P = 0.05

## **1.6 Post-operative visual outcome in relation to pre-operative cataract grade**

### **1.6.1 In SICS group versus Phacoemulsification group**

Different degrees of post-operative (PO) visual acuity (Snellen's units) in relation to pre-operative cataract grade were compared in patients who had undergone SICS and in those who had undergone phacoemulsification cataract surgery (Table 6). Of three patients in the SICS group who had had preoperative NC2 cataracts, two achieved PO visual acuity of 6/9 and one achieved 6/12, while of eight patients in the Phaco group who had had preoperative NC2 cataracts, two each achieved PO visual acuities of 6/6, 6/9, 6/12 and  $\geq 6/24$  (Table 6).

Fifty-one patients in the SICS group who had had preoperative NC3 grade cataracts achieved PO visual acuities of 6/6 (13 [25.5%] of 51 patients), 6/9 (21 [41.2%]), 6/12 (seven [13.2%]), 6/18 (four [7.8%]) and 6/24 or worse (six [11.8%] of 51 patients) whereas 117 patients in the Phaco group who had had preoperative NC3 grade cataracts achieved PO visual acuities of 6/6 (30 [25.6%] of 117 patients), 6/9 (51 [43.6%]), 6/12 (14 [11.97%]), 6/18 (eight [6.8%]) and 6/24 or worse (six [11.97%] of 117 patients) (Table 6); these differences were not statistically significant ( $\chi^2$  [d.f.=4]=0.19; P=0.99).

**Table 6: Post-operative visual acuity versus preoperative nuclear cataract grades\* in small incision or phacoemulsification cataract surgery at a tertiary eye hospital.**

Grade of nuclear cataract (LOCS)	Post-operative (PO) visual acuity (Snellen) SICS group (total no. = 130)						PO visual acuity(Snellen) Phaco group (total no. = 390)						Overall Total
	6/6	6/9	6/12	6/18	$\geq$ 6/24	Group total	6/6	6/9	6/12	6/18	$\geq$ 6/24	Group total	
	NC 2	-	2	1	-	-	3	2	2	2	-	2	
NC 3	13	21	7	4	6	51	30	51	14	8	14	117	168
NC 4	8	14	8	5	3	38	41	45	20	10	16	132	170
NC 5	1	3	1	1	2	8	11	8	7	2	5	33	41
MC/HMC	4	9	9	2	6	30	28	41	11	11	9	100	130
Group total	26	49	26	12	17	130	112	147	54	31	46	390	520

Abbreviations: LOCS= Lens Opacities Classification System; no. = number; SICS = small incision cataract surgery; NC=nuclear cataract; MC/HMC= mature/hypermature cataract;

Statistical Analysis ( $\chi^2$  [degree of freedom {d.f.}= 4] with Yates' correction where necessary)

1. NC 2 cataract : No. of patients in SICS group vs. Phaco group with PO visual acuities (statistical analysis not done)
2. NC 3 cataract : No. of patients in SICS group vs. Phaco group with PO visual acuities;  $\chi^2 = 0.19$ ; P= 0.99
3. NC 4 cataract : No. of patients in SICS group vs. Phaco group with PO visual acuities;  $\chi^2 = 3.23$ ; P= 0.5
4. NC 5 cataract: No. of patients in SICS group vs. Phaco group with PO visual acuities;  $\chi^2 = 2.36$ ; P= 0.67
5. **MC/HMC cataract: No. of patients in SICS group vs. Phaco group with PO visual acuities 6/6 and 6/9 vs.  $\geq$  6/12. Yates'  $\chi^2$  (d.f.=1)=5.47 P= 0.02**



Thirty-eight patients in the SICS group who had had preoperative NC4 grade cataracts achieved PO visual acuities of 6/6 (eight [21%] of 38 patients), 6/9 (14 [36.8%]), 6/12 (eight [21%]), 6/18 (five [13.2 %]) and 6/24 or worse (three [8%] of 38 patients) whereas 132 patients in the Phaco group who had had preoperative NC4 grade cataracts achieved PO visual acuities of 6/6 (41 [31.1%] of 132 patients), 6/9 (45 [34.1%]), 6/12 (20 [15.2%]), 6/18 (10 [7.6 %]) and 6/24 or worse (16 [12 %] of 132 patients) (Table 6); these differences were not statistically significant ( $\chi^2$  [d.f.=4]=3.23; P=0.5). Similarly, of eight patients in the SICS group and 33 patients in the Phaco group who had had preoperative NC5 grade cataracts, the PO visual acuities achieved were 6/6 (in one of 8 [SICS] and 11 [33.3%] of 33 Phaco group patients), 6/9 (in three [37.5%] SICS patients and in eight [24.2%] Phaco patients), 6/12 (in one of eight [SICS] and in seven [21.2%] of 33 Phaco group patients), 6/18 (in one of eight [SICS] and in two [6.1%] of 33 Phaco group patients) and 6/24 or worse (in two [25%] of eight patients [SICS] and in five [15.2%] of 33 [Phaco group] patients) (Table 6); these differences were not statistically significant ( $\chi^2$  [d.f.=4]=2.36; P=0.67).

Interestingly, significant differences between the SICS and Phaco groups were observed when PO visual acuities were assessed against pre-

operative mature and hypermature cataract grades (Table 6). In the Phaco group, of 100 individuals who exhibited preoperative mature or hypermature cataracts, 69 (69%) achieved PO visual acuities of 6/6 or 6/9 while 31 (31%) achieved PO visual acuities of 6/12, 6/18 or 6/24 and worse ; in contrast, in the SICS group, of 30 individuals who had had preoperative mature or hypermature cataracts, 13 (43.3%) achieved PO visual acuities of 6/6 or 6/9 while 17 (56.7%) achieved PO visual acuities of 6/12 or worse (Table 6). This difference was statistically significant ( $\chi^2$  [d.f.=1]=5.47; P=0.02).

### **1.6.2 In males versus females**

Different degrees of PO visual acuity (Snellen's units) were also assessed in relation to pre-operative cataract grade from another viewpoint, that is, gender of the patients (Table 7). Interestingly, significant differences between males and females were observed when PO visual acuities were assessed against pre-operative NC3 cataract grade (Table 7). Of 58 males who had had preoperative NC3 grade cataracts, 24 (41.4 %) achieved a PO visual acuity of 6/6, 13 (22.4 %) achieved 6/9, 12 (20.7 %) achieved 6/12, four (6.9 %) achieved 6/18 and five (8.6%) achieved a PO visual acuity of 6/24 or worse; in contrast, of 110 females who had had preoperative NC3 grade cataracts, 19 (17.3 %) achieved a PO visual acuity

of 6/6, 59 (53.6 %) achieved 6/9, nine (8.2 %) achieved 6/12, eight (7.3 %) achieved 6/18 and 15 (13.6%) achieved a PO visual acuity of 6/24 or worse (Table 7). These differences were highly statistically significant ( $\chi^2$  [d.f.=4]=22.8; P=0.00014).

With reference to individuals with preoperative NC4 grade cataracts, PO visual acuities did not differ significantly between males and females (Table 7). With reference to preoperative NC5 grade cataracts in 10 males and 31 females, PO visual acuities achieved were 6/6 (in one [10%] of 10 males and in 11 [35.5 %] of 31 females), 6/9 (in two [20%] of 10 males and in nine [29.0 %] of 31 females), 6/12 (in two [20%] males and in six [19.4 %] females), 6/18 (in one [10%] of 10 males and in two [6.5%] of 31 females) and 6/24 or worse (in four [40%] of 10 males and three [9.7%] of 31 females) (Table 7); these differences approached statistical significance ( $\chi^2$  [d.f.=4]=3.66; P=0.06). With reference to individuals with preoperative mature or hypermature grade cataracts, PO visual acuities did not differ significantly between males and females (Table 7).

**Table 7: Post-operative visual acuity versus different preoperative nuclear cataract grades\* in males and females undergoing cataract surgery at a tertiary eye hospital.**

Grade of nuclear cataract (LOCS)	Post-operative visual acuity (Snellen)						Post-operative visual acuity(Snellen)						Overall Total
	Males (total no. = 161)						Females (total no. = 390)						
	6/6	6/9	6/12	6/18	≥ 6/24	Group total	6/6	6/9	6/12	6/18	≥ 6/24	Group total	
NC 2	0	0	0	0	1	1	2	4	3	0	1	10	11
NC 3	24	13	12	4	5	58	19	59	9	8	15	110	168
NC 4	16	11	8	3	4	42	33	48	20	12	15	128	170
NC 5	1	2	2	1	4	10	11	9	6	2	3	31	41
MC/HMC	10	18	9	5	8	50	22	32	11	8	7	80	130
Group total	51	44	31	13	22	161	87	152	49	30	41	359	520

*Abbreviations:* LOCS= Lens Opacities Classification System; no. = number; NC=nuclear cataract ; MC/HMC= mature/hypermature cataract; PO=post-operative

*Statistical Analysis* ( $\chi^2$  [degree of freedom {d.f.}= 4] with Yates' correction where necessary)

1. NC 2 cataract (statistical analysis not done)
2. **NC 3 cataract: No. of males vs. no. of females with PO visual acuities;  $\chi^2 = 22.8$ ; P= 0.0001**
3. **NC 4 cataract** : No. of males vs. no. of females with PO visual acuities;  $\chi^2 = 3.4$ ; P= 0.5
4. **NC 5 cataract: No. of males vs. no. of females with PO visual acuities;  $\chi^2 = 3.66$ ; P= 0.06** (with Yates' correction, p=0.1)
5. **MC/HMC cataract:** No. of males vs. no. of females with PO visual acuities  $\chi^2 = 2.6$ ; P= 0.63

## **1.7 Post-operative first day and post-operative seventh day visual acuity**

In the 39 male patients who had undergone SICS, the 7<sup>th</sup> post-operative day mean visual acuity (decimals),  $0.82 + 0.25$ , was significantly better than the 1<sup>st</sup> post-operative day mean visual acuity of  $0.67 + 0.3$  (paired 't' [d.f.=38] = 3.9 ; P= 0.0003) (Table 8). In the 91 female patients who had undergone SICS, the 7<sup>th</sup> post-operative day mean visual acuity (decimals) of  $0.82 + 0.27$  was significantly better than the 1<sup>st</sup> post-operative day mean visual acuity of  $0.54 + 0.24$  (paired 't' [d.f.=90] = 11.71 ; P < 0.0001) (Table 8).

Similarly, in the 122 male patients who had undergone phacoemulsification cataract surgery, the 7<sup>th</sup> post-operative day mean visual acuity (decimals) of  $0.86 + 0.25$ , was significantly better than the 1<sup>st</sup> post-operative day mean visual acuity of  $0.63 + 0.3$  (paired 't' [d.f.=121] = 12.3 ; P < 0.0001) (Table 8) while in the 268 females who had undergone phacoemulsification cataract surgery, the 7<sup>th</sup> post-operative day mean visual acuity (decimals) of  $0.87 + 0.24$  was significantly better than the 1<sup>st</sup> post-operative day mean visual acuity of  $0.64 + 0.3$  (paired 't' [d.f.=267] = 18.75 ; P < 0.0001) (Table 8).

**Table 8: Mean post-operative 1<sup>st</sup> day versus mean post-operative 7<sup>th</sup> day visual acuity values in patients undergoing small incision or phacoemulsification cataract surgery at a tertiary eye care hospital**

Surgical group	Gender of patients	Number of patients	Mean $\pm$ SD visual acuity (decimals) at		Statistical Analysis (Student 't'[(paired)])
			1 <sup>st</sup> Post-operative day	7 <sup>th</sup> Post-operative day	
SICS	Males	39	0.67 $\pm$ 0.3	0.82 $\pm$ 0.25	t = 3.9 (d. f.=38); P =0.0003
	Females	91	0.54 $\pm$ 0.24	0.82 $\pm$ 0.27	t = 11.71 (d. f.=90); P =<0.0001
Phaco	Males	122	0.63 $\pm$ 0.3	0.86 $\pm$ 0.25	t= 12.3 (d. f.=121); P < 0.0001
	Females	268	0.64 $\pm$ 0.3	0.87 $\pm$ 0.24	t= 18.75 (d.f.=267); P < 0.0001

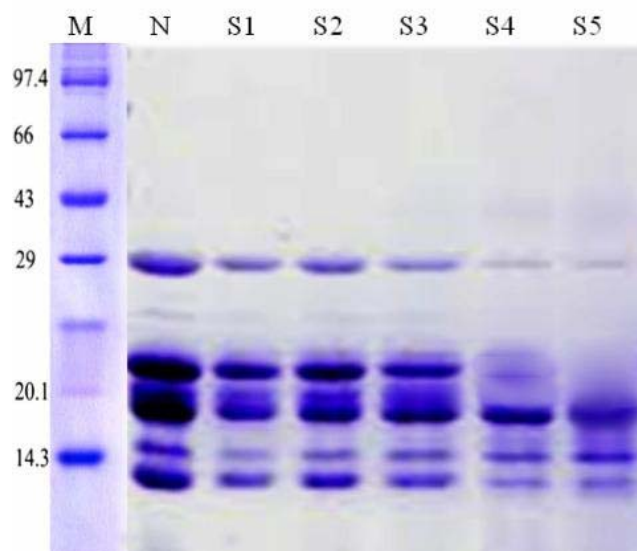
Abbreviation SICS= small incision cataract surgery; SD=standard deviation ; d.f.= degree of freedom

## 2. BIOCHEMICAL ASPECTS

### 2.1 SDS-PAGE

After SDS-PAGE, the gel was imaged and analyzed with QuantityOne software. In the normal lens nuclei (Lane N), most proteins were 12 to 30 kDa (Fig. 14). In the cataractous lens samples (Lanes S1, S2, S3, S4 and S5), the band intensities at ~20 kDa visibly ( $P < 0.05$ ) showed a gradual reduction in intensity, when compared with the band intensity of normal lens nuclei. With increasing nuclear sclerosis (NC2 to NC5 to mature/hypermature cataracts, corresponding to lanes S1 to S5), the staining intensity of bands corresponding to 29 and 20 kDa proteins was found to notably decrease (Fig. 14). In mature/hypermature cataractous lens samples (lane S5), many proteins were found to be missing in the gel, when compared to the pattern in normal lenses (lane N). In contrast, the distribution of proteins in NC2 cataractous lens samples (lane S1) was similar to that obtained in samples from normal lenses (lane N). These perceived differences in band staining intensity, as assessed by the naked eye, were reinforced by intensity analysis of the SDS-PAGE images using computer software.

**Figure 14 : Sodium dodecyl sulphate polyacrylamide gel electrophoretic image of various grades of cataract**



M - protein molecular weight marker

N - normal human lens (9 bands)

S1 - human nuclear cataract, Grade 2 (7 bands)

S2 - human nuclear cataract, Grade 3 (6 bands)

S3 - human nuclear cataract, Grade 4 (5 bands)

S4 - human mature cataract (4 bands)

S5 - human hypermature cataract (4 bands)



## 2.2 Two dimensional gel electrophoresis

To further confirm the protein degradation observed on SDS-PAGE, two dimensional gel electrophoresis (2-DE) was performed and the gel spots corresponding to  $\alpha$ -crystallin from the different grades of cataractous lenses were selected (Fig. 15). These spots were then subjected to MALDI-TOF analysis in order to ascertain the crystallin identification (Table 9). Alpha-crystallin was identified in all the lens samples, although the concentration varied.

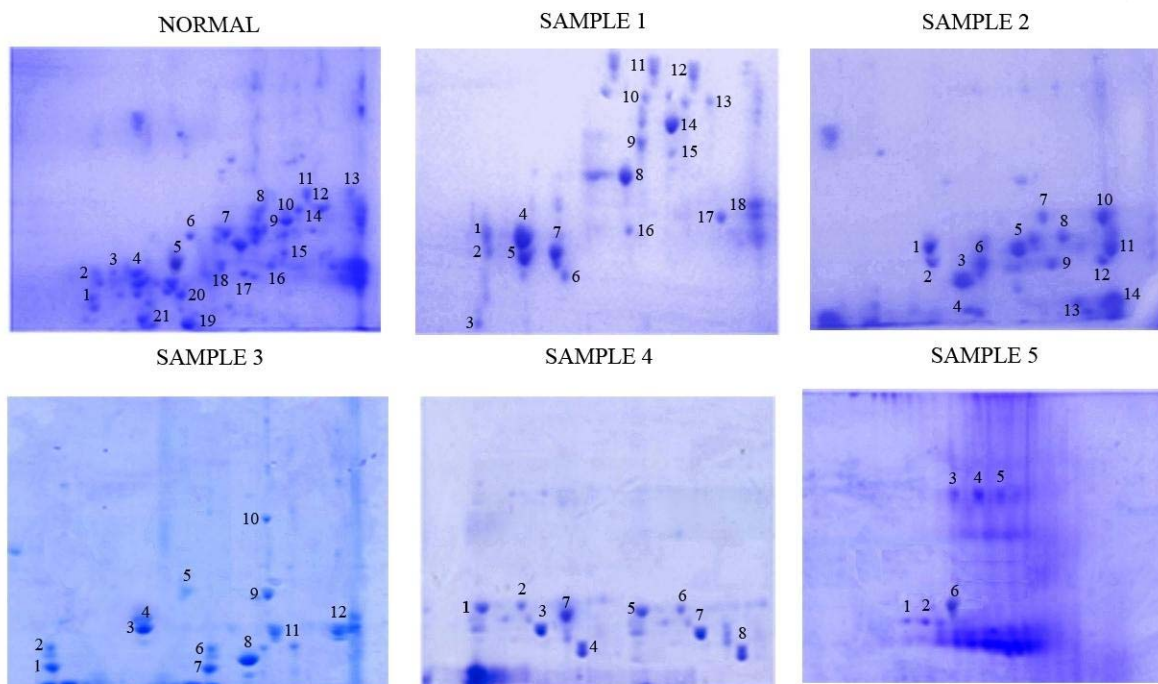
**Table 9. Identification of crystallin protein by mass spectrometry (MALDI)**

<b>Protein fraction and Spot Number</b>	<b>Crystallin ID [Accession Number]</b>	<b>Matched masses*</b>	<b>Number of Residues covered</b>	<b>% coverage</b>
5 (normal lens)	$\alpha$ CRY5 (P02511)	14	20	51
7 (cataract lens)	$\alpha$ CRY5 (P02511)	12	15	41
6 (cataract lens)	$\alpha$ CRY5 (P02511)	8	38	59
4 (cataract lens)	$\alpha$ CRY5 (P02511)	14	38	73
7 (cataract lens)	$\alpha$ CRY5 (P02511)	14	43	82
6 (cataract lens)	$\alpha$ CRY5 (P02511)	13	34	63

MALDI=Matrix-assisted laser desorption/ionization

\*matched masses included trypsin digestion products with 0-2 missed cuts and partial oxidation of methionine. Mass tolerance was 200 ppm.

**Figure 15 : Two-dimensional gel electrophoretic images of various grades of cataract**



Sample 1 - human nuclear cataract, Grade 2 (7 bands)

Sample 2 - human nuclear cataract, Grade 3 (6 bands)

Sample 3 - human nuclear cataract, Grade 4 (5 bands)

Sample 4 - human mature cataract (4 bands)

Sample 5 - human hypermature cataract (4 bands)

# *Discussion*

## DISCUSSION

Cataract continues to be the leading cause of global blindness, except in First World countries<sup>72</sup>. In 2002, the WHO estimated that 50% of global blindness was due to cataract<sup>5,6,8</sup>. The emerging demographic structure of the population worldwide, shows an increasing proportion of elderly patients<sup>73,74,75</sup>. This enhanced life- expectancy in Third World countries suggests that the total number of cataract- blind individuals is likely to increase. It has been reported to be responsible for 50-80% of bilateral blindness<sup>76,77</sup>. In one study conducted on an older population in a rural area of southern India, subjects with age-related cataract had an associated decrease in the quality of life and vision function, independent of presenting visual acuity in the better eye<sup>76,78</sup>. Hence it becomes vital to tackle cataract which causes potentially reversible visual loss when detected early, and thus improve the quality of life in the elderly.

Irrespective of its etiology or subtype, cataract remains a condition that cannot be alleviated medically. At present, only one approved and widely accepted treatment is performed, which involves the surgical removal of the cataractous lens and its replacement with an intraocular plastic lens<sup>72</sup>. Cataract surgery itself has been revolutionised over the last 20 years. The modern day life-style, which dictates the need for excellent

vision, has led to increased expectations of best results following cataract surgery<sup>79</sup>. From being a surgical procedure that required long hospital stay, with prolonged visual rehabilitation, this has now been transformed into a short day-care procedure with immediate, tangible benefits. Although cataract surgery is associated with a degree of ocular morbidity, it is now possible to offer cataract surgery at an earlier stage of cataract maturation, therein sparing patients from an extended period of severe visual impairment. It hence becomes mandatory to judge the best time to refer and operate on a patient with cataracts

The present study is thus an attempt to discern the frequency of occurrence of different types of senile cataract (based on the Lens Opacification Classification System (LOCS)Version III in patients presenting at a tertiary eye care hospital over a defined time period.It also aims identify putative correlations between types of senile cataract and clinical parameters such as age, gender, presence of systemic diseases (such as diabetes mellitus, hypertension and hypercholesterolemia), presence of ocular problems (refractive errors, glaucoma).

Patients who were enrolled in the study underwent monocular cataract surgery either by SICS (SICS group) or by phacoemulsification (Phaco group), as described earlier. There were 130 patients in SICS group and 390 patients in the Phaco group. Interestingly, the number of females

undergoing cataract surgery (91 in SICS group, 268 in Phaco group) exceeded the number of males (39 in SICS and 122 in Phaco groups). Although this difference was not statistically significant, it was consistent with the results of an Australian study (Blue Mountain Eye Study) in which the 10-year incidence of cataract was significantly higher in women than in men<sup>80</sup>. The Aravind Comprehensive Eye study (Nirmalan PK *et al.*, 2004)<sup>77</sup> reported a significantly lower prevalence of cataract (age-adjusted) in males.

A meta-analysis of cataract surveys in Third World countries reported that the cataract surgical coverage rate in the population was approximately 1.5 times higher for males than for females<sup>81</sup>. In spite of the lower cataract surgical coverage rate, 63% of all cataract surgeries were done in females; had females received cataract surgery at the same rate as males, cataract blindness prevalence would be reduced by a median of 12.5%<sup>82</sup>. Cataract extractions may be performed more frequently in women than in men, by a 2:1 ratio<sup>83,84</sup>, since women apparently live longer<sup>85</sup>, or due to a higher prevalence of cataract in women<sup>86</sup> or because men tolerate a larger visual loss than women before they request surgery<sup>86,87</sup>.

The mean ages of patients enrolled in this study was, 60.95 + 4.44 years in males and 61.97 + 4.82 years in females in the SICS group. In the phaco group, the mean age of male patients was 61.66 + 4.61 years and

females 61.56 + 4.33 years (Table 1); however, these differences were statistically insignificant. The increase in prevalence of cataract with increasing age has been investigated in several epidemiological studies. Kaluzny JJ *et al.*, 1993<sup>88</sup>, in their study on 1075 men and 1247 women with senile cataract, reported a statistically significant difference between the mean ages of the men (63 years) and the women (65 years). Hodge W *et al.*, 1995<sup>36</sup> describes increasing incidence of cataract with increasing age. In the Beaver Dam Study from 1988-90<sup>39,90</sup>, Klein *et al.* noted cataract formation combined with visual acuity decrease to 20/30 or worse in 25% of females and 135 of males aged more than 75 years.

In the frequently-cited Framingham eye study<sup>91</sup>, of the 2675 inhabitants investigated, lenticular opacification occurred in 80% of those aged more than 75 years; in 46%, vision was also decreased to 20/30 or worse. In a study on 5000 inhabitants in Sweden<sup>92</sup>, cataract prevalence was estimated to be 24% for women and 145 for males; inclusion of any previous cataract surgery raised prevalence to 42% in females and 27% in males.

Co-morbid systemic conditions noted in the patients in the two groups were hypertension, diabetes mellitus, hypercholesterolemia and coronary artery disease (Table 2). Overall, co-morbid conditions occurred in 60 (46%) of 130 patients in the SICS group and in 158 (40.5%) of 390

patients in the phacogroup (Table 2); this difference was statistically insignificant. Interestingly, co-morbid conditions occurred significantly more frequently in males (78 [48.4 %] of 161) than in females (140 [39%] of 359) studied ( $X^2$  [d.f.=1] = 4.47 ; P = 0.03) (Table 2). In the present study, hypertension was the most frequently associated co-morbid condition, affecting 57% of SICS individuals and 47% of phaco individuals. (Fig 2). Hypertension and diabetes mellitus were the most frequent co-morbid conditions in females in both groups, affecting 41% and 36%, respectively in the SICS group, and 43% and 35% in the phaco group (Table 2). Interestingly, hypertension was seen significantly more frequently in males (39 [24%] of 161 males) than in females (59 [16.4%] of 359 females) in the current study ( $X^2$  [d.f.=1] = 4.4 ; P=0.036).

Burgess *et al.*,<sup>93</sup> in his study on 136 cataract patients, found systemic hypertension and diabetes mellitus to increase the risk of cataract occurrence. In the study by Behera *et al.*<sup>94</sup> on 1627 individuals (49% males and 51% females), the prevalence of hypertension and diabetes mellitus was 20.59% and 5.95% respectively, with hypertension occurring more frequently in females (24.5% of 832 females) than in males (13.8% of 795 males). However, the prevalence of diabetes that was reported, was nearly equal in both males and females i.e. 4.7% and 4.4% respectively. The



study concluded that cataract cases are usually associated with chronic systemic diseases like hypertension and diabetes.

In the current study, although diabetes mellitus occurred more frequently in females (49 [13.6%] of 359) than in males (17 [10.6%] of 161), this difference was statistically insignificant. Cataracts more frequently in diabetics than in non-diabetics and at a younger age.<sup>39,96</sup>

The most common type of cataract in diabetics including both the SICS and phaco groups was Posterior subcapsular cataract (43.9% [29 out of 66]). The Beaver Dam Eye study<sup>96</sup> that reported similar results. Avraham *et al.*,<sup>97</sup> confirmed the association of PSCC with diabetes ; these authors noted PSCC presence at multiple degrees of glucose bintolerance and hypothesized a possible independent role for non-enzymatic glycosylation. The occurrence of cataract in diabetics may impair the recognition of sight-threatening diabetic retinopathy at an early stage and there exists the risk of deterioration of retinopathy that may go unrecognized. Cataract surgery may also worsen diabetic maculopathy. Although, cataract surgery used to be deferred in diabetic patients until vision was severely disrupted, current day cataract surgical techniques can be done when the patient experiences symptoms and well before a serious impairment of the view of the retinal fundus.

The present study used the LOCS classification to grade cataracts into nuclear cataracts NC1, NC2, NC3, NC4, NC5 and end stage cortical cataracts as mature or hypermature (MC/HMC) cataracts (Table 3). In the SICS group (total 130 patients) three patients (2%) had NC2, 52 (40%) had NC3, 38 (29%) had NC4, eight (6%) had NC5 and 29 (23%) had mature / hypermature cataracts; corresponding percentages in the phaco group (total 390 patients) were 2%, 30%, 34%, 8% and 26%, respectively (Table 3). These differences were not statistically significant ( $\chi^2$  [d.f.=4]=4.8; P=0.31).

In males NC2 cataracts were noted in 0.4 %, NC3 cataracts in 37%, NC4 cataracts in 26%, NC5 cataracts in 6.2% and mature / hypermature cataracts in 30.4%; in female patients, the percentages were 3%, 31%, 35%, 9% and 22%, respectively. Interestingly, these gender differences approached statistical significance ( $\chi^2$  [d.f.=4] = 10.6, P = 0.03 : with Yates' correction,  $\chi^2$  = 8.63, P = 0.07) (Table 3).

According to Kleine *et al.*,<sup>39,90</sup> there was an increasing incidence of nuclear cataract with increase in age. Women were more likely to have nuclear cataract than men, even after age adjustment. Women were more severely affected than men. The Blue mountain eye study<sup>83</sup> done in 2003 was a 5 year follow up study that examined 2335 patients; nuclear cataract followed by cortical cataract was found to be the most frequently occurring

type of lens opacity. Here also, women were found to have a significantly higher incidence of nuclear and cortical cataracts and cataract surgeries. Hormonal factors were cited to explain this higher difference.

In the present study, of the 520 patients who were enrolled, 390 patients underwent manual SICS and 390 underwent cataract extraction by phacoemulsification. The 130 patients who underwent SICS had a post-operative mean visual acuity (decimals) of  $0.58 + 0.02$ , which was significantly higher than the pre-operative mean visual acuity of  $0.07 + 0.01$  (unpaired 't' [d.f.=258] = 21.6 ;  $P < 0.0001$ ) (Table 4). This significant improvement was seen in both males and females in the SICS group (Table 4). Similarly, in the 390 patients who underwent phacoemulsification surgery, the post-operative mean visual acuity ( $0.64 + 0.01$ ) was significantly better than the pre-operative mean visual acuity of  $0.07 + 0.01$  (unpaired 't' [d.f.= 776) = 38.7 ;  $P < 0.0001$ ) (Table 5). This significant improvement was seen in both males and females (Table 5).

Minassian *et al.*<sup>98</sup> and Karki *et al.*,<sup>99</sup> compared extra-capsular cataract extraction (ECCE) and SICS; they found that both were good for community cataract surgery, but that SICS gave a statistically better visual outcome. Phacoemulsification however remains the standard of care for cataract extraction in the western world. Advantages of phacoemulsification include the reduction in morbidity from cataract

surgery by reduction of the incision size, with subsequent faster recovery and decreased risk of complications including endophthalmitis. In addition, the learning curve for this procedure seems to be remarkably small.

Singh *et al.* (2009)<sup>46</sup> sought to elucidate the outcomes of phacoemulsification versus SICS for cataract surgery in immature cataract. In this prospective randomized controlled trial performed on approximately 90 patients with immature senile cataract in each group taken for phacoemulsification or SICS, the groups did not differ significantly in terms of demographic details and in pre-operative vision. More than 66% of individuals in the phaco group and more than 75% of individuals in the SICS group had good visual acuity (6/6-6/18) the day after surgery, the difference again not being of statistical significance. Vision less than <6/60 was considered poor, and this was noted in 6% of individuals in the phaco group and in 1% of individuals in the SICS group. Average visual acuity in the phaco group was  $0.43 \pm 0.27$ , and in the SICS group was  $0.47 \pm 0.24$ . Interestingly, the average surgical time was statistically less in the SICS group as compared to the phacogroup. These authors concluded that SICS with the intraocular placement of polymethmethacrylate lens is a relevant surgery for treatment of immature cataracts in Third World countries.

Meeks *et al.* (2013)<sup>41</sup> reported on a retrospective cohort evaluation of safety and effectiveness of phaco cataract extraction and extracapsular extraction performed by beginning resident surgeons in Texas, USA. Data were collected for cases performed over almost a 6-year period during which, initially the first primary surgeon cases does extracapsular cataract extraction and, later, the first primary surgeon cases performs phacoemulsification. Posterior chamber IOLs were placed in all but two phacoemulsification cases and in four patients in whom extracapsular cataract extraction was performed. These authors concluded that phacoemulsification cataract extraction can be taught safely and effectively to residents with no cataract surgery experience as a primary surgeon.

Rohit C Khanna *et al.*,<sup>100</sup> compared the surgical outcomes of SICS and phacoemulsification performed by ophthalmology trainees. SICS group individuals were significantly older and with worse pre-operative visual acuity; however, BCVA  $\geq$  6/12 was achieved in more than 84% of individuals in both groups.

With reference to mean pre-operative visual acuities, differences between the groups were not significant. The differences in the pre-operative mean visual acuity in the SICS group [0.08 + 0.01] and the Phaco group [0.06 + 0.01] in males with P=0.21 as well as the values of the SICS group [0.07 + 0.01] and the Phaco group [0.07 + 0.01] in females

were not statistically significant. Mean post-operative visual acuity between males in SICS and Phaco groups was not significant ; however, phaco group females had a significantly better mean post-operative visual acuity than SICS group females in the present study (Tables 3, 4).

Gogate *et al.* (2007)<sup>44</sup> compared costs of SICS and phaco surgeries, in a hospital setting in India using standard norms. The participants included 400 patients and four surgeons and it reported that the cost of phacoemulsification cataract surgery was higher due to the cost of the foldable lens used. Moreover, phacoemulsification required additional expenditure to cover the financial depreciation of the instrument and the replacement of faulty or old parts. These authors concluded that SICS is cheaper, equally safe and yields a comparable visual outcome with that of phaco.

Ruit *et al.* (2007)<sup>45</sup> aimed to evaluate the success and visual outcomes following phacoemulsification with that following SICS in 108 individuals with significant cataracts. Both surgical techniques yielded comparable surgical outcomes with few complications. The day after surgery, both groups had almost similar levels of uncorrected visual acuity (UCVA) while a lower degree of corneal oedema occurred in the SICS group. In the six-month follow-up period, almost 90% of the individuals who had undergone SICS exhibited UCVA 20/60 or more, while 98%

exhibited best-corrected visual acuity of 20/60 or more versus 85% of the individuals in the phacoemulsification group with UCVA of 20/60 or more and 98% with BCVA of 20/60 or more; the difference did not yield statistical significance. Interestingly, SICS took a shorter time to perform than did phacoemulsification. These authors concluded that both surgical techniques resulted in good visual recoveries with fewer complications. However, they also opined that since SICS can be performed in a shorter time duration, is less costly and independent of high-end technology unlike phacoemulsification, SICS might be a more relevant option to use to treat advanced cataracts in the developing countries.

The prevalence of cataract surgery as well as factors in association with post-surgical outcomes of vision in migrant Indians living in Singapore was studied by Gupta *et al.* (2013)<sup>48</sup>. A population-based study was conducted on 3, 400 Indian immigrants residing in Singapore. The age- and gender-standardized prevalence of cataract surgery was 9.7% in Singapore resident Indians, while the post-operative visual impairment, as defined by best corrected visual acuity, was found to occur in 10.9% of the eyes studied. These authors concluded that socioeconomic variables and migration did not significantly contribute to the prevalence of cataract surgery, and that diabetic retinopathy was a major cause of postoperative visual impairment in migrant Indians living in Singapore.

Biochemical analysis of the cataractous lenses was done using SDS-PAGE, with the gel being imaged and analyzed with QuantityOne software. The normal lens nuclei constituted Lane N, with most proteins having molecular weight of 12 to 30 kDa. The band intensities of the cataractous lens samples, showed a statistically significant reduction in intensity, which was evident visibly, when compared with the band intensity of normal lens nuclei. With increasing nuclear sclerosis, the staining intensity of bands corresponding to 29 and 20 kDa proteins was found to notably decrease. In mature/hypermature cataractous lens samples, many proteins were found to be missing in the gel, when compared to the pattern in normal lenses. In contrast, the distribution of proteins in NC2 cataractous lens samples was similar to that obtained in samples from normal lenses. The results of the present study conform to the findings by Abbott *et al.*,<sup>21</sup> who describes two-dimensional electrophoresis (2-DE) and mass spectrometric analysis (MS) as important tools in the promising field of proteomics; these are greatly useful in determining the modifications in lenticular crystalline proteins that contribute to cataractogenesis. 2-DE is capable of simultaneously resolving complex mixtures of modified crystallins. These resolved crystallins can be then quantified by image analysis. These perceived differences in band



staining intensity, as assessed by the naked eye, may be subjected to intensity analysis of the SDS-PAGE images using computer software.

To further confirm the protein degradation observed on SDS-PAGE, two dimensional gel electrophoresis (2-DE) was performed in the present study and the gel spots corresponding to  $\alpha$ -crystallin from the different grades of cataractous lenses were selected. These spots were then subjected to MALDI-TOF analysis in order to ascertain the crystallin identification. Varying concentrations of alpha-crystallin was identified in all the lens samples.

# *Summary*

## SUMMARY

The present dissertation studied the clinical and biochemical aspects of senile cataracts in patients presenting with diminished vision due to lenticular opacification at the Institute of Ophthalmology, Joseph Eye Hospital, Tiruchirapalli, Tamilnadu, over a period of 11 months (December 1, 2011 to October 31, 2012). Of the 604 patients enrolled, analysis could be performed for the clinical aspects of the study only on 520 patients.

Patients who were enrolled in the study underwent monocular cataract surgery either by small incision cataract surgery (SICS) (130 patients) or by phacoemulsification (390 patients). Patients in the SICS group and phaco group were, essentially, age-and gender-matched. Hypertension was, by far, the most frequent co-morbid condition in males in both SICS and phaco groups, affecting 57% in the SICS group and 47% in the phaco group. Hypertension was also more common in males and although diabetes mellitus occurred more frequently in females, this difference was not statistically significant. Various grades of nuclear cataract (NC), posterior subcapsular cataract (PSCC) and end-stage cortical cataracts namely mature (MC) and hypermature (HMC) were seen in the patients. These cataracts were graded according to the LOCS classification

III. NC3 (40%) and NC4 (34%) were the most common types of cataracts in the SICS and phaco group respectively. NC 3 (37%) and NC4 (35%) were the most common subtypes among males and females. Interestingly, these gender differences approached statistical significance. PSCC cataracts were noted (in association with nuclear cataracts) in 71 males and 179 females. PSCC 1 cataract was the most common grade of PSC (46.5%) and in females (49.7%).

The mean pre-operative visual acuities showed no significant differences between the SICS and phaco groups. The post-operative mean visual acuity (decimals) in the SICS and phaco groups was  $0.58 \pm 0.02$  and  $0.64 \pm 0.01$ , respectively and it was significantly better than the pre-operative mean visual acuity in both the SICS and phaco groups ( $0.07 \pm 0.01$  and  $0.07 \pm 0.01$ ) respectively. This significant improvement was seen in both males and females. Though there was no post-operative mean visual acuity difference between SICS and phaco in males, such a difference existed in females with better visual acuity in the phaco group. When all patients were considered, the post-operative mean visual acuity in the Phaco group was higher than that in the SICS group, and this difference approached statistical significance with  $P = 0.06$ .

The frequency of surgical and post-operative complications, pre-existing pathology and combined surgery was compared between the SICS and Phaco groups. Combined surgery was needed for 13 (10.0 %) of 130 SICS group patients and 17 (4.4 %) of 390 phaco group patients, this difference was statistically significant ( $P=0.03$ ).

Different degrees of post-operative (PO) visual acuity (Snellen's units) in relation to pre-operative cataract grade were compared in patients who had undergone SICS and in those who had undergone phacoemulsification cataract surgery. Interestingly, significant differences between the SICS and Phaco groups were observed, when PO visual acuities were assessed against pre-operative mature and hypermature cataract grades. In the Phaco group, of 100 individuals who exhibited preoperative mature or hypermature cataracts, 69 (69%) achieved PO visual acuities of 6/6 or 6/9 while 31 (31%) achieved PO visual acuities of 6/12, 6/18 or 6/24. This difference was statistically significant ( $P=0.02$ ).

The biochemical analysis was done after SDS-PAGE, and the gel was imaged and analyzed with Quantity One software. In the normal lens nuclei (Lane N), most proteins were 12 to 30 kDa (Fig 14). In the cataractous lens samples (Lanes S1, S2, S3, S4 and S5), the band intensities at ~20 kDa visibly ( $P < 0.05$ ) showed a gradual reduction in

intensity, when compared with the band intensity of normal lens nuclei. With increasing nuclear sclerosis, the staining intensity of bands corresponding to 29 and 20 kDa proteins was found to notably decrease (Fig 14). In mature/hypermature cataractous lens samples, many proteins were absent in the gel, when compared to the pattern in normal lenses. In contrast, the distribution of proteins in NC2 cataractous lens samples (lane S1) was similar to that obtained in samples from normal lenses (lane N). These perceived differences in band staining intensity, as assessed by the naked eye, were reinforced by intensity analysis of the SDS-PAGE images using computer software. To further confirm the protein degradation observed on SDS-PAGE, two dimensional gel electrophoresis (2-DE) was performed and the gel spots corresponding to  $\alpha$ -crystallin from the different grades of cataractous lenses were selected (Fig. 15). These spots were then subjected to MALDI-TOF analysis in order to ascertain the crystallin identification. Alpha-crystallin was identified in all the lens samples, although the concentration varied.

*Conclusion*

## CONCLUSION

The results of the present study suggest that both SICS and phacoemulsification yield satisfactorily similar visual outcomes. Females undergoing phacoemulsification may achieve better visual outcomes than males. Aside from this, there are no other obvious factors (grade of cataract, associated ocular and systemic comorbid conditions, combined surgery) that would favour choosing one procedure rather than the other. Other considerations such as cost, may influence the final decision. But a study of these influences was outside the scope of the present study. Additional studies are needed on a larger cohort of patients over a longer time duration to identify possible factors that may favour one procedure over the other. Additional biochemical studies on various grades of human cataract may help in identifying factors triggering cataractogenesis; an understanding of these biochemical factors may pave the way for medical management of cataract.



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*Proforma*

## **PROFORMA**

**IP no:**

**Age:**

**Sex:**

**Occupation :**

**Socioeconomic status:**

**Heredity:**

**Personal history:**

h/o dehydrational crisis or malnutrition

h/o alcohol

h/o smoking

**Visual complaints:**

- decreased v/a:
- glare:
- myopic shift

## **Clinical assessment**

**Systemic associations:**

hypertension

diabetes mellitus

hypercholesterolemia

coronary artery disease

allergy

**Ocular associations:**

Glaucoma

High myopia

**Eye to be operated:**

**Pre-operative vision:**

**Type of cataract:**

**Stage :**

**Surgical procedure:**

**Intra- or post-operative complications**

**Post-operative vision:**

**Vision in the other eye:**

**Cataract type if any in the other eye:**

# *Master Chart*



## MASTER CHART - SICS GROUP

IP NO	age	sex	decreased v/a		HTN	DM	hyperchol estremia	CAD	EYE OP	preop vn	pre-op decimal	type of cataract		postop Vn	post dec	
1053001	62	M	6//6	3//60					LE	3//60	0.05	NS3	PSCC3	6//6	1	
1052974	56	M	1//60	1//60					LE	1//60	0.02	NS2	PSCC2	6//6	1	
1053000	57	M	5//60	6//12					RE	5//60	0.08	NS2		6//6	1	
1052976	62	M	3//60	2//60	P			P	LE	2//60	0.04	NS2	PSCC2	6//12	0.5	
1052098	56	M	6//6	6//24	P		P		LE	6//24	0.25	NS2	PSCC1	6//6	1	
1053191	59	M	6//24	6//24					RE	6//24	0.25	NS2		6//9	0.63	
1053544	60	M	6//60	PL+					LE	PL+	0	MC		6//6	1	
1053525	60	M	6//18	6//6					RE	6//18	0.33	NS2		6//6	1	
1053547	63	M	4//60	6//12		P			RE	4//60	0.07	NS3	PSCC1	6//6	1	
1053558	62	M	6//60	6//18					RE	4//60	0.07	NS3		6//6	1	
1053543	62	M	2//60	1//60					LE	1//60	0.02	MC		6//24	0.25	pcr+sfiol
1054270	54	M	1//60	6//9					RE	1//60	0.02	NS2	PSCC3	6//9	0.63	
1054091	56	M	5//60	6//60	P			P	LE	5//60	0.08	NS2	PSCC2	6//6	1	
1054085	59	M	6//12	1//60					LE	1//60	0.02	NS2	PSCC2	6//6	1	
1044739	60	M	6//18	PL+	P	P			LE	PL+	0	MC		6//12	0.5	sk
1027277	64	M	6//12	5//60					LE	5//60	0.08	NS2	PSCC1	6//12	0.5	
1054656	70	M	6//60	NO PL					RE	6//60	0.01	NS2	PSCC1	6//6	1	
1054729	59	M	6//12	6//18					LE	6//18	0.33	NS2	PSCC2	6//6	1	
1054705	57	M	6//18	6//36					LE	6//36	0.17	NS3		6//12	0.5	
1054000	54	M	6//18	2//60	P				LE	2//60	0.04	NS4		6//60	0.1	ARMD
1054704	71	M	6//60	6//12					RE	6//60	0.1		PSCC2	6//12	0.5	
1055353	58	M	5//60	5//60	P	P		P	RE	5//60	0.08	NS2		6//12	0.5	
1055354	59	M	6//12	6//60					LE	6//60	0.1	NS2	PSCC1	6//18	0.33	
1054703	69	M	6//36	PL+	P				LE	PL+	0	MC		5//60	0.8	
1055202	65	M	4//60	2//60					LE	2//60	0.04	NS3	PSCC1	6//18	0.33	
1055355	58	M	6//12	PL+	P				LE	PL+	0	MC		6//24	0.25	
1055466	64	M	2//60	5//60					RE	2//60	0.04	MC		6//18	0.33	
1055425	62	M	6//12	CFC	P				LE	PL+	0	MC		6//12	0.5	
1055499	62	M	6//36	6//60		P			LE	6//60	0.1	NS2		6//12	0.5	
1055500	66	M	6//24	6//12					RE	6//24	0.25	NS2		6//6	1	
1055511	60	M	PL+	6//36					RE	PL+	0	MC		6//9	0.63	
1055501	67	M	PL+	6//36					RE	PL+	0	NS3		6//9	0.63	
1005794	52	M	6//24	6//18					RE	6//24	0.25	NS2		6//9	0.63	
1055788	59	M	5//60	6//18					RE	5//60	0.08	NS3	PSCC	6//9	0.63	
1055663	64	M	2//60	1//60					LE	1//60	0.02	NS3		6//6	1	
1056307	61	M	6//12	5//60					LE	5//60	0.08	NS3		6//18	0.33	dense SK
1056423	64	M	6//12	2//60					LE	2//60	0.04	NS3		6//12	0.5	combined
1043828	67	M	6//24	PL+	P				LE	PL+	0	MC		6//9	0.63	
1056424	57	M	1//60	5//60	P		P		RE	1//60	0.02	HMC		6//9	0.63	pxf
1052980	48	F	1//60	05//60	P			P	LE	1//60	0.02	MC		6//6	1	

1052979	60	F	6//18	1//60				LE	1//60	0.02	NS3	PSCC1	6//6	1	
1052981	58	F	1//60	0.5//60			P	LE	1//60	0.02	NS4		6//6	1	
1052978	70	F	5//60	2//60				LE	2//60	0.04	NS2	PSCC3	6//9	0.63	
1052977	67	F	1//60	4//60	P	P	P	RE	1//60	0.02	NS3	PSCC2	6//9	0.63	
1053151	68	F	PL+	6//6				RE	PL+	0	MC		6//18	0.33	dense sk
1038982	59	F	6//36	4//60				LE	4//60	0.07	NS2	PSCC2	6//9	0.63	
1053179	70	F	6//36	1//60				LE	1//60	0.02	NS3	PSCC	6//9	0.63	
1053178	66	F	6//36	4//60				LE	4//60	0.07	NS3	PSCC	6//6	1	
1053180	63	F	6//36	3//60		P		LE	3//60	0.05	NS2	PSCC2	6//9	0.63	combined
1053141	61	F	6//12	1//60				LE	1//60	0.02	NS4		6//9	0.63	
1053167	65	F	5//60	6//60				RE	5//60	0.08	NS2	PSCC2	6//9	0.63	
1053177	52	F	6//18	PL+		P		LE	PL+	0	MC		6//9	0.63	
1053190	58	F	6//36	6//24				RE	6//36	0.17	NS3	PSCC1	6//6	1	
1053050	56	F	PL+	PL+				RE	PL+	0	HMC		6//9	0.63	
1011070	60	F	5//60	6//9				RE	5//60	0.08	NS2	PSCC2	6//9	0.63	
1033501	61	F	6//9	6//24				LE	6//24	0.25	NS3		6//6	1	
1039633	66	F	6//12	PL+				LE	PL+	0	HMC		6//9	0.63	
1053314	67	F	PL+	1//60				RE	PL+	0	MC		6//9	0.63	
1053555	72	F	6//24	5//60				LE	5//60	0.08	NS4		6//18	0.33	combined
1053556	69	F	3//60	6//12				RE	3//60	0.05	NS4	PSCC2	6//9	0.63	
1053492	65	F	HM	6//6				RE	1//60	0.02	MC		6//6	1	
1053537	54	F	6//36	3//60	P	P		LE	3//60	0.05	NS3	PSCC3	6//9	0.63	
1053538	57	F	CFC	3//60				RE	PL+	0	MC		6//9	0.63	
1053548	58	F	3//60	6//24				RE	3//60	0.05	NS3	PSCC2	6//18	0.33	
1053552	59	F	4//60	6//24				RE	4//60	0.07	NS2	PSCC3	6//9	0.63	
1052653	59	F	PL+	PL+	P		P	LE	6//60	0.1	MC		6//12	0.5	
1053557	61	F	6//9	HM				LE	PL+	0	NS3	PSCC	6//9	0.63	combined
1046112	63	F	3//60	1//60	P			LE	1//60	0.02	NS2	PSCC2	6//6	1	
1053553	63	F	HM	5//60				RE	PL+	0	HMC		5//60	0.08	dense sk
1054154	61	F	2//60	PL+				LE	PL+	0	MC		6//60	0.1	brvo
1054232	60	F	5//60	6//24		P		RE	5//60	0.08	NS2		6//9	0.63	
1054174	67	F	6//18	6//12				RE	6//18	0.33	NS2	PSCC1	6//9	0.63	
1054230	66	F	6//18	6//24				LE	6//24	0.25	NS2	PSCC1	6//9	0.63	
1054231	70	F	3//60	6//12	P			RE	3//60	0.05	NS2	PSCC1	6//9	0.63	
1054229	49	F	2//60	2//60		P		RE	2//60	0.04	NS3		6//12	0.5	
1054234	65	F	5//60	5//60				LE	5//60	0.08	NS1	PSCC3	6//9	0.63	
1054152	61	F	6//60	6//9				RE	6//60	0.1	NS2	PSCC1	6//9	0.63	
1054233	60	F	6//18	6//12				RE	6//18	0.33	NS2		6//9	0.63	
1054359	65	F	6//36	3//60				LE	3//60	0.05	NS3	PSCC2	6//9	0.63	
1052128	62	F	6//18	6//12				RE	6//18	0.33	NS2		6//9	0.63	
1054113	60	F	5//60	6//12	P	P		RE	5//60	0.08	NS2	PSCC2	6//9	0.63	
1054543	66	F	6//60	3//60				LE	3//60	0.05	NS3	PSCC	6//24	0.25	combined
1054538	61	F	5//60	HM				LE	1//60	0.02	NS3	PSCC1	6//9	0.63	
1054536	56	F	6//36	PL+				LE	PL+	0	MC		6//12	0.5	
1054342	59	F	2//60	NO PL				RE	2//60	0.04	NS2	PSCC1	6//6	1	

1054532	67	F	6//12	6//60				LE	6//60	0.1	NS1	PSCC2	6//12	0.5	
1054417	65	F	HM	6//12	P	P		RE	1//60	0.02	MC		6//9	0.63	
1054659	68	F	CFC	6//18				RE	PL+	0	NS3	PSCC2	6//36	0.17	
1055143	59	F	6//24	6//36		P	P	LE	6//36	0.17	NS2		6//18	0.33	
1054744	73	F	6//18	6//24				LE	6//24	0.25	NS2	PSCC1	6//6	1	
1055235	69	F	HM	PL+				LE	PL+	0	MC		6//12	0.5	
1055230	69	F	6//12	5//60				LE	5//60	0.08	NS2	PSCC1	6//24	0.25	dense sk
1055232	59	F	6//36	6//12				RE	6//36	0.17	NS2	PSCC2	6//9	0.63	
1055288	58	F	6//9	6//18				LE	6//18	0.33	NS1	PSCC2	6//9	0.63	
1055242	60	F	6//18	6//36				LE	6//36	0.17	NS2		6//12	0.5	
1055204	60	F	6//60	6//36	P		P	RE	6//60	0.1	NS2	PSCC2	6//36	0.17	brvo
1036572	62	F	6//18	3//60		P		LE	3//60	0.05	NS3		6//9	0.63	
1055356	59	F	3//60	6//60				RE	3//60	0.05	NS3		6//18	0.33	
1055357	64	F	6//60	3//60				LE	3//60	0.05	NS2		6//36	0.17	
1055359	63	F	6//60	6//60				LE	6//60	0.1	NS3		6//12	0.25	
1055360	58	F	5//60	6//18				RE	5//60	0.08	NS2		6//9	0.33	
1055358	58	F	3//60	6//12	P			RE	3//60	0.05	NS4		6//12	0.5	
1055223	63	F	6//18	PL+				LE	PL+	0	MC		6//24	0.25	
1055274	57	F	PL+	6//24				RE	PL+	0	NS4		1//60	0.02	
1055361	74	F	5//60	5//60				RE	5//60	0.08	NS2	PSCC	6//9	0.63	
1055506	61	F	6//24	1//60	P			LE	1//60	0.02	TC		6//12	0.25	
1055498	65	F	1//60	2//60	P			RE	1//60	0.02	NS3		6//12	0.25	
1055513	62	F	6//36	1//60				LE	1//60	0.02	NS2		6//18	0.33	
1055512	60	F	6//24	3//60				LE	3//60	0.05	NS3		6//9	0.63	
1055502	63	F	PL+	0.5//60				RE	PL+	0	MC		6//12	0.5	
1055641	59	F	5//60	6//24				RE	5//60	0.08	NS2	PSCC	6//24	0.25	
1055659	57	F	0.5//60	1//60				RE	1//60	0.02	NS2		6//24	0.25	
1055572	59	F	6//60	6//9				RE	6//60	0.1	NS3		6//12	0.5	drusen
1055362	58	F	5//60	5//60		P		RE	5//60	0.08	NS2		3//60	0.05	
1055793	69	F	5//60	PL+				LE	PL+	0	MC		6//6	1	
1055792	64	F	6//12	CFC				LE	PL+	0	NS4	PSCC1	6//9	0.63	
1055844	59	F	6//12	5//60	P			LE	5//60	0.08	NS3		6//9	0.63	
1055795	60	F	6//24	1//60	P	P		LE	1//60	0.02	NS3	PSCC	5//60	0.08	VMT
1055790	62	F	6//60	PL+				LE	PL+	0	NS3		6//18	0.33	
1055789	67	F	5//60	5//60				RE	5//60	0.08	NS2		6//6	1	
1056421	63	F	1//60	6//36	P		P	RE	1//60	0.02	NS3		6//12	0.5	
1056313	66	F	6//36	1//60				LE	1//60	0.02	NS2		6//12	0.5	
1056347	59	F	6//36	2//60				LE	2//60	0.04	NS2		6//9	0.63	
1056309	58	F	6//36	5//60				LE	5//60	0.08	NS2		6//18	0.33	COMBINED
1056349	59	F	6//36	2//60				LE	2//60	0.04	NS3	PSCC	6//9	0.63	
1056317	60	F	5//60	3//60	P	P	P	LE	3//60	0.05	NS3	PSCC	6//9	0.63	
1056411	62	F	6//18	6//60				LE	6//60	0.1	NS2		6//12	0.5	
1056413	60	F	PL+	1//60				RE	3//60	0.05	MC		6//12	0.5	
1056362	62	F	3//60	6//24				RE	3//60	0.05	NS3		6//12	0.5	combined
1056420	57	F	3//60	6//60		P		RE	3//60	0.05	NS3		6//9	0.63	

## MASTER CHART - PHACO GROUP

MR.NO.	AGE	SEX	RE	LE	HTN	DM	CHOL	CAD	EYE Operated	pre-op	snellen	grade	grade	post-op V/A	snellen	complic	
1001422	60	M	2//60	1//60					LE	0.02	1//60	NS3	PSCC3	1	6//6		
1001538	56	M	3//60	6//12					RE	0.05	3//60	NS2	PSCC2	1	6//6		
1001547	57	M	2//60	2//60					RE	0.04	2//60	NS2		1	6//6		
1001588	62	M	5//60	5//60	P			p	LE	0.08	5//60	NS2	PSCC2	0.5	6//12		
1001755	56	M	6//12	5//60	P	P	P		LE	0.08	5//60	NS2	PSCC1	1	6//6		
1001925	59	M	6//60	3//60					LE	0.05	3//60	NS2		0.63	6//9		
1002106	61	M	6//24	6//36					LE	0.17	6//36	NS3		1	6//6		
1002112	64	M	5//60	6//60					RE	0.08	5//60	NS2	PSCC2	1	6//6		
1002113	63	M	6//60	1//60					LE	0.02	1//60	NS3	PSCC1	1	6//6		
1002114	62	M	HM	PL+					LE	0	PL+	HMC		1	6//6		
1002115	62	M	6//60	6//36					RE	0.1	6//60	NS4		0.25	6//24	PCR+SFIOL	
1002116	52	M	6//36	PL+					LE	0	PL+	MC		0.63	6//9		
1002118	56	M	6//18	PL+	P	P		p	LE	0	PL+	MC		1	6//6		
1002119	65	M	4//60	2//60					LE	0.04	2//60	NS3	PSCC2	1	6//6		
1020699	59	M	6//12	PL+	P				LE	0	PL+	MC		0.5	6//12	pxf	
1020757	64	M	2//60	5//60					RE	0.04	2//60	NS3	PSCC2	0.5	6//12		
1020770	70	M	PL+	0.5//60					RE	0	PL+	MC		1	6//6		
1020784	59	M	5//60	6//24					RE	0.08	5//60	NS2	PSCC2	1	6//6		
1020872	57	M	0.5//60	1//60					RE	0.02	1//60	NS3	PSCC2	0.5	6//12		
1020881	54	M	6//60	6//9	P	P			RE	0.1	6//60	NS4		0.1	6//60	SFIOL	
1020901	67	M	5//60	5//60					RE	0.08	5//60	NS3	PSCC1	0.5	6//12		
1020929	62	M	6//12	5//60	P		P	p	LE	0.08	5//60	NS2	PSCC1	0.5	6//12		
1020936	65	M	6//24	6//60					LE	0.1	6//60	NS3	PSCC1	0.33	6//18	combined	
1020981	58	M	6//36	2//60	P	P			LE	0.04	2//60	NS2	PSCC3	0.5	6//12		
1021143	59	M	6//36	5//60					LE	0.08	5//60	NS2	PSCC1	0.33	6//18	Drusen at fovea	
1021163	69	M	6//24	PL+	P				LE	0	PL+	MC		0.08	5//60		
1021164	65	M	PL+	1//60					RE	0	PL+	HMC		0.33	6//18		
1021185	58	M	3//60	6//60	P				RE	0.05	3//60	MC		0.25	6//24	combined	
1021203	64	M	6//6	3//60					LE	0.05	3//60	MC		0.33	6//18		
1021205	62	M	6//18	1//60	P				LE	0.02	1//60	MC		0.5	6//12		
1021231	62	M	1//60	0.5//60					LE	0.02	0.5//60	NS4	PSCC1	0.5	6//12		
1021243	66	M	5//60	2//60					LE	0.04	2//60	NS3	PSCC1	1	6//6		
1021258	60	M	1//60	4//60					RE	0.02	1//60	MC		0.63	6//9		
1021259	64	M	6//12	1//60					LE	0.02	1//60	MC		0.63	6//9		
1021260	52	M	6//24	6//24					RE	0.25	6//24	NS2		0.63	6//9		
1021263	59	M	PL+	PL+					RE	0	PL+	HMC		0.63	6//9		
1021265	64	M	6//12	PL+					LE	0	PL+	HMC		0.04	2//60	dense SK	
1021266	61	M	HM	6//6					RE	0.02	HM	NS4		1	6//6		
1021267	64	M	6//18	6//6					RE	0.33	6//18	NS3		1	6//6		
1021268	67	M	4//60	6//12	P	P			RE	0.07	4//60	MC		0.63	6//9		
1021274	57	M	6//60	6//18	P		P		RE	0.1	6//60	NS2	PSCC1	0.63	6//9		
1021275	58	M	2//60	PL+					LE	0	PL+	HMC		1	6//6		
1021276	62	M	2//60	2//60					RE	0.04	2//60	NS3	PSCC1	1	6//6		
1021277	59	M	5//60	5//60	P				LE	0.08	5//60	NS2	PSCC2	1	6//6		
1021278	61	M	6//60	6//9					RE	0.1	6//60	NS4		0.63	6//9	PC RENT	
1021279	70	M	6//18	PL+		P		p	LE	0	PL+	MC		1	6//6		
1021315	61	M	5//60	HM					LE	0.02	HM	HMC		0.63	6//9		

1021319	56	M	4//60	6//24					RE	0.07	4//60	NS2	PSCC3	1	6//6		
1021363	67	M	6//60	1//60					LE	0.05	1//60	NS3	PSCC1	0.63	6//9		
1021368	65	M	HM	PL+					LE	0	PL+	HMC		0.33	6//18		
1021373	70	M	6//12	5//60	P			P	LE	0.08	5//60	NS3	PSCC2	1	6//6		
1021390	71	M	6//18	3//60					LE	0.05	3//60	MC		0.5	6//12	BRVO	
1021400	60	M	6//18	PL+	P				LE	0	PL+	MC		0.63	6//9		
1021401	60	M	4//60	2//60					LE	0.04	2//60	NS3	PSCC2	0.5	6//12		
1021402	62	M	PL+	6//24					RE	0	PL+	HMC		0.63	6//9		
1021404	58	M	5//60	5//60					LE	0.08	5//60	NS3	PSCC2	0.63	6//9		
1021405	74	M	5//60	6//24					RE	0.08	5//60	NS2	PSCC2	0.5	6//12		
1021406	64	M	0.5//60	1//60					LE	0.02	0.5//60	NS3	PSCC1	1	6//6		
1021416	62	M	6//60	6//9	P				RE	0.1	6//60	NS4		0.5	6//12	SFIOL	
	62	M	PL+	6//36					RE	0	PL+	MC		0.63	6//9		
1022065	60	M	5//60	PL+		P	P		LE	0	PL+	MC		1	6//6		
1022150	62	M	6//60	3//60					LE	0.05	3//60	NS3	PSCC2	0.17	6//36		
1022171	60	M	5//60	HM					LE	0.02	HM	MC		0.5	6//12		
1022349	59	M	CFC	6//18					RE	0.02	CFC	MC		0.5	6//12		
1022367	52	M	6//60	6//12					RE	0.1	6//60	NS3	PSCC1	0.17	6//36		
1022465	67	M	HM	PL+					LE	0	PL+	MC		0.17	6//36	BRVO	
1022478	66	M	6//36	6//12					RE	0.33	6//36	NS3		0.63	6//9		
1022492	59	M	6//18	6//36					LE	0.33	6//36	NS1	PSCC1	0.08	5//60		
1022506	59	M	6//18	3//60	P				LE	0.05	3//60	NS2	PSCC3	0.33	6//18		
1022507	60	M	3//60	6//60					RE	0.05	3//60	NS2	PSCC2	0.1	6//60		
1022509	62	M	5//60	5//60					RE	0.08	5//60	NS2	PSCC1	0.25	6//24		
1022515	64	M	6//12	6//60					LE	0.1	6//60	NS3	PSCC1	0.63	6//9		
1022517	67	M	6//60	3//60	P			p	RE	0.1	6//60	NS2	PSCC1	0.33	6//18		
1022565	67	M	5//60	5//60		P			RE	0.08	5//60	NS2	PSCC1	0.5	6//12		
1022577	59	M	PL+	6//36					RE	0	PL+	MC		0.17	6//36		
1022578	70	M	6//24	1//60	P	P			LE	0.02	1//60	NS3	PSCC2	1	6//6		
1022691	61	M	6//24	3//60					LE	0.05	3//60	NS2	PSCC2	0.63	6//9		
1022710	58	M	6//60	6//9					RE	0.1	6//60	NS2	PSCC2	0.04	2//60		
1022749	67	M	4//60	6//18	P			p	RE	0.07	4//60	NS4		0.33	6//18	ARMD	
1022754	72	M	5//60	6//18	P		P	p	RE	0.08	5//60	NS3	PSCC1	0.63	6//9		
1022755	69	M	6//24	1//60					LE	0.02	1//60	MC		0.63	6//9		
1022844	60	M	6//36	1//60					LE	0.02	1//60	NS3	PSCC1	1	6//6		
1022905	59	M	5//60	3//60					LE	0.05	3//60	NS2	PSCC3	1	6//6		
1022917	61	M	6//18	6//60	P				LE	0.1	6//60	NS2	PSCC2	1	6//6		
1022926	63	M	6//12	2//60					LE	0.04	2//60	MC		0.63	6//9	PC RENT	
1023075	49	M	6//60	PL+		P			LE	0	PL+	MC		0.63	6//9		
1023223	54	M	4//60	6//12					RE	0.07	4//60	MC		0.63	6//9		
1023716	59	M	2//60	2//60					RE	0.07	4//60	NS2	PSCC1	0.5	6//12		
1023774	65	M	6//60	6//9					RE	0.1	6//60	NS3	PSCC1	1	6//6		
1023806	70	M	1//60	6//9					RE	0.02	1//60	NS3	PSCC3	0.5	6//12		
1023818	59	M	5//60	6//60	P		P		RE	0.08	5//60	MC		0.33	6//18		
1025283	59	M	6//12	5//60					LE	0.08	5//60	MC		0.5	6//12	BRVO	
1025396	60	M	6//60	NO PL	P				RE	0.1	6//60	NS2	PSCC2	0.5	6//12		
1025447	62	M	6//12	6//18					LE	0.33	6//18	NS2	PSCC2	0.08	5//60		
1025449	59	M	6//18	6//36					RE	0.17	6//36	NS2		0.25	6//24		
1025521	59	M	CFC	6//18					RE	0.02	CFC	NS3	PSCC2	0.1	6//60		
1025524	63	M	6//24	6//36					LE	0.17	6//36	NS2		0.63	6//9		
1025763	59	M	6//60	6//36					LE	0.1	6//60	NS2	PSCC2	0.63	6//9		
1025764	57	M	6//18	3//60					LE	0.05	3//60	NS3		0.5	6//12		

1025766	54	M	3//60	6//60	P				RE	0.05	3//60	NS4		0.25	6//24	SFIOL	
1025774	68	M	5//60	5//60					RE	0.08	5//60	NS3	PSCC2	0.25	6//24		
1025932	59	M	6//60	3//60		P	P		LE	0.05	3//60	NS2		0.63	6//9		
1025986	65	M	3//60	6//12					LE	0.05	3//60	NS3	PSCC2	0.63	6//9		
1026012	69	M	6//36	PL+					LE	0	PL+	MC		0.63	6//9		
1026135	59	M	6//24	6//12					RE	0.05	6//24	NS2	PSCC1	1	6//6		
1026137	64	M	PL+	6//36					RE	0	PL+	NS2		0.63	6//9		
1026145	63	M	PL+	0.5//60					RE	0	PL+	MC		1	6//6		
1026152	57	M	0.5//60	1//60					RE	0.02	1//60	NS4		0.63	6//9		
1026226	74	M	PL+	6//36					RE	0	PL+	NS2	PSCC	1	6//6		
1026235	62	M	5//60	PL+	P				LE	0	PL+	MC		1	6//6		
1026250	62	M	6//12	CFC					LE	0.02	CFC	NS2		1	6//6		
1026271	66	M	6//12	5//60					LE	0.08	5//60	NS2		1	6//6		
1026274	60	M	6//24	6//18					RE	0.25	6//24	MC		1	6//6		
1026276	61	M	5//60	6//18	P				RE	0.08	5//60	TC		0.63	6//9		
1026139	58	M	6//36	5//60		P			LE	0.08	5//60	NS2		0.5	6//12	combined	
1026276	61	M	5//60	6//9					RE	0.08	5//60	NS3		0.63	6//9		
1026279	59	M	6//9	6//24					LE	0.25	6//24	NS2		0.63	6//9		
1026285	58	M	6//12	PL+					LE	0	PL+	NS2		1	6//6		
1026287	59	M	PL+	1//60					RE	0	PL+	NS3	PSCC	0.63	6//9		
1026288	60	M	6//24	5//60	P	P	p		LE	0.08	5//60	NS3	PSCC	0.63	6//9		
1026135	59	M	6//24	6//12					RE	0.05	6//24	NS2	PSCC1	1	6//6		
1026137	64	M	PL+	6//36					RE	0	PL+	NS2		0.63	6//9		
1001418	59	F	3//60	5//60	P				RE	0.05	3//60	NS4	PSCC1	1	6//6		
1001424	60	F	2//60	PL+					LE	0	PL+	NS3	PSCC1	1	6//6		
1001435	58	F	5//60	6//24			P		RE	0.08	5//60	NS4		1	6//6		
1001470	72	F	6//18	6//12					RE	0.1	6//60	NS2	PSCC3	0.63	6//9		
1001528	66	F	6//18	6//24	P	P	P		LE	0.25	6//24	NS3	PSCC2	0.63	6//9		
1001664	63	F	6//60	6//9					RE	0.1	6//60	NS3	PSCC	0.33	6//18	PC RENT	PXF
1001664	59	F	1//60	6//9					RE	0.02	1//60	NS2	PSCC2	0.63	6//9		
1001679	70	F	5//60	6//60					RE	0.08	5//60	NS3	PSCC	0.63	6//9		
1001704	66	F	6//12	1//60					LE	0.02	1//60	NS3	PSCC	1	6//6		
1001718	63	F	6//18	6//60		P			LE	0.1	6//60	NS2	PSCC2	0.63	6//9		
1001719	61	F	6//18	PL+					LE	0	PL+	NS4		0.63	6//9		
1001728	65	F	6//36	3//60					LE	0.05	3//60	NS2	PSCC2	0.63	6//9		
1001784	52	F	6//18	PL+	P	P		P	LE	0	PL+	MC		0.63	6//9		
1001924	58	F	5//60	6//12					RE	0.08	5//60	NS3	PSCC1	1	6//6		
1001944	56	F	5//60	HM					LE	0.02	HM	MC		0.63	6//9		
1001957	60	F	6//36	PL+					LE	0	PL+	NS3	PSCC2	0.63	6//9		
1001967	61	F	2//60	NO PL					RE	0.04	2//60	NS3	PSCC1	1	6//6	POAG	Combined
1001969	66	F	6//12	6//60					LE	0.1	6//60	NS3		0.63	6//9		
1001973	67	F	HM	6//12					RE	0.02	HM	MC		0.63	6//9		
1001957	58	F	6//60	NO PL					RE	0.1	6//60	NS4		0.33	6//18		
1001987	69	F	6//12	6//36					LE	0.17	6//36	NS2	PSCC1	0.63	6//9		
1001994	65	F	6//18	6//36					LE	0.17	6//36	NS3		1	6//6		
1002008	54	F	6//18	2//60	P	P		P	LE	0.04	2//60	NS3	PSCC3	0.63	6//9		
1002018	57	F	CFC	6//18					RE	0.02	CFC	MC		0.63	6//9		
1002096	58	F	6//60	6//12					RE	0.1	6//60	NS3	PSCC1	0.33	6//18		
1002111	59	F	6//18	5//60					LE	0.08	5//60	NS2	PSCC2	0.63	6//9		
1002121	59	F	6//12	5//60	P			P	LE	0.08	5//60	MC		0.5	6//12		
1002137	61	F	6//36	6//12					RE	0.17	6//36	NS3		0.63	6//9	COMBINED	POAG
1002139	63	F	6//9	6//18	P				LE	0.33	6//18	NS2		1	6//6		

1002143	63	F	6//18	6//36					LE	0.17	6//36	NS3		0.08	5//60	DENSE SK	
1002173	61	F	6//18	3//60					LE	0.05	3//60	MC		0.1	6//60	BRVO	
1002180	60	F	3//60	6//60			P		RE	0.05	3//60	MC		0.63	6//9		
1002197	67	F	5//60	5//60					RE	0.08	5//60	NS2	PSCC3	0.63	6//9		
1002187	66	F	6//12	6//60					LE	0.1	6//60	NS2	PSCC1	0.63	6//9		
1002221	70	F	6//60	3//60	P			P	LE	0.05	3//60	NS2	PSCC2	0.63	6//9		
1002269	48	F	6//60	6//60			P		RE	0.1	6//60	NS3		0.5	6//12		
1002274	65	F	5//60	6//18					RE	0.08	5//60	NS3	PSCC2	0.63	6//9		
1002275	61	F	3//60	6//12					RE	0.05	3//60	NS2	PSCC1	0.63	6//9		
1002341	60	F	PL+	6//24					RE	0	PL+	MC		0.63	6//9		
1002354	65	F	5//60	5//60					LE	0.08	5//60	NS3	PSCC2	0.63	6//9		
1002366	62	F	6//12	CFC					LE	0.02	CFC	MC		0.63	6//9		
1002368	60	F	6//36	6//60	P		P		LE	0.1	6//60	NS2	PSCC2	0.63	6//9		
1002373	65	F	6//24	6//12					RE	0.25	6//24	NS3		0.63	6//9	COMBINED	
1002379	61	F	PL+	6//36					RE	0	PL+	MC		0.63	6//9		
1002385	56	F	6//24	1//60					LE	0.02	1//60	NS4	PSCC1	0.5	6//12		
1002410	59	F	1//60	2//60					RE	0.02	1//60	NS3	PSCC2	1	6//6		
1002442	67	F	6//36	1//60					LE	0.02	1//60	NS2	PSCC2	0.5	6//12		
1002476	65	F	6//24	3//60	P		P		LE	0.05	3//60	NS3	PSCC1	0.63	6//9		
1002566	68	F	PL+	6//36					RE	0	PL+	MC		0.17	6//36	SFIOL	PXF
1002640	59	F	5//60	PL+			P	P	LE	0	PL+	MC		0.33	6//18		
1002650	57	F	6//12	CFC					LE	0.02	CFC	NS4	PSCC2	1	6//6		
1002660	59	F	5//60	6//18					RE	0.08	5//60	NS3	PSCC1	0.5	6//12		
1002703	54	F	6//24	1//60					LE	0.1	1//60	NS3	PSCC1	0.25	6//24		
1002717	59	F	6//60	PL+					LE	0	MC			0.63	6//9		
1002724	58	F	2//60	1//60					LE	0.02	1//60	NS3	PSCC2	0.63	6//9		
1002752	60	F	5//60	5//60					LE	0.08	5//60	NS3	PSCC2	0.5	6//12		
1002773	60	F	1//60	6//36	P				RE	0.02	1//60	NS3	PSCC2	0.17	6//36	DENSE SK	
1002778	62	F	6//36	1//60			P		LE	0.02	1//60	NS3		0.63	6//9		
1002855	59	F	6//12	5//60					LE	0.08	5//60	NS3	PSCC1	0.33	6//18		
1002944	64	F	6//36	2//60					LE	0.04	2//60	NS2	PSCC2	0.17	6//36		
1002950	63	F	5//60	3//60					LE	0.05	3//60	NS3	PSCC2	0.5	6//12		
1003030	58	F	6//18	6//60					LE	0.1	6//60	NS2	PSCC1	0.63	6//9		
1003060	58	F	6//12	2//60	P				LE	0.04	2//60	NS4		0.5	6//12		
1003081	63	F	1//60	5//60					RE	0.02	1//60	MC		0.25	6//24	DENSE SK	
1003099	57	F	3//60	6//24					RE	0.05	3//60	NS3	PSCC1	0.1	6//60		
1003146	57	F	1//60	05//60					RE	0.02	1//60	NS2	PSCC3	0.63	6//9		
	61	F	1//60	1//60					LE	0.02	1//60	MC		1	6//6		
1011483	65	F	5//60	6//12	P				RE	0.08	5//60	NS3	PSCC1	1	6//6		
1011522	62	F	3//60	2//60					LE	0.04	2//60	NS4		1	6//6		
1011528	60	F	PL+	6//6					RE	0	PL+	MC		0.63	6//9		
1011529	63	F	6//36	4//60					LE	0.07	4//60	MC		0.5	6//12		
1011530	59	F	6//36	1//60					0	0.02	1//60	NS3	PSCC	0.25	6//24		
1011541	57	F	6//36	4//60					LE	0.07	4//60	NS3	PSCC2	0.25	6//24		
1011650	59	F	6//36	3//60					LE	0.05	3//60	NS3	PSCC2	0.5	6//12		
1011690	58	F	5//60	6//60			P		RE	0.08	5//60	PSCC2		0.17	6//36		
1011691	69	F	6//6	6//24					LE	0.25	6//24	NS2		1	6//6		
1011694	64	F	6//18	PL+					LE	0	PL+	MC		0.63	6//9		
1011711	59	F	6//36	6//24	P				RE	0.17	6//36	NS3		0.63	6//9		
1011819	63	F	5//60	6//9	P		P		RE	0.08	5//60	NS3	PSCC	0.02	CFC		
1011860	62	F	6//9	6//24					LE	0.25	6//24	NS3		0.33	6//18		
1011879	65	F	PL+	1//60					RE	0	PL+	MC		1	6//6		

1011889	63	F	6//24	5//60	P		P	P	LE	0.08	5//60	NS3	PSCC2	1	6//6		
1011898	66	F	3//60	6//12					RE	0.05	3//60	NS4	PSCC	1	6//6		
1011924	59	F	6//36	3//60					LE	0.05	3//60	NS2	PSCC2	0.63	6//9		
1011925	58	F	CFC	3//60					RE	0.02	CFC	MC		0.33	6//18	COMBINED	
1011932	59	F	3//60	6//24					RE	0.05	3//60	NS3	PSCC	0.63	6//9		
1011933	60	F	6//60	PL+		P			LE	0	PL+	MC		0.63	6//9		
1011934	62	F	4//60	6//24					RE	0.07	4//60	NS2	PSCC2	1	6//6		
1011948	60	F	PL+	PL+					RE	0	PL+	MC		1	6//6		
1011949	62	F	6//9	HM					LE	0.02	HM	NS4		1	6//6		
1011952	57	F	3//60	1//60		P			LE	0.02	1//60	NS3	PSCC1	0.63	6//9		
1011958	54	F	HM	5//60					RE	0.02	HM	MC		1	6//6		
1011969	57	F	2//60	1//60					LE	0.02	1//60	NS3	PSCC3	1	6//6		
1011973	60	F	5//60	6//24			P		RE	0.08	5//60	NS4		1	6//6		
1011974	59	F	6//18	6//12					RE	0.33	6//18	NS2		0.63	6//9		
1011975	60	F	6//18	6//24	P	P	P		LE	0.25	6//24	NS3		1	6//6		
1011975	63	F	3//60	6//12					RE	0.05	3//60	NS2	PSCC2	1	6//6		
1011991	63	F	1//60	6//9					RE	0.02	1//60	NS3	PSCC2	0.63	6//9		
1011992	63	F	5//60	6//60					RE	0.08	5//60	NS3	PSCC1	1	6//6		
1011993	62	F	6//12	1//60					LE	0.02	1//60	NS3	PSCC1	1	6//6		
1011994	61	F	6//18	3//60		P			LE	0.05	3//60	NS3	PSCC2	0.5	6//12		
1011996	60	F	6//18	PL+					LE	0	PL+	MC		0.33	6//18		
1011997	67	F	6//36	3//60					LE	0.05	3//60	NS2	PSCC2	0.63	6//9		
1012009	66	F	6//12	5//60	P		P		LE	0.08	5//60	NS2	PSCC1	0.63	6//9		
1012014	49	F	5//60	6//12					RE	0.08	5//60	NS3	PSCC1	0.63	6//9		
1012015	65	F	6//60	3//60					LE	0.05	3//60	NS2	PSCC3	0.63	6//9		
1012021	54	F	6//36	PL+					LE	0	PL+	MC		1	6//6		
1012078	56	F	2//60	6//24					RE	0.04	2//60	NS3	PSCC2	0.63	6//9		
1012113	59	F	6//12	6//60					LE	0.1	6//60	NS3		1	6//6		
1012114	60	F	HM	6//12					RE	0.02	HM	MC		0.63	6//9		
1012116	60	F	6//60	6//36					RE	0.1	6//60	NS4		0.63	6//9		
1012148	65	F	6//12	6//18					LE	0.33	6//18	NS2		0.63	6//9		
1012149	64	F	6//18	6//36					LE	0.17	6//36	NS3		1	6//6		
1012158	62	F	6//18	2//60	P	P			LE	0.04	2//60	NS3	PSCC3	0.63	6//9		
1012167	60	F	CFC	6//18					RE	0.02	CFC	MC		0.63	6//9		
1012168	66	F	6//60	6//12					RE	0.1	6//60	NS3		0.33	6//18		
1012172	61	F	6//24	6//36					LE	0.17	6//36	NS3		0.63	6//9		
1012188	59	F	5//60	6//60					RE	0.08	5//60	NS2	PSCC2	0.63	6//9		
1012243	59	F	6//36	6//12					RE	0.17	6//36	NS3		0.63	6//9	COMBINED	
1012261	57	F	6//9	6//18	P				LE	0.33	6//18	NS2		1	6//6		
1012262	54	F	6//18	6//36					LE	0.17	6//36	NS3		1	6//6	DENSE SK	
1012270	68	F	6//60	6//36					RE	0.1	6//60	NS3		1	6//6	PCR+SFIOL	
1012296	59	F	3//60	6//60		P			RE	0.05	3//60	NS2	PSCC3	0.63	6//9		
1012304	73	F	5//60	5//60					RE	0.08	5//60	NS2	PSCC1	1	6//6		
1012305	62	F	6//12	6//60					LE	0.1	6//60	NS2	PSCC1	0.08	5//60		
1012306	65	F	6//60	3//60	P				LE	0.05	3//60	NS3	PSCC1	0.25	6//24		
1012306	69	F	6//60	6//60		P			RE	0.1	6//60	NS3		0.1	6//60		
1012307	69	F	5//60	6//18					RE	0.08	5//60	NS3	PSCC1	0.63	6//9		
1012340	59	F	3//60	6//12					RE	0.05	3//60	NS2	PSCC2	0.63	6//9		
1012345	58	F	6//36	PL+					LE	0	PL+	MC		0.63	6//9		
1012396	59	F	6//12	PL+					LE	0	PL+	MC		0.63	6//9		
1012422	59	F	2//60	5//60					RE	0.04	2//60	NS3	PSCC1	1	6//6		
1012448	63	F	6//36	6//60		P			LE	0.1	6//60	NS2	PSCC2	0.63	6//9		



1012451	58	F	6//24	6//12					RE	0.25	6//24	NS3	PSCC	0.5	6//12	COMBINED	
1012454	58	F	PL+	6//36					RE	0	PL+	MC		0.63	6//9		
1012457	69	F	6//24	1//60					LE	0.02	1//60	MC		0.5	6//12		
1012479	63	F	1//60	2//60					RE	0.02	1//60	NS3	PSCC2	0.63	6//9		
1012565	65	F	6//36	1//60					LE	0.02	1//60	NS4	PSCC2	0.63	6//9		
1012572	57	F	6//24	3//60	P	P			LE	0.05	3//60	MC		0.63	6//9		
1012667	58	F	PL+	0.5//60					RE	0	PL+	MC		0.63	6//9		
1022036	66	F	5//60	5//60					RE	0.08	5//60		PSCC2	1	6//6		
1022121	61	F	6//12	CFC					LE	0.02	CFC	MC		0.5	6//12		
1022142	65	F	6//12	5//60	P		P		LE	0.08	5//60	NS2	PSCC1	0.1	6//60		
1022254	63	F	6//36	PL+					LE	0	PL+	MC		0.33	6//18		
1022296	59	F	2//60	1//60					LE	0.02	1//60	MC		1	6//6		
1022319	57	F	6//12	6//60					LE	0.1	6//60	NS1	PSCC2	0.5	6//12		
1022330	59	F	HM	6//12					RE	0.02	HM	NS2	PSCC3	0.33	6//18		
1022337	67	F	6//60	NO PL	P				RE	0.1	6//60	NS2	PSCC1	0.5	6//12		
1022341	58	F	6//12	6//60			P		LE	0.1	6//60	NS3		0.25	6//24	DENSE SK	
1022343	69	F	6//18	6//60					LE	0.1	6//60	NS3	PSCC1	0.63	6//9		
1022346	64	F	6//18	2//60	P	P			LE	0.02	2//60	NS2	PSCC2	0.63	6//9		
1022415	59	F	6//24	6//36					LE	0.33	6//36	NS3		0.63	6//9		
1022426	60	F	6//18	6//24					LE	0.25	6//24	NS2		0.33	6//18		
1022427	62	F	5//60	6//60					RE	0.08	5//60	NS4		0.5	6//12		
1022427	64	F	6//60	1//60					LE	0.02	1//60	MC		0.33	6//18		
1022470	63	F	6//12	5//60					LE	0.08	5//60	NS3	PSCC1	0.5	6//12		
1022490	61	F	6//9	6//18					LE	0.17	6//18		PSCC2	0.5	6//12		
1022493	58	F	6//60	6//36					RE	0.1	6//60	NS2	PSCC1	0.25	6//24		
1022521	57	F	6//60	6//60					RE	0.1	6//60	NS3		0.5	6//12		
1022536	60	F	5//60	6//18					RE	0.08	5//60	NS2	PSCC2	0.5	6//12		
1022538	62	F	3//60	6//12					RE	0.05	3//60	NS3	PSCC3	1	6//6		
1022541	57	F	6//36	PL+					LE	0	PL+	MC		0.63	6//9		
1022546	62	F	6//60	PL+					LE	0	PL+	MC		1	6//6		
1022561	60	F	4//60	2//60					LE	0.04	2//60	NS2	PSCC3	1	6//6		
1022563	58	F	PL+	6//24					RE	0	PL+	MC		1	6//6		
1022564	70	F	6//12	PL+					LE	0	PL+	MC		0.63	6//9		
1022548	56	F	2//60	5//60					LE	0.08	5//60	NS3	PSCC1	0.25	6//24		
1022568	57	F	6//12	CFC					LE	0.02	CFC	NS4	PSCC1	0.25	6//24		
1022569	62	F	6//36	6//60	P		P		LE	0.1	6//60	NS3		0.5	6//12		
1022570	68	F	6//24	6//12					RE	0.25	6//24	NS1	PSCC1	0.63	6//9		
1022659	66	F	1//60	2//60					RE	0.02	1//60	NS3	PSCC1	0.63	6//9		
1022669	63	F	6//36	1//60					LE	0.02	1//60	NS2	PSCC3	0.63	6//9		
1022696	65	F	PL+	0.5//60	P		P		RE	0	PL+	MC		0.63	6//9		
1022706	56	F	5//60	6//24					RE	0.08	5//60	NS4		0.02	CFC		
1022709	52	F	0.5//60	1//60					RE	0.02	0.5//60	NS3	PSCC1	0.33	6//18		
1022721	59	F	PL+	6//36					RE	0	PL+	MC		1	6//6		
1022722	56	F	5//60	5//60					LE	0.08	5//60	NS3	PSCC1	1	6//6		
1022737	60	F	5//60	PL+	P	P			LE	0	PL+	MC		1	6//6		
1022744	61	F	6//12	CFC					LE	0.02	CFC	MC		1	6//6		
1022748	66	F	6//12	5//60					LE	0.08	5//60	NS3	PSCC1	0.63	6//9		
1022767	65	F	6//60	PL+					LE	0	PL+	MC		1	6//6		
1022768	54	F	2//60	1//60			P		LE	0.02	1//60	MC		1	6//6		
1022787	57	F	5//60	5//60	P				LE	0.08	5//60	NS2	PSCC2	0.63	6//9		
1022841	58	F	1//60	6//36					RE	0.02	1//60	NS2	PSCC3	0.63	6//9		
1022860	59	F	6//12	5//60			P		LE	0.08	5//60	NS4		1	6//6		

1022864	60	F	6//36	2//60					LE	0.04	2//60	NS2	PSCC3	0.63	6//9		
1022871	63	F	6//36	5//60	P	P	P		LE	0.08	5//60	NS3	PSCC1	1	6//6		
1022883	62	F	6//36	2//60					LE	0.04	2//60	NS2	PSCC2	1	6//6		
1022949	63	F	6//24	PL+					LE	0	PL+	MC		0.63	6//9		
1022952	62	F	1//60	5//60					LE	0.02	1//60	NS3	PSCC2	0.63	6//9		
1022953	61	F	PL+	1//60					RE	0	PL+	MC		0.63	6//9		
1022963	60	F	3//60	6//24			P		RE	0.05	3//60	NS2	PSCC2	1	6//6		
1022965	67	F	3//60	6//60					RE	0.05	3//60	NS4		1	6//6		
1023032	66	F	CFC	3//60					RE	0.02	CFC	MC		0.63	6//9		
1023048	70	F	3//60	6//24	P		P		RE	0.05	3//60	NS2	PSCC2	0.5	6//12		
1023075	65	F	4//60	6//24					RE	0.07	4//60	NS3	PSCC1	0.5	6//12	COMBINED	
1023203	61	F	6//18	6//6					RE	0.33	6//18		PSCC1	0.63	6//9		
1023279	56	F	6//60	6//18					RE	0.1	6//60	NS2	PSCC2	0.63	6//9		
1023517	59	F	PL+	PL+					LE	0	PL+	MC		0.63	6//9		
1023518	60	F	6//9	HM					LE	0.02	HM	MC		0.5	6//12		
1023556	60	F	3//60	1//60					LE	0.02	1//60	MC		1	6//6		
1023565	65	F	HM	5//60					RE	0.02	HM	NS4	PSCC1	0.5	6//12		
1023571	64	F	2//60	1//60					LE	0.02	1//60	NS4	PSCC2	0.63	6//9		
1023574	62	F	2//60	PL+					LE	0	PL+	MC		1	6//6		
1023587	60	F	5//60	6//24	P	P	P		RE	0.08	5//60	NS3	PSCC1	1	6//6		
1023650	66	F	6//18	6//12					RE	0.33	6//18	NS1	PSCC1	0.5	6//12		
1023651	61	F	6//18	6//24					LE	0.25	6//24	NS3		0.1	6//60		
1023653	56	F	3//60	6//12					RE	0.05	3//60	NS3	PSCC2	0.17	6//36		
1023751	67	F	5//60	5//60					RE	0.33	6//18		PSCC1	0.33	6//18		
1025275	57	F	6//12	1//60					LE	0.02	1//60	NS3	PSCC	0.5	6//12	COMBINED	
1025276	54	F	6//18	6//12	P				LE	0.33	6//18	NS2	PSCC2	0.25	6//24		
1025281	68	F	6//18	PL+					LE	0	PL+	MC	,	0.63	6//9	DENSE SK	
1025282	71	F	6//36	3//60					LE	0.05	3//60	MC		0.63	6//9	PCR+SFIOL	
1025289	73	F	6//18	6//12			P		RE	0.33	6//18	NS2		0.17	6//36		
1025290	62	F	5//60	6//12					RE	0.08	5//60	NS2	PSCC1	0.63	6//9		
1025296	65	F	6//60	3//60					LE	0.05	3//60	NS2	PSCC1	0.33	6//18		
1025298	69	F	5//60	HM	P				LE	0.02	HM	NS2	PSCC1	0.5	6//12		
1025304	69	F	6//36	PL+			P		LE	0	PL+	NS3		0.33	6//18		
1025306	59	F	2//60	NO PL					RE	0.04	2//60	NS1	PSCC3	0.17	6//36		
1025307	58	F	6//12	6//60					LE	0.1	6//60	NS2	PSCC1	0.5	6//12		
1025345	60	F	HM	6//12					RE	0.02	HM	NS2	PSCC3	0.63	6//9		
1025450	58	F	6//18	2//60	P				LE	0.04	2//60	MC		0.33	6//18		
1025523	64	F	6//60	6//12					RE	0.1	6//60	NS2	PSCC1	0.25	6//24		
1025556	58	F	6//18	6//24	P	P			LE	0.25	6//24	NS2	PSCC2	0.33	6//18		
1025610	66	F	5//60	6//60					LE	0.08	5//60	NS3	PSCC	0.5	6//12	COMBINED	
1025630	61	F	6//60	1//60					LE	0.02	1//60	NS3	PSCC1	0.5	6//12		
1025663	56	F	HM	PL+					LE	0	PL+	MC		1	6//6		
1025664	59	F	6//12	5//60					LE	0.08	5//60	NS2	PSCC1	0.63	6//9		
1025726	67	F	6//36	6//12					RE	0.17	6//36	NS1	PSCC2	1	6//6		
1025740	65	F	6//9	6//18	P	P			LE	0.33	6//18	MC		1	6//6		
1025763	70	F	6//18	6//36					LE	0.17	6//36	NS2	PSCC1	1	6//6		
1025801	71	F	6//12	6//60					LE	0.1	6//60		PSCC2	0.5	6//12		
1025932	73	F	6//60	6//60					LE	0.1	6//60	NS2	PSCC1	0.17	6//36		
1025961	62	F	5//60	6//18	P		P		RE	0.08	5//60	NS2	PSCC1	1	6//6		
1026038	69	F	6//18	PL+					LE	0	PL+	NS2	PSCC1	0.63	6//9		
1026062	59	F	4//60	2//60					LE	0.04	2//60	NS2	PSCC2	0.63	6//9		

1026066	58	F	PL+	6//24					RE	0	PL+	MC		0.02	CFC	Glaucomatous optic atrophy
1026071	60	F	6//12	PL+					LE	0	PL+	NS2		0.33	6//18	
1026094	60	F	5//60	5//60	P				RE	0.08	5//60	NS2	PSCC2	0.04	2//60	
1026096	62	F	2//60	5//60		P			RE	0.04	2//60	NS3		1	6//6	
1026131	59	F	6//12	CFC					LE	0	CFC	NS3		1	6//6	
1026132	58	F	6//36	6//60		P			LE	0.1	6//60	NS2		1	6//6	
1026127	63	F	6//24	1//60					LE	0.02	1//60	NS3		0.33	6//18	COMBINED
1026128	58	F	1//60	2//60					RE	0.02	1//60	NS2		0.63	6//9	
1026129	58	F	6//36	1//60					LE	0.02	1//60	NS4		0.63	6//9	
1026131	69	F	6//24	3//60					LE	0.05	3//60	MC		1	6//6	
1026146	65	F	5//60	6//24					RE	0.08	5//60	NS3	PSCC1	0.63	6//9	
1026196	58	F	6//60	6//9	P				RE	0.1	6//60	MC		1	6//6	
1026227	64	F	5//60	5//60					RE	0.08	5//60	MC		0.63	6//9	
1026279	65	F	6//24	1//60	P				LE	0.02	1//60	NS3		1	6//6	
1026285	62	F	6//60	PL+					LE	0	PL+	NS2		1	6//6	
1026287	60	F	2//60	1//60					LE	0.02	1//60	NS3		1	6//6	
1026289	63	F	5//60	5//60					RE	0.08	5//60	MC		1	6//6	
1026135	59	F	1//60	6//36					RE	0.02	1//60	NS2	PSCC	0.63	6//9	
1026137	57	F	6//36	1//60					LE	0.02	1//60	NS2		0.63	6//9	
1026139	59	F	6//12	5//60					LE	0.08	5//60	NS3		1	6//6	
1026138	67	F	6//36	2//60					LE	0.04	2//60	NS3		1	6//6	
1026141	69	F	6//36	2//60					LE	0.02	2//60	MC		0.33	6//18	old BRVO
1026145	64	F	5//60	3//60					LE	0.05	3//60	NS4	PSCC1	0.63	6//9	
1026149	59	F	6//18	6//60	P				LE	0.1	6//60	NS3		0.63	6//9	
1026152	52	F	6//12	2//60					LE	0.04	2//60	NS2		1	6//6	
1026198	59	F	6//24	PL+					LE	0	PL+	NS3	PSCC	0.63	6//9	
1026226	60	F	1//60	5//60	P	P			RE	0.02	1//60	NS3	PSCC	0.63	6//9	
1026220	62	F	PL+	1//60					RE	0	PL+	NS3		0.63	6//9	
1026235	64	F	6//18	PL+					LE	0	PL+	NS3		1	6//6	
1026250	67	F	6//36	6//24					RE	0.17	6//36	NS2		0.63	6//9	
1026271	63	F	6//24	6//24	P		P	P	RE	0.25	6//24	NS3		1	6//6	
1026274	66	F	PL+	PL+					LE	0	PL+	NS2		0.63	6//9	