CORNEAL ENDOTHELIAL CELL .LOSS AFTER MANUAL SMALL INCISION CATARACT SURGERY AND PHACOEMULSIFICATION AT MUHIMBILI NATIONAL HOSPITAL, DAR ES SALAAM

By

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A Dissertation Submitted in partial Fulfillment of the Requirements for
The Degree of Masters of Medicine (Ophthalmology) of
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CERTIFICATION

The undersigned certify that they have read and hereby recommend for acceptance by Muhimbili University of Health and Allied Sciences a dissertation entitled: **Corneal endothelial cell loss after manual small incision cataract surgery and phacoemulsification at Muhimbili National Hospital, Dar es Salaam**, in partial fulfillment of the requirements for the degree of Master of Medicine (Ophthalmology) of the Muhimbili University of Health and Allied Sciences.

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DECLARATION & COPYRIGHT

I, Dr Ng'hungu Lufunga, declare that this is my original work and that it has not been presented and will not be presented to any other University for a similar or any other degree award.

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Finally, I would like to glorify the name of the Almighty God, for giving me health, power and the ability to accomplish this work.

DEDICATION

To my wife Felista Boniphace, my son George and my two daughters Marietha and Catherine for their patience, understanding and encouragement despite that they were missing me most of the time.

ABSTRACT

Background: Cataract is the world's leading cause of blindness with around 20 million people bilaterally blind due to cataract[1]. The definitive treatment of cataract is surgical removal of the opacified lens which is aimed at restoring normal vision [3, 5]. However, every cataract extraction even in experienced hands is associated with some endothelial cell damage. Endothelial cell loss if sufficiently severe can cause endothelial cell density to fall below the threshold level needed to maintain corneal transparency. This can lead to the development of late corneal decompensation which ultimately leads to poor visual outcome which has been reported to occur in about 1% of cataract extractions [7, 11, 27, 28, 29].

Broad objective: To assess the corneal endothelial cell loss after manual small incision cataract surgery and phacoemulsification at Muhimbili National Hospital, Dar es Salaam.

Methodology: A hospital based descriptive, prospective cohort study was conducted between April 2012 and January 2013. A total of 127adult patients with 131eyes who underwent cataract surgery at MNH were enrolled. Patients were examined whereby demographic data, pre and post operative corneal endothelial cell count as well as post operative visual acuity were recorded. Corneal endothelial cell count was determined by use of a TOPCON SP.2000P specular microscope.

Results: All 131 eyes assessed by specular microscope had preoperative corneal endothelial cell count ranging from 1500 – 3000 cells/mm^2 which decreased with increasing age ($P<0.01$). Eighty nine eyes (67.9%) underwent MSICS while 42 eyes (32.1%) underwent phacoemulsification with 84 eyes (64.1%) been for males and 47 eyes (35.9%) for females. Fifty four (60.7%) out of 89 eyes lost significant cornea endothelial cells on day 1 with average cell loss of 611.1 cells/mm^2 while 33 eyes (37.1%) had a significant cell loss 1 month post MSICS. In the phacoemulsification group, 19 (45.2%) out of 42 eyes had a significant cornea endothelial cell loss on day 1 with an average endothelial cell loss of 552.6 cells/mm^2 while 12 eyes (28.6%) lost cells 1 month post phacoemulsification. An accelerated decrease in corneal endothelial cell count in MSICS and phaco groups one month post cataract surgery

was also noted in 17 eyes (19.1%) in MSICS group and 3 eyes (7.14%) in phaco group. However, there was no statistically significant association between postoperative visual acuity and different levels of corneal endothelial count on first day and one month post cataract surgery with P values of 0.111 and 0.142 on first day, 0.716 and 0.213 after one month for MSICS and phacoemulsification respectively.

Conclusion: The loss of cornea endothelial cells post cataract surgery at MNH is significant although not to an extent of causing cornea oedema.

Recommendations

1. Emphasis on preoperative corneal endothelial cell evaluation should be done.

2. Further study is proposed to identify the factors responsible for corneal endothelial cell loss as well as long term follow up of post cataract surgery patients.

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ABBREVIATIONS:

AC – Anterior chamber

BCVA – Best corrected visual acuity

ECCE – Extra capsular Cataract Extraction

ICCE – Intra capsular Cataract Extraction

IOL – Intraocular Lens

MNH – Muhimbili National Hospital

MSICS – Manual small incision cataract surgery

MUHAS – Muhimbili University of Health and Allied Sciences

Phaco - Phacoemulsification

Postop – Post operative

VA – Visual acuity

WHO – World Health Organization

1.0 INTRODUCTION

Cataract is the world's leading cause of blindness with around 20 million people blind due to cataract. It is a major cause of severe visual impairment leading to bilateral blindness. In developing countries 50 – 90% of all blindness in people who are above 50 years of age is caused by cataracts[1].

A cataract is a clouding of the lens of the eye that may lead to loss of vision. Cataracts result from many conditions, the most common cause is the natural aging process. This is a result of degenerative changes on the specific lens proteins within the lens which are altered leading to gradual clouding of the lens. Other causes include eye injury, chronic eye diseases and other systemic diseases, such as diabetes mellitus[2].

The definitive treatment of cataract is surgical removal of the opacified lens[3]. Cataract surgery is the most common major eye surgery and comprises 80% of eye surgeries done in eye units[4]. The aim of cataract surgery is to prevent blindness by restoring normal vision[5] as well as improving the quality of life. Studies have shown that successful cataract surgery in previously blind individual improves a household's economic status[6].

Every cataract extraction even in experienced hands and modern techniques involves some corneal manipulation and endothelial damage. Severe damage leads to late corneal decompensation, which has been reported to occur in about 1% of cataract extractions[7]. The damage to the corneal endothelium leads to corneal endothelial cell loss which if severe can eventually lead to the development of corneal edema[8]. This leads to the loss of corneal transparency and poor visual outcome post cataract surgery.

Corneal transparency is regulated by a corneal endothelial pump that can be interfered by surgical trauma. The corneal endothelium is the innermost layer of the cornea made up of hexagonal cell monolayer that does not divide significantly i.e does not regenerate when damaged. The corneal endothelial layer possesses the ability to pump water out of the corneal stroma against an osmotic gradient since it contains an active Na^+/K^+ pump[8, 9, 10]. Its cell count varies from 3500 – 4000 cells/mm^2 in infancy to about 2400 to 3200 cells/mm^2 in adults[11].

Studies have shown that, for the cornea endothelium to perform the function of maintaining corneal clarity it must consist of healthy cells above a minimum cell density which is reported to range from 400 – 700cells/mm^2. If the average cell count per square millimetre falls below this limit, it is likely that stromal oedema and bullous keratopathy will result following cataract surgery [8, 10, 11, 12]. Therefore, precautions have to be taken not to damage the corneal endothelium during cataract surgery since damage to the endothelium will lead to further reduction in endothelial cell density [13, 14].

According to the available literatures, corneal endothelial cell density reduction may be a result of a number of factors. Cataract extraction has been reported by several studies to be one of the factors contributing to the reduction of corneal endothelial cell density. The mechanisms are related to surgical manipulation of the surgeons resulting in corneal endothelial damage and consequently leading to corneal endothelial cell loss [13, 14].

Other factors leading to corneal endothelial cell loss include, (a) Aging process [10, 15] as in most individuals the cell density decreases from birth to death due to the senile degenerative changes of the corneal endothelium, (b) Glaucoma, literatures have shown that elevated intraocular pressure result in the gradual loss of endothelial cells and a progressive loss in endothelial function which is said to be not the direct result of high pressure but rather due to some metabolic disturbance, such as prolonged low oxygen concentration in the aqueous humor[16], (c) Intraocular inflammation is said to be another factor contributing to endothelial cell loss, endothelial cells appear to suffer little intracellular damage as a result of the inflammatory cell invasion whereby in the most advanced cases the inflammatory cells dislodge individual endothelial cells and cause them to float free in the aqueous humor[17], (d) Blunt ocular trauma in which specular microscopic studies have shown that posterior annular keratopathy, occurring after blunt corneal trauma in humans, represents a contusion injury and consists of disrupted and swollen endothelial cells[18,19] and (f) Fuchs' endothelial dystrophy[11]. Hence, preoperative assessment of corneal endothelium as well as ruling out the existence of such conditions is important for the surgery to be successful.

Different studies which have been conducted to evaluate the corneal endothelial cell count after cataract extraction have shown a variation in the prevalence of corneal endothelial cell

loss. In early studies, cataract extraction performed by phacoemulsification or a similar method generally resulted in greater endothelial cell loss (30 to 40%) than more conventional forms of cataract extraction [20, 21]. Recent studies have shown a prevalence of corneal endothelial cell loss in micro incision cataract surgery of about 9.5%, whereas that after standard phacoemulsification of about 7.6% [22].

Although several studies have been conducted with regard to corneal endothelial cell loss after cataract extraction, there is no published data yet to point out the overall prevalence of corneal endothelial cell loss in our environment and/ or country. This necessitated for studies to be conducted in our environment for the assessment of the corneal endothelium preoperatively and post operatively as it would help to provide baseline information for future studies. It would also contribute towards improvement of quality of care at MNH by improving postoperative visual outcome.

At Muhimbili National Hospital few patients undergo endothelial cell count preoperatively and no studies had been carried out before to evaluate the cornea endothelial cell loss postoperatively so the extent to which intraocular surgeries cause corneal endothelial damage was not known at the hospital. The courage to learn new techniques of cataract surgery is also increasing as in other parts of the world[23], hence a study to evaluate the health status of cornea endothelium both preoperatively and postoperatively at the hospital was proposed.

The main objective of the current study was to assess the corneal endothelial cell count pre and post cataract surgeries at Muhimbili National Hospital. The study has evaluated and quantified the corneal endothelial cell loss related to this commonest intraocular surgery at MNH. Therefore, the study has raised awareness to the surgeons on the importance of pre and post operative evaluation of patients for cataract surgery which is going to contribute towards the efforts of the hospital on improving the quality of cataract surgery.

2.0 LITERATURE REVIEW

Corneal endothelial cell loss remains a well known undesirable complication of cataract surgery that may in severe cases result in a negative impact on visual outcome after cataract surgery. This occurs when the average cell count per square millimetre falls below 1000, which is likely to cause stromal oedema and bullous keratopathy [8, 10, 11, 12] consequently resulting in poor visual outcome after cataract surgery.

It had been shown that all intraocular surgeries result in some corneal endothelial cell drop-out[24]. Cataract surgery has been one of intraocular surgeries that produces some corneal endothelial cell loss [13, 14]. Although the corneal endothelial cell loss is said to be caused mostly by mechanical injury at operation[25], endothelial cells are also vulnerable to mild trauma like irrigation during surgery by comparatively high volumes which could have a long-term effect on the rate of cell loss[26].

It is understood that the corneal endothelium is responsible for the maintenance of the corneal transparency due to the presence of an active Na^+/K^+ ATP pump which pumps water out of the stroma and hence keeping it relatively dehydrated[8, 9, 10]. The damage to corneal endothelium with consequent reduction in endothelial cells may lead to an ineffective pumping of water from the corneal stroma leading to corneal edema which ultimately leads to poor visual outcome [11, 27, 28, 29].

The mechanisms through which cataract surgery cause corneal endothelial cell loss and eventually leading to cornea edema include direct trauma of the cornea endothelium resulting from touching of the corneal endothelium during surgery by instruments [14, 30, 31, 32]. Instruments cause a direct damage to the corneal endothelium leading to a reduction in corneal endothelial cells due to the fact that when the corneal endothelium is damaged it does not regenerate but just heals by sliding of the endothelial cells to fill the gap left by the damaged cells[9, 11].

Insertion of intraocular lenses (IOL) is another mechanism through which corneal endothelial cell loss can occur during cataract surgery where by the IOL can accidentally touch the endothelium [14, 30, 31, 32]. Due to the strong adhesive force between the methacrylate lens and the endothelial cell membrane, they stick together when they come in contact with one another; the separation of these two surfaces causes the cell membrane to be torn from endothelial cells resulting into the damage of the corneal endothelial cells [33, 34, 35]. However, a study on reduction in endothelial cell density following cataract extraction and intraocular lens implantation showed that the major cause of endothelial cell loss is operative trauma to the endothelium whereby cataract extraction with lens implantation but without endothelial contact does not result in a \significant corneal endothelial cell reduction than the cataract extraction alone[36]. When different types of intraocular lens are compared, studies have shown the implantation of an anterior chamber lens to be more traumatic than that of a posterior chamber lens[37, 38]. But, this has been reduced by the modification of available implants and surgical techniques to an acceptable level of surgical risk.

The volume of fluid irrigated into the eye at the time of surgery as well as the mechanical contact with nuclear fragments also determines the extent of corneal endothelial cell loss by causing some trivial injuries to the endothelium [39, 40, 41]. The types of irrigating solutions have also a detrimental effect to the cornea endothelium. Studies on corneal perfusion tudies have shown that endothelial structure and function is best maintained when solutions that resemble aqueous humor in composition are used. Irrigating solutions not satisfying the basal metabolic requirements of the corneal endothelium can rapidly produce adverse endothelial changes and corneal edema [26, 42]. The individual surgeon techniques and skill is also reported to be among the determinants of corneal endothelial cell loss following cataract surgery [39, 40, 41].

The risk of corneal endothelial cell loss is also increased by surgical complications like posterior capsular rupture and vitreous loss. Other factors like ultrasonic vibrations or contact of cornea endothelium with other ocular tissues due to flat anterior chamber or vitreous touch [14, 30, 31, 32] contribute also to the increased risk for corneal endothelial cell loss after cataract surgery.

Other mechanisms involved include cataract surgery incision size and location e.g after cataract surgery, endothelial damage and cell loss is greatest in the superior cornea because the incision is placed superiorly and the lens extraction itself is most traumatic to the superior endothelium .Since endothelial wound healing occurs primarily by migration of adjacent cells rather than by cell replication, a vertical disparity in cell density persists for many years [43].

Although cataract surgery can cause corneal endothelial cell loss postoperatively which can ultimately lead to corneal oedema which can compromise the visual outcome, preoperative assessment of the corneal endothelial cell count is vital since a clear cornea with normal thickness is not necessarily associated with normal endothelial morphology or cell density. Cornea oedema is more likely to occur when preoperative cell density is below the range of 400 - 700cells/mm^2 but unlikely to occur when density is greater than 1000cells/mm^2 [8, 11, 12].

Literatures have shown the existence of conditions which can cause corneal endothelial cell loss other than cataract surgery. These include: (a) Aging - central endothelium changes as a function of age [15]. In most individuals the cell density decreases from birth to death, (b) Glaucoma -persistently elevated intraocular pressure is said to result in the gradual loss of endothelial cells and a progressive loss in endothelial function [16], (c) Intraocular inflammation - endothelial cells suffer little intracellular damage as a result of the inflammatory cell invasion [17], (d) Blunt ocular trauma[18, 19] and (e) Fuchs' endothelial dystrophy[11]. Hence, preoperative assessment of corneal endothelium as well as ruling out the existence of such conditions is important for the surgery to be successful.

There are mainly two types of cataract surgeries, namely intracapsular and extracapsular cataract extraction. The extracapsular cataract extraction can be in the form of conventional extra capsular cataract extraction (ECCE), manual small incision cataract surgery or phacoemulsification [8, 9] of which each technique can result in different rate of endothelial cell loss depending on individual surgeon techniques and skills which vary widely [39, 40].

Studies have shown an increased risk of severe cell loss with phacoemulsification in patients with hard cataracts suggesting that phacoemulsification may not be the optimal procedure in

these cases, and that ECCE should be preferred. However no significant difference in overall corneal endothelial cell loss exists between these two operative techniques [41, 44, 45]. The increased risk of trauma of corneal endothelium during cataract surgery by phacoemulsification is attributed to high total ultrasound energy required when the lens nucleus is hard and hence increasing the risk for corneal endothelial cell loss [39, 40, 41]. But contradictory results have been obtained in another study which showed significant difference in endothelial cell reduction (9.17% in ECCE group, and 4.72% in Phaco group) concluding that under the same conditions phacoemulsification caused reduced operative trauma of the corneal endothelium[4].

Variations in the prevalence of corneal endothelial cell loss after cataract extraction have been shown to exist in different studies. In early studies, cataract extraction performed by phacoemulsification or a similar method generally resulted in greater endothelial cell loss (30 to 40%) than the more conventional forms of extraction [20, 21]. A recent study showed a prevalence of corneal endothelial cell loss in micro-incision cataract surgery of about 9.5%, whereas that after standard phacoemulsification of about 7.6% [22]. Another study on modern techniques of cataract extraction (eg, phacoemulsification) showed that they are associated with endothelial cell loss of about 4-10%; however, this depends on an individual patient of which wide variations in cell loss can occur [46] depending for example on the nature of the cataract.

Due to the fact that corneal endothelium is among the determinant of good outcome of cataract surgery by playing an important role in maintaining the cornea transparency hence during surgery measures to protect the integrity of corneal endothelium have to be taken. The routine use of viscoelastic agents has resulted in a dramatic decrease in endothelial cell loss and offers a practical and effective means of protecting the cornea from inadvertent trauma during cataract surgery [47]. Studies have indicated that using currently available apparatus and viscoelastic materials, cell loss has been reduced to 15% or less in intraocular procedures [48].

As it has been pointed out earlier the importance of corneal endothelium in the maintenance of cornea transparency, severe damage to the corneal endothelium during cataract surgery resulting in significant endothelial cell loss can lead to the development of an irreversible corneal edema [49] which represents an important cause of visual disability following routine cataract surgery. Sometimes, patients with relatively few endothelial cell counts undergoing cataract surgery may require extended periods to resolve their corneal edema, chiefly because only a few endothelial cells are pumping fluid out of the stroma and into the anterior chamber [11]. This emphasizes the importance of good technique to avoid endothelial cell damage at the time of surgery [50].

In order to understand the nature of corneal endothelium in terms of cell morphology and density, different methods have been employed to study it. One of these methods is specular microscopy which is the study of the changes in different layers of the cornea under very high magnification (100 times greater than slit-lamp biomicroscopy) i.e mainly used to assess the endothelium. It analyzes images of the corneal endothelium with respect to cellular size, shape, density and distribution. The most important indication for specular microscopy is the evaluation of the functional reserve of the cornea endothelium prior to intraocular surgery. Other indications include evaluation of the donor cornea regarding suitability for penetrating keratoplasty and demonstration of pathology particularly cornea guttata, Descemet membrane irregularities and posterior polymorphous dystrophy. Specular microscope can also distinguish the morphological features of altered cells from the rest of the endothelium [8, 11, 12].

Other methods used to study the corneal endothelium are ultrasound pachymetry and optical pachymetry which both measure the corneal thickness. Different diseases result in corneal edema which is associated with central corneal thickening as the cornea begins to swell. The measurements of corneal thickness are helpful in monitoring the progression of a disease process (eg, Fuchs dystrophy), as well as in assessing therapeutic regimens (eg, topical steroid use in corneal graft rejection). Optical pachymetry helps in measuring the depth of cornea pathology (eg, scars, other lesions) when the full thickness of the corneal stroma is not involved and it is necessary for therapeutic reasons to estimate the depth of the pathology[12].

Each of the methods for the assessment of corneal endothelium have their advantages and limitations or draw backs. Specular microscope has been able to give the corneal endothelial cell count and morphological features of altered cells from the rest of the endothelium [8, 11, 12]. It has an advantage over ultrasonic pachymetry and optical pachymetry in the evaluation of the degree of surgical trauma and corneal endothelial status. This is because the functional capacity of the endothelium is important and corneal cell depletion is not reflected in corneal thickness measurements until there has been substantial loss of corneal endothelial cells [51]. For example, corneas which have incurred moderate reductions in the number of corneal endothelial cells of otherwise healthy endothelial cells, the corneal thickness does not increase. A healthy endothelium is thus able to maintain corneal dehydration over a large range of endothelial cell counts hence been able to maintain a clear cornea [8, 11, 12, 51].

However the information obtained from specular microscopy is basically morphological and gives little indication of the functional capacity of the endothelium to maintain corneal deturgescence compared to pachymetry which measures corneal thickness which serves as a parameter for assessing overall endothelial function in corneas with a diseased endothelium or with borderline low endothelial cell counts [51, 52] since corneal thickness increases when there is an inefficient pumping activity of the corneal endothelium due to either diseased endothelium or reduced corneal endothelial cells below the lower limit required for the maintenance of a clear cornea [8, 11, 12]. Also it is possible to perform specular microscopy only when the cornea is optically clear. Thus the presence of oedema, scarring, or infiltration of the cornea prevents the examination of the underlying endothelium [52].

3.0 STATEMENT OF THE PROBLEM

Corneal endothelial cell loss is one of the complications of cataract surgery. When the corneal endothelial cell loss is severe, it is associated with poor visual outcome after cataract surgery. This result from severe damage to the corneal endothelium during cataract surgery resulting to significant endothelial cell loss that can lead to the development of an irreversible corneal edema[49] resulting into visual disability following routine cataract surgery.

Cornea oedema is more likely to occur when endothelial cell density is below the range of 400 - 700cells/mm^2 but unlikely to occur when density is greater than 1000cells/mm^2 [8, 11, 12]. This is due to the fact that, for the cornea endothelium to perform the function of maintaining corneal clarity it must consist of healthy cells above a minimum cell density which are capable of pumping fluids out of the stroma. This emphasizes the need for preoperative assessment of the corneal endothelial cell count as it gives a clue on the visual outcome of the patient due to the high chance of development of severe cornea oedema if the endothelial cell count is very low preoperatively.

At Muhimbili National Hospital few patients undergo endothelial cell count preoperatively and no studies had been carried out to assess the corneal endothelial cell loss postoperatively before this study. Therefore, the extent to which intraocular surgeries caused corneal endothelial damage was not known at the hospital.

A study to assess the corneal endothelial cell count pre and post cataract surgery at Muhimbili National Hospital was therefore proposed. The study which has evaluated and quantified the corneal endothelial cell loss related to this commonest intraocular surgery at MNH.

4.0 RATIONALE OF THE STUDY

This was the first study to be conducted at Muhimbili National Hospital. Therefore, it has provided baseline data for future studies related to pre and post operative corneal endothelial cell count. The study had also raised awareness to the surgeons on the importance of pre and post operative evaluation of patients for cataract surgery. This is going to contribute towards the efforts of the hospital on improving the quality of cataract surgery.

5.0 OBJECTIVES

5.1 Broad objective

- To assess the corneal endothelial cell loss after manual small incision cataract surgery and phacoemulsification at Muhimbili National Hospital, Dar es Salaam.

5.2 Specific objectives

1. To determine the pre-operative corneal endothelial cell count among patients undergoing cataract surgery by age.
2. To determine the corneal endothelial cell loss after manual small incision and phacoemulsification cataract surgery by age.
3. To determine the association between endothelial cell loss and post operative visual acuity.

6.0 METHODOLOGY:

6.1 Study area

The study was conducted at Muhimbili National Hospital (MNH) which is a National Referral Hospital and University Teaching Hospital with 1,500 bed facility, attending 1,000 to 1,200 outpatients per week, admitting 1,000 to 1,200 inpatients per day. An average of 245 patients undergoes cataract surgery per year at the hospital i.e with an average of 200 patients undergoing manual small incision cataract surgery (MSICS) and about 45 patients undergoing phacoemulsification. For example, according to the theatre register book in the year 2010, a total of 231 patients underwent cataract surgery with 208 patients undergoing MSICS and 28 patients undergoing phacoemulsification. While in the year 2011 a total of 259 patients underwent cataract surgery with 215 patients undergoing manual small incision cataract surgery and 44 patients undergoing phacoemulsification.

The hospital is located within Dar es Salaam city, which has a population of more than 4 million people.

6.2 Study population

All adult patients who were diagnosed to have cataract and planned for cataract surgery at MNH eye clinic.

6.3 Study design

A hospital based descriptive, prospective cohort study.

6.4 Inclusion criteria

All adult patients with cataracts who underwent either manual small incision cataract surgery or phacoemulsification both males and females were included. Only patients who consented were included.

6.5 Exclusion criteria

Patients with history of previous intraocular surgery, significant corneal opacification, cornea dystrophy, uveitis, glaucoma or vitreous loss during surgery were excluded.

6.6 Sampling and Sample size

Convenient sampling[53] was used. All adult patients with cataracts undergoing either manual small incision cataract surgery or phacoemulsification at Muhimbili National Hospital during the study period were included.

6.7 Procedures

All adult patients who were diagnosed to have cataract and planned for cataract surgery during the study period were informed about the study. An informed consent form was filled by each patient before a patient was included in the study. A thorough history was taken followed by a general and systemic examination which was performed by the investigator. All information was recorded on a structured questionnaire.

Ocular examination was performed starting with visual acuity using Snellens chart/illiterate e-chart. Topcon slit lamp bio-microscope was used for examination of the anterior segment of the eye including the eyelids, conjunctiva, cornea, anterior chamber, iris, pupil, lens and anterior vitreous. Intraocular pressure measurement was done by using a Topcon pneumotonometer.

Patients were allocated to either manual small incision cataract surgery group or phacoemulsification according to the grade of cataracts they presented with. Grading of cataract was done using WHO – simplified cataract grading system (2002) whereby cataracts grade2 up to grade 3 whether it was nuclear, cortical or posterior subcapsular cataract was fit for phacoemulsification while grade 9 was for manual small incision cataract surgery.

Corneal endothelial cell count assessment was determined by use of a non contact TOPCON SP.2000P specular microscope, the only machine which was available at the department at the time of study which gives reliable and reproducible values for endothelial cell density at high magnification [8, 11, 12, 57]. The machine enumerates the number of endothelial cells, allowing an objective assessment of the patient's endothelium which guides the physician to make a mutual decision. It is useful for the selection of eyes for intraocular surgery in general and foreseeing the prognosis in cases of donor corneas. The photographs were taken a day before surgery and were repeated on day one and one month after cataract surgery. The endothelial cell count was obtained by matching the corneal endothelial cell pictures with the scale on the photograph (figure1 and 2).

Examination of the posterior segment was done by slit lamp bio-microscope using 78D/90D volk lenses preoperatively for patients who did not have dense or bilateral cataracts and post operatively for all patients after dilatation of both pupils using cyclopentolate or tropicamide eye drops.

The best corrected visual acuity in the better eye of 6/4 - 6/18 was graded as normal, that of <6/18 to ≥3/60 was graded as visual impairment and visual acuity of less than 3/60 was regarded as blind. Refraction was done at one month post surgery.

Manual small incision cataract surgeries were performed mostly by residents (about 70%) while phacoemulsification were performed by experienced surgeons or ophthalmologist in

training for phacoemulsification procedure.

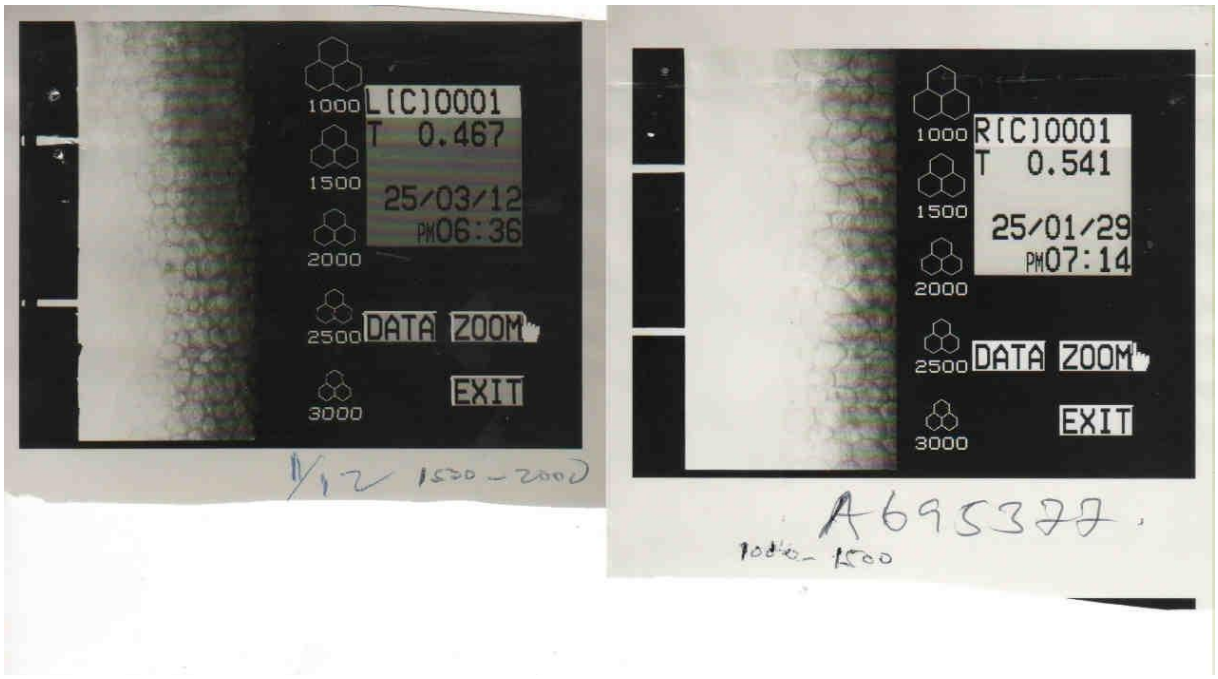


Figure 1: Corneal endothelial photographs showing endothelial counts between 1500 – 2000cells/mm^2 (Left) and 1000 – 1500cells/mm^2 (Right).

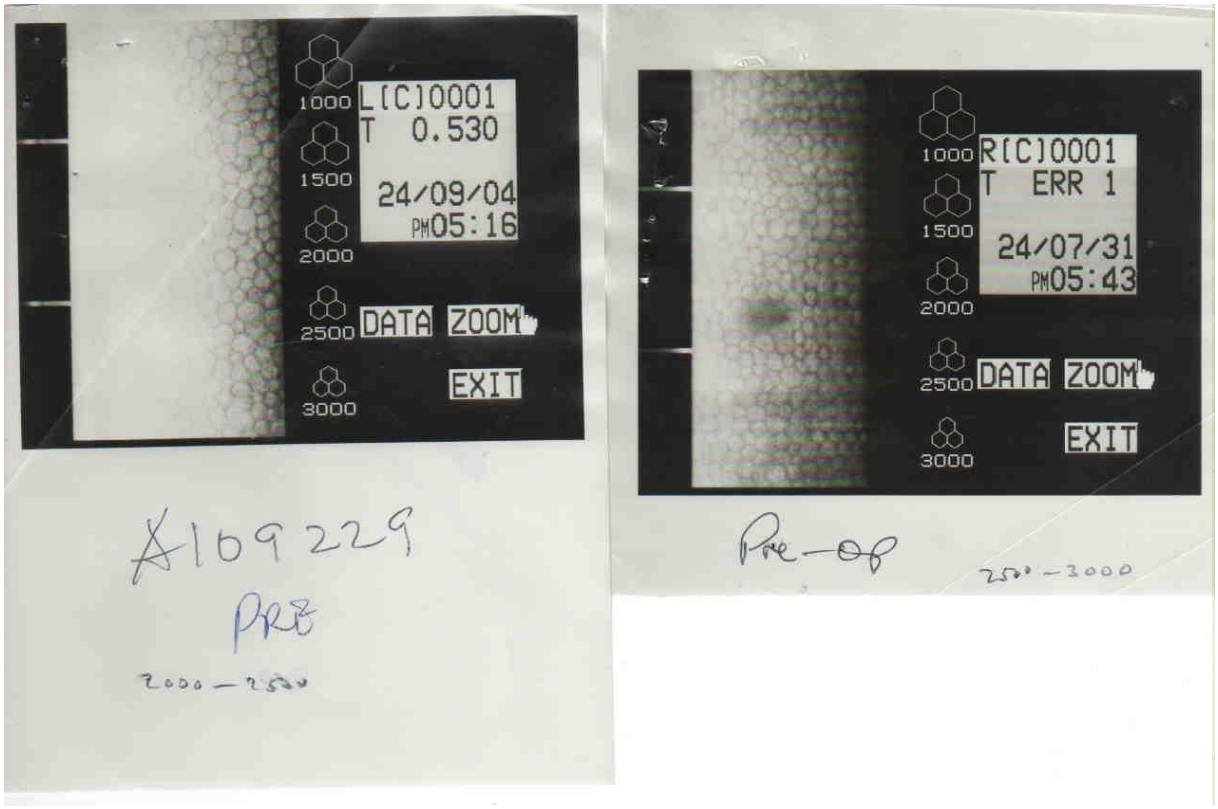


Figure 2: Cornea endothelial photographs showing endothelial counts between 2000 – 2500cells/mm^2 (Left) and 2500 – 3000cells/mm^2 (Right).

6.8 Ethical issues

Application for approval to the MUHAS Research and Publications Committee was done before commencement of the study. Patients were informed about the study, those who consented were included. However no patient was denied of appropriate and adequate treatment upon not consenting. All patients' information was kept confidentially and was not accessed by people not concerned with the study.

6.9 Data management and analysis

All the collected data were recorded into the checklist for storage of information and were checked by the research team for completeness and consistency. Data master sheets were used to process and analyze collected data by using tables and figures. Also data collected were entered using SPSS (Statistical Package for Social Sciences) version 16 and analysis was done using CHI SQUARE and P values of mantel hanzel statistics.

6.10 **Definition of terms**

Normal vision is defined by WHO as visual acuity of 6/4 to 6/18 in the better eye with best correction.

Visual impairment is defined by WHO as presenting visual acuity of <6/18 to ≥3/60 in the better eye with best correction.

Blindness is defined by WHO as presenting visual acuity of less than 3/60 in the better eye with best correction.

NB: For the case of this study it was referring to vision in the operated eye whether it had normal vision, visually impaired or blind.

Significant cornea endothelial cell loss is defined as change of cornea endothelial cell count from higher to lower level e.g from 2500 – 3000 cells/mm^2 to 2000 – 2500 cells/mm^2 e.t.c.

7.0 RESULTS

A total of 151 patients with cataract were planned for cataract surgery. Of these, 127 patients with 131 eyes met the criteria and were recruited and included in the final analysis of the study. Four patients had bilateral cataract surgery done. The age range was 26 – 90 with the mean age of 64.56 years. There were more males than females with M: F ratio of 2:1. Majority were in the age group 46 – 65years. Majority of patients underwent MSICS (Table1 and Figure3).

Table 1: Age and sex distribution of the study population (N=127).

Age group (Years)	Sex					
	Male		Female		Total	
	N	%	N	%	N	%
26 – 45	2	2.5	3	6.5	5	3.9
46 – 65	40	49.4	26	56.5	66	52.0
66 – 85	38	46.9	16	34.8	54	42.5
>85	1	1.2	1	2.2	2	1.6
Total	81	63.8	46	36.2	127	100

Figure 3: Distribution of type of surgery among the operated eyes (N=131)

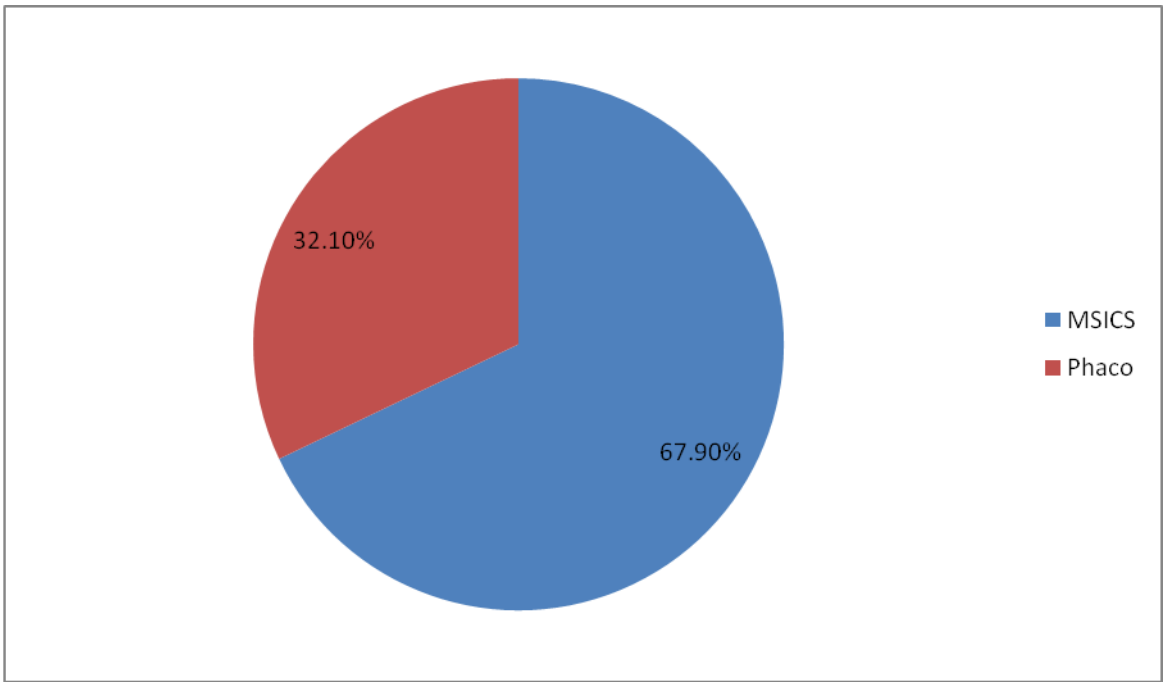


Table 2: Preoperative corneal endothelial cell count by age (N=131).

Age group (Years)	Corneal endothelial cell count (cells/mm^2)							
	1500 – 2000		2000 – 2500		2500 – 3000		Total	
	N	%	N	%	N	%	N	%
26 – 45	0	0.0	1	1.2	4	11.1	5	3.8
46 – 65	5	50.0	40	47.1	23	63.9	68	51.9
66 – 85	3	30.0	44	51.8	9	25.0	56	42.7
>85	2	20.0	0	0.0	0	0.0	2	1.5
Total	10	7.6	85	64.9	36	27.5	131	100

Preoperative cornea endothelial cell count ranged from 1500 – 3000 cells/mm^2 which decreased significantly with increasing age ($P < 0.01$).

Table 3: Cornea endothelial cell loss on day 1 and one month post MSICS by age

Age group (Years)	Lost cells Day1			Lost cells 1 month		
	Yes	No	Total	Yes	No	Total
26 - 45	2 (3.7%)	0 (0.0%)	2 (2.2%)	1 (3.0%)	1 (1.8%)	2 (2.2%)
46 -65	25 (46.3%)	22 (62.9%)	47 (52.8%)	17 (51.5%)	30 (53.6%)	47 (52.8%)
66 - 85	26 (48.1%)	12 (34.3%)	38 (42.7%)	14 (42.4%)	24 (42.9%)	38 (42.7%)
>85	1 (1.9%)	1 (2.9%)	2 (2.2%)	1 (3.0%)	1 (1.8%)	2 (2.2%)
Total	54 (60.7%)	35 (39.3%)	89 (100%)	33 (37.1%)	56 (62.9%	89 (100%)

Fifty four (60.7%) out of 89 eyes lost significant cornea endothelial cells on day 1 with average cell loss of 611.1 cells/mm^2 while 33 eyes (37.1%) had a significant cell loss 1 month post MSICS. Cornea endothelial cells ranged from 1000 - 3000 cells/mm^2 and 1000 - 2500 cells/mm^2 on day1 and one month post MSICS respectively. Cornea endothelial cell loss did not vary with age ($P>0.05$) on day one while there was a statistically significant decrease of cornea endothelial cell count with increasing age one month post MSICS (P value 0.035).

Table 4: Correlation of visual acuity and corneal endothelial cell count on the first day post-MSICS

Corneal endothelial cell count (cells/mm^2)	Postop VA day1									
	<3/60-NPL		<6/60 - 3/60		<6/18 - 6/60		6/4 - 6/18		Total	
	N	%	N	%	N	%	N	%	N	%
1000 – 1500	0	0.0	2	28.6	5	71.4	0	0.0	7	7.9
1500 – 2000	1	2.6	2	5.3	28	73.7	7	18.4	38	42.7
2000 – 2500	5	13.2	3	7.9	24	63.2	6	15.8	38	42.7
2500 – 3000	0	0.0	1	16.7	2	33.3	3	50.0	6	6.7
Total	6	6.7	8	9.0	59	66.3	16	18.0	89	100

Sixteen eyes (18%) had normal VA on the first day and there was no statistically significant variation in VA among different levels of corneal endothelial count. (P value 0.111).

Table 5: Correlation of best corrected visual acuity and corneal endothelial cell count 1month post MSICS

Corneal endothelial cell count (cells/mm^2)	VA one month post MSICS							
	<6/60 - 3/60		<6/18 - 6/60		6/4 - 6/18		Total	
	N	%	N	%	N	%	N	%
1000 – 1500	1	4.3	4	17.4	18	78.3	23	25.8
1500 – 2000	2	5.0	4	10.0	34	85.0	40	44.9
2000 – 2500	1	3.8	6	23.1	19	73.1	26	29.2
Total	4	4.5	14	15.7	71	79.8	89	100

Seventy one (79.8%) out of 89eyes had normal vision with no statistically significant variation of VA between different levels of cornea endothelial cell count ($P > 0.05$).

Table 6: Cornea endothelial cell loss on day 1 and one month post phacoemulsification by age

	Lost cells day1			Lost cells 1 month		
Age group (Years)	Yes	No	Total	Yes	No	Total
26 - 45	3 (15.8%)	0 (0.0%)	3 (7.1%)	0 (0.0%)	3 (10.0%)	3 (7.1%)
46 -65	6 (31.6%)	13 (56.5%)	19 (45.2%)	8 (66.7%)	11 (36.7%)	19 (45.2%)
66 - 85	10 (52.6%)	10 (43.5%)	20 (47.6%)	4 (33.3%)	16 (53.3%)	20 (47.6%)
Total	19 (45.2%)	23 (54.8%)	42 (100%)	12 (28.6%)	30 (71.4%)	42 (100%)

Nineteen (45.2%) out of 42 eyes had a significant cornea endothelial cell loss on day 1 with an average endothelial cell loss of 552.6 cells/mm^2 while 12 eyes (28.6%) lost cells 1 month post phacoemulsification. Cornea endothelial cell count ranged from 1000 – 3000 cells/mm^2 on both day 1 and one month post phaco. Cornea endothelial cell loss did not vary significantly with increasing age ($P>0.05$) both on day one and one month post phacoemulsification.

Table 7: Correlation of visual acuity and corneal endothelial cell count on the first day post Phacoemulsification

Corneal endothelial cell count (cells/mm^2)	Postop VA day1							
	<6/60 - 3/60		<6/18 - 6/60		6/4 - 6/18		Total	
	N	%	N	%	N	%	N	%
1000 – 1500	0	0.0	3	75.0	1	25.0	4	9.5
1500 – 2000	0	0.0	6	50.0	6	50.0	12	28.6
2000 – 2500	0	0.0	8	38.1	13	61.9	21	50.0
2500 – 3000	1	20.0	2	40.0	2	40.0	5	11.9
Total	1	2.4	19	45.2	22	52.4	42	100

Twenty two eyes (52.4%) had normal vision with no statistical significant variation of VA between different levels of corneal endothelial cell count (P value 0.142).

Table 8: Correlation of best corrected visual acuity and corneal endothelial cell count 1month post phacoemulsification

Corneal endothelial cell count (cells/mm^2)	VA 1 month post phaco					
	<6/18 - 6/60		6/4 - 6/18		Total	
	N	%	N	%	N	%
1000 – 1500	0	0.0	7	100	7	16.7
1500 – 2000	1	6.7	14	93.3	15	35.7
2000 – 2500	3	16.7	15	83.3	18	42.9
2500 – 3000	1	50.0	1	50.0	2	4.8
Total	5	11.9	37	88.1	42	100

Thirty seven (88.1%) out of 42eyes had normal vision with no statistical significant difference in variation of VA between different levels of corneal endothelial cell count (P value 0.213).

8.0 DISCUSSION

The findings of this study confirmed the evidence of other reports which revealed that as individual ages, the corneal endothelial cell count declines. The study revealed a preoperative statistically significant decrease in corneal endothelial cell count with increasing age (P value 0.000). Previous studies have shown a variation of the corneal endothelial cell count by age. The corneal endothelial cell count varies from 3500 – 4000 cells/mm^2 in infancy to about 2400 to 3200 cells/mm^2 in adults [11]. Further more evidence from literature reveals that as an individual age[3] the corneal endothelial count declines by 30 -50% between the ages of 20 and 80years. The mechanisms leading to endothelial cell loss with increasing age are not well known though some evidences associated it with aging degenerative changes [10, 15]. All eyes which were recruited in this study had corneal endothelial count ranging from 1500 - 3000 cells/mm^2 which was above the lower limit which is said to be capable of maintaining the corneal deturgescence. Hence, post operative bullous keratopathy was less likely expected to occur [8, 11, 12].

Studies have shown surgical trauma at the time of cataract surgery can be associated with a marked reduction in endothelial cell counts [13, 14, 22, 45]. The results of this study have also shown fifty four (60.7%) out of 89 eyes in MSICS group and nineteen (45.2%) out of 42 eyes in phacoemulsication group lost significant cornea endothelial cells on day 1 post surgery. The available literatures show enormous factors responsible for the corneal endothelial cell loss following cataract surgery. Some of these factors are related to pre-operative selection and evaluation of patients and other majorities are intra-operative factors. One example of the common preoperative factors is the overlooking on the preoperative examination, not performing specular microscopy preoperatively to identify patients with low endothelial count or those with corneal guttata hence the emphasis of this study is to create awareness to clinicians to perform specular microscopy to help quantify endothelial reserve and to aid in risk assessment. The technique phacoemulsification has been documented to be associated with endothelial cell loss of about 4-10%. Other factors documented to relate with endothelial cell loss are endothelial cell loss has been correlated with cataract incision size and location,

density of nucleus, total ultrasound energy used, volume of fluid irrigated into the eye at the time of surgery and individual surgeon techniques and skill [14, 30, 31, 32, 39, 40] . These mentioned factors could probably be among the factors responsible for the endothelial cell loss in the current study although a specific study need to be done to find out the responsible factors for the endothelial cell loss at this institution.

Endothelial cell loss if sufficiently severe can cause endothelial cell density to fall below the threshold level needed to maintain corneal transparency. The threshold of endothelial cell density varies considerably amongst individuals. The minimum cell density necessary for adequate function is in the range of 400 - 1000 cells/mm².The maintenance of relative stromal dehydration requires this critical number of functioning endothelial cells, without which the cornea becomes edematous and cloudy. The cornea endothelium's chief function is to remove fluid from the cornea stroma allowing the cornea to remain optically clear [8, 9, 10]. Hence, the cornea may remain clear after significant endothelial cell loss if the average cell count per square millimeter does not fall below the range of 400 - 1000cells/mm^2.

The current study reports no single patient/eye with endothelial count less than 1000cells/mm^2 postoperatively giving less chance for patients/eyes to have significant corneal oedema compromising visual outcome. This is evidenced by the normal BCVA seen one month post cataract surgery whereby in both groups i.e MSICS and phacoemulsification, majority of patients/eyes had normal BCVA and there was no statistical significant association between VA and different levels of corneal endothelial counts (P value of 0.716 and 0.213 for MSICS and phacoemulsification respectively). When the cell density falls below the minimum level necessary for adequate function, the remaining cornea endothelial cells fails to maintain it in a relatively dehydrated state leading to the development of bullous keratopathy [8, 10, 11] which ultimately leads to poor visual outcome [11, 27, 28, 29]. The bullous keratopathy creates irregularity at the optical tear film air – interface contributing to degraded optical performance of the cornea and can compromise visual acuity.

The current study showed that 33 eyes (37.1%) had a significant cell loss 1 month post MSICS with 17 eyes (19.1%) having continued cornea endothelial cell loss while 12 eyes

(28.6%) lost cells 1 month post phacoemulsification with 3 eyes (7.14%) having continued endothelial cell loss. Different reports have also shown that there is a continued corneal endothelial cell losses at an average rate of 2.5% per year for at least 10 years after surgery, with or without an IOL implantation. The causes of accelerated decrease in endothelial cell density after cataract extraction are still not understood but this has been noted to be attributed to the subclinical inflammation post surgery [54, 55, 56].

Patients in the current study were followed up for one month which was a very short time to be able to tell the ultimate effect of cataract surgery on the cornea endothelial count. Hence, a further study is proposed in terms of long term follow up at this institution. However, the results of this study should alert the surgeons on the importance of preoperative cornea endothelial cell count evaluation. Also being aware of the continued endothelial cell loss post surgery [54, 55, 56], patients with low level of endothelial count although they might have clear cornea should be operated with minimal manipulations. Due to the possibility of development of corneal oedema resulting from cornea endothelial cell loss in a long run, patients should be warned on chances of poor vision when the cornea endothelial count falls below lower limit which is capable of maintaining cornea transparency [8, 11, 12].

The age of the patient have been reported in some studies to be among the risk factors for increased corneal endothelial cell loss post cataract surgery [41, 58] however, in the current study no statistically significant decrease in postoperative corneal endothelial cell count with increasing age was observed in both MSICS and phacoemulsification group (P value 0.083 and 0.34 respectively). But, one month post cataract surgery there was a statistically significant decrease (P value 0.035) in corneal endothelial cell count among different age groups in MSICS group while in phacoemulsification group it did not show any statistically significant decrease.

The statistically significant decrease in corneal endothelial count with increasing age in the MSICS group can be explained by the fact that in practice at MNH, MSICS are mostly done by residents where more manipulations are expected while phacoemulsification are done by

experienced surgeons or ophthalmologist in training for phaco who would probably perform less manipulations. Another explanation may be related to the wound size big enough to cause more corneal insult in the MSICS group on the pre existing low endothelial cells count among older patients. However, due to the variability in the surgeons skills and technique as well as small sample size in this study, it is difficult to draw conclusion from these results hence further study to look on whether there was more continued endothelial cell loss with increasing age post MSICS than post phacoemulsification is proposed.

The current study showed that proportionally there were more MSICS as compared to phacoemulsification .This can be explained by the fact that MNH is a teaching hospital and MSICS is the main type of cataract surgery which is taught to the residents in training.

9.0 CONCLUSION

The loss of cornea endothelial cells post cataract surgery at MNH is significant although not to an extent of causing cornea oedema.

10.0 RECOMMENDATIONS

1. Emphasis on preoperative corneal endothelial cell evaluation should be done.

2. Further study is proposed to identify the factors responsible for corneal endothelial cell loss as well as long term follow up of post cataract surgery patients.

11. 0 STUDY LIMITATIONS

Being a hospital based study sometimes surgeries had to stop due to lack of consumables like viscoelastic hence limiting the attainment of the intended sample size. Small sample size also affected the study which needed a large sample size for proper comparison with the general population. Surgeries been performed by different surgeons did not allow comparison between loss of cornea endothelial cells in MSICS and that in phacoemulsification since every surgeon have different techniques and hence corneal endothelial cell loss would vary also.

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Appendix 1

Questionnaire: English version

SN……. File No: ……..

Mobile phone number:……

1. Age ……
2. Sex a) Male b)Female
3. Level of education
   a)primary b) secondary c)tertiary d) others
4. Is there any pain in your eyes
   a) Yes b) No
5. Status of the lens (Grade of cataract) ……..
6. Type of surgery the patient is to undergo,
   a)MSICS b)phacoemulsification
7. Preoperative corneal endothelial cell count ……
8. Preoperative VA ……
9. Postoperative corneal endothelial cell count on;
   a) Day 1 ……..
   b) One month ……
10. Postoperative ocular examination

    a) Day 1

    VA …….. AC ……

    Conjunctiva ……. Pupil …….

    Cornea …… IOL ……..

    b) One month

    VA ……. Conjunctiva ……..

    Cornea …… AC ……

    Pupil …… IOL …….

    Fundus …….

11. Cause of poor visual outcome if any ……

Appendix II

Questionnaire: Swahili Version

Namba ya dodoso: …….. Namba ya Jalada ……

Namba ya simu: …….

1. Umri ……
2. Jinsia a) Kiume b) Kike
3. Kiwango cha Elimu
   a)Msingi b) Sekondari c)Elimu ya juu d) Nyinginezo (Taja) ……
4. Kuna maumivu yoyote kwenye macho
   b) Ndiyo b) Hapana
5. Hali ya lenzi (kiwango cha mtoto wa jicho) ……
6. Aina ya upasuaji ambao mgonjwa atafanyiwa
   a)MSICS b) phaco
7. Kiwango cha chembe chembe za kioo cha jicho kabla ya upasuaji……
8. Uwezo wakuona kabla ya upasuaji …….
9. Kiwango cha chembe chembe za kioo cha jicho baada ya upasuaji
   a)Baada ya siku 1 ……..
   b)Baada ya mwezi mmoja ……
10. Uchunguzi wa macho baada ya upasuaji

    a) Baada ya siku 1

    Uwezo wa kuona …….. Chumba cha mbele……

    Konjanktiva ……. Mboni …….

    Kioo ch a jicho …… IOL ……..

    b) Baada ya mwezi mmoja

    Uwezo wa kuona …….. Chumba cha mbele……

    Konjanktiva ……. Mboni …….

    Kioo ch a jicho …… IOL ……..

    Fundus ……

11. Sababu za uoni hafifu baada ya upasuaji kama zipo ……..

Appendix III

Informed Consent Form: English Version

MUHIMBILI UNIVERSITY OF HEALTH AND ALLIED SCIENCES

SCHOOL OF MEDICINE,

Department of ophthalmology

INFORMED CONSENT FORM

ID-NO | | | | | | | | | | | |

Consent to participate in a research study

- Greetings. I a m Dr Ng'hungu Lufunga , a postgraduate student, doing a masters of medicine in ophthalmology at Muhimbili University of Health and Allied Sciences. Iam doing research with the objective of evaluation of the corneal endothelial cell loss after manual small incision cataract surgery and phacoemulsification among patients seeking eye care services at Muhimbili National Hospital, Dar es Salaam

Purpose of the study: To evaluate the corneal endothelial cell loss after manual small incision cataract surgery and phacoemulsification among patients seeking eye care services at Muhimbili National Hospital, Dar es Salaam

Participants of the study

All adult patients with cataracts undergoing either manual small incision cataract surgery or phacoemulsification will participate in the study. Participants will have ocular examinations whereby non invasive procedures will be used.

Confidentiality

All the participants who will join the study their names will not be required but will be identified by use of number. The information obtained during data collection will be kept under strict locked environment where it is only the researcher will have access and will be destroyed after the dissertation have been submitted and accepted for the award of my postgraduate degree.

Risk

No harm is expected to occur because of joining in the study.

Benefits

The results of this research will help to improve the quality of care of these patients.

Right to withdrawal

Joining in this study is completely your choice. You can withdrawal at any particular moment even after signing the consent form. You can even refuse to respond to any question in the questionnaire or review guide.

Whom to contact

In case of any concern or question about the study you can contact the researchers, Dr Ng'hungu Lufunga,Dr Anna Sanyiwa,Dr John Kisimbi and Prof Kinabo Nicholas at Muhimbili University, P.O. BOX 65001, Dar es Salaam. You can also contact Prof M. Aboud the Chairperson of the Muhimbili University Senate Research and Publications Committee, P.O.BOX 65001, Dar es Salaam, for any matters concerning ethical violation of the study.

I ……..have read the contents in this form. My questions have been answered and I agree to participate in this study.

Signature of participant……..

Signature of researcher/research assistant…….

Appendix IV

Informed Consent Form: SWAHILI VERSION

Salaam,mimi ni Dr Ng'hungu Lufunga,mwanafunzi wa shahada ya uzamili ya udaktari,Idara ya macho,chuo kikuu cha Afya Muhimbili.Nafanya utafiti kuangalia kiwango cha upotevu wa chembe chembe za kioo cha jicho wakati wa upasuaji wa mtoto wa jicho, hospitali ya taifa Muhimbili.

Lengo la utafiti

Kuangalia kiwango cha upotevu wa chembe chembe za kioo cha jicho wakati wa upasuaji wa mtoto wa jicho, hospitali ya taifa Muhimbili.

Usiri

Majina ya washiriki wa utafiti huu hayatahitajika badala yake zitatumika namba za utambuzi. Habari zote zitakazo kusanywa wakati wa utafiti zitatumika kwa wahusika wa utafiti tu na zitaharibiwa baada ya ripoti ya utafiti kuwa imekubalika kwa ajili kutunukiwa shahada ya uzamili.

Washiriki wa utafiti

Wagonjwa wote wenye mtoto wa jicho watakaofanyiwa upasuaji wa mtoto wa jicho watashiriki kwenye utafiti huu. Washiriki watafanyiwa uchunguzi wa macho wa kawaida na kwa kutumia vifaa ambavyo havihatarishi hali ya macho.

Madhara

Hakuna madhara yanayotarajiwa kwa washiriki wa utafiti.

Faida

Matokeo ya utafiti huu yatasaidia kuboresha huduma za macho kwa wagonjwa wenye mtoto wa jicho na jamii kwa ujumla.

Haki ya kujitoa

Ushiriki katika utafiti ni wa hiyari, mshiriki yoyote ana haki ya kuamua kujitoa katika utafiti wakati wowote kujitoa hakutaathiri kiwango cha huduma kwa mgonjwa.

Mawasiliano

Kama kuna shida yoyote au maswali kuhusu utafiti huu unaweza kuwasiliana na watafiti Dr Ng'hungu Lufunga, Dr Anna Sanyiwa, Dr John Kisimbi na Prof. Kinabo Nicholas wa Chuo Kikuu ch Muhimbili S.L.P. 65001, DSM. Pia unaweza kuwasiliana na Prof M. Aboud mwenyekiti wa kamati ya utafiti na machapisho Chuo Kikuu cha Muhimbili, S.L.P.65001, DSM.

Mimi …….nimesoma na kuelewa maelezo yaliyo kwenye fomu hii na maswali yangu yamejibiwa na ninakubali kushiriki kwenye utafiti huu.

Sahihi ya mshiriki……..

Sahihi ya mtafiti/mtafiti msaidizi…….