Study of Endothelial Cell Count in Patients after Penetrating Keratoplasty by Clinical Specular Microscope

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Abstract: Title: To study the endothelial cell count in patients after penetrating keratoplasty by clinical specular microscope. Purpose: To analyse postoperative endothelial cell density (ECD) and its rate of decline postoperatively after one week, one month, 3 months and 6 months following penetrating keratoplasty. To study the various factors associated with the decrease in endothelial cell loss for better postoperative outcome. Materials and Methods: A prospective, observational study was carried out on 50 adult patients admitted for penetrating keratoplasty having various corneal pathologies. Pre-operative history taking, assessment of visual acuity, detailed anterior segment examination including intraocular pressure and preoperative endothelial cell count was attempted in all the patients. A penetrating keratoplasty was done in all the patients and the patients were followed up on post-operative first week, first month, third month and sixth month. Results: There was a significant Endothelial Cell Count (ECC) loss post a major procedure like Penetrating keratoplasty ranging from 6.45% to 15.47% at the end of 6 months when compared to the annual 0.6% loss of ECC in normal eyes. Conclusion: Re grafts/re-penetrating keratoplasties show a larger margin of ECC loss. Grafts of donors <45 years of age, phakic donors and smaller donor size of <8.5 mm was associated with lower ECC loss. Additional per-op procedures like synechiolysis, corneal suture removal, anterior vitrectomy showed a lower ECC count at the end of 6 months. Post-operative procedures like laser membranectomy had no significant role in the loss of ECC.

Keywords: Endothelial cell count (ECC), Penetrating keratoplasty, Clinical specular microscope.

1. Introduction

Corneal transplantation or keratoplasty is one of the most common transplantation surgeries performed in humans. Keratoplasty is the main treatment modality for visual rehabilitation in eyes with deteriorated corneal clarity. Although newer modalities of keratoplasty have come into play but penetrating keratoplasty (PK) still remains the gold standard in corneal conditions involving full thickness of the cornea¹. PK is a full-thickness transplant procedure, in which a trephine of an appropriate diameter is used to make a full-thickness resection of the patient’s cornea, followed by placement of a full-thickness donor corneal graft. Interrupted and/or running sutures are placed in radial fashion at equal tension to minimize post-operative astigmatism. Later, the sutures are removed selectively to reduce the amount of astigmatism present.

Indications for Penetrating Keratoplasty² are –

1) Optical keratoplasty is performed to improve vision. Important indications include bullous keratopathy, keratoconus, various dystrophies like Fuchs corneal dystrophy, posterior polymorphous corneal dystrophy, degenerations, endothelial failure, corneal endothelial dysfunction and scarring.

2) Tectonic grafting may be carried out to restore or preserve corneal integrity in eyes with severe structural changes such as stromal thinning and descemetocele.

3) Therapeutic corneal transplantation may afford removal of diseased corneal tissue in eyes unresponsive to antimicrobial therapy.

The primary goal after corneal transplantation is preservation of a clear graft which is maintained with the help of corneal endothelium. PK can visually rehabilitate many of those who suffer from visual impairment due to corneal diseases. After successful PK, the transplanted corneal endothelial cells remain viable as a true chimera for years. Corneal endothelial cell density is a commonly reported indicator of the outcome for corneal grafts clarity.²

Specular microscopy is a non-invasive photographic technique that allows you to visualize and analyse the corneal endothelium. Using computer-assisted morphometry, modern specular microscopes analyse the size, shape and population of the endothelial cells. The instrument projects light onto the cornea and captures the image that is reflected from the optical interface between the corneal endothelium and the aqueous humour. The reflected image is analysed by the instrument and displayed as a specular photomicrograph. In clinical practice, specular microscopy is the most accurate way to examine the corneal endothelium. In this study a constructive analysis is done on the endothelial cell count post penetrating keratoplasty and the various factors affecting the cell loss pre and post operatively.

2. Objectives

1) To study postoperative endothelial cell density (ECD) and its rate of decline postoperatively after one week, one month, 3 months and 6 months following penetrating keratoplasty.

2) To study the various factors associated with the decrease in endothelial cell loss.
3. Materials and methods

This was a prospective, observational study done with the approval from Institutional Ethics Committee. In the current study, 50 cases, admitted on the basis of inclusion and exclusion criteria discussed below and operate d for penetrating keratoplasty between the period July 2016 to July 2018, were analysed for ECC by clinical specular microscope.

Inclusion criteria:
Using non-contact specular microscopy, endothelial cell counts were obtained at one week, one month, 3 months and 6 months in 50 patients who underwent penetrating keratoplasty with or without additional procedures at Department of Ophthalmology, P.D.U. GOVT. Medical College, Rajkot. These included patients with
- Corneal opacities
- Failed grafts following various keratoplasties
- Bullous keratopathy
- Corneal dystrophy
- Corneal ulcer

Exclusion criteria:

a) Systemic
- Death of unknown cause
- Death with neurologic disease of unestablished cause
- Active viral encephalitis
- Hepatitis b surface antigen positive donors
- HTLV-I AND HTLV-II infection
- Hepatitis C seropositive donors
- Acquired immunodeficiency syndrome or HIV seropositivity
- Rabies
- Active viral hepatitis
- Creutzfeldt-Jakob disease.
- Subacute sclerosing panencephalitis
- Progressive multifocal leukoencephalopathy
- Reye’s syndrome
- Death from unknown cause including unknown encephalitis
- Congenital rubella
- Active septicemia including endocarditis
- Acquired immunodeficiency high risk behavioral features including homosexuals, intravenous drug abusers, prostitutes and hemophilics
- Leukemia (blast form)
- Lymphoma and lymphosarcoma.

b) Ocular
a. Intrinsic eye disease
b. Prior refractive procedures

All the cases underwent a detailed history taken pre-operatively and a detailed pre-operative as well as post-operative (1st week, 1st month, 3rd month and 6th month) ocular examination. It consisted of assessing Unaided Visual Acuity (VA), Anterior segment examination with a Slit Lamp Biomicroscope; and endothelial cell count with a clinical specular microscope. An written informed valid consent was taken before undergoing penetrating keratoplasty.

4. Results and Analysis

In the current study maximum number of penetrating keratoplasties (46%) took place in the range of 41-60 age group. Pseudophakic bullous keratopathy (36%) and corneal opacity (26%) were the most common indications included in this study. Triple procedure (46%) and pseudophakic pkp(40%) constituted most of the procedures in this study as compared to phakic pkp(2%), aphakic pkp(2%) and re-pkp(10%). The majority of the donor tissues taken in this study were under 45 years of age (88%).

Furthermore, endothelial cell counts (ECC) were compared between the values obtained at the end of 6 months with that obtained at the end of the 1st month. Optical repkp constituted the maximum average ECC loss (14.20%) and aphakic pkp the minimum average ECC loss (8.53%).

Maximum average ECC loss was seen in donors above 50 years of age. (12.44%).

Pseudophakic donors (13.41%) had a greater ECC loss compared to phakic donors (10.89%). It was also noted that there was no significant variation in the ecc loss when the death to enucleation time was within 6 hours.

In the current study donors graft size of 8.5(10.79%) or less constituted a lower ECC loss as compared to a larger donor graft size of 10.5(12.43%) or more. Procedures like...
synchondrosis, corneal suture removal and anterior vitrectomy had a significant role in the loss of ECC as compared to a simple procedure involving betadine wash only.

In our study we noted that per vasculature proximity and retention of major portion of end of the study. This is related to the fact that smaller grafts have low chances of allograft rejection due to less limbal vasculature proximity and retention of major portion of healthy endothelium.

In our study we noted that per-operative procedures like synechiolysis (11.54%), corneal suture removal (12.06%) and anterior vitrectomy (12%) had a significant role in the loss of ECC as compared to a simple procedure involving routine basic surgical procedure (10.8%) or iv mannitol (10.8%) only. This signified that complicated cases had a greater chance of post-operative inflammation and damage to endothelial cell which led to poor graft survival when compared to routine penetrating keratoplasties. It was also noted that such procedures majorly took place in re graft procedures(re-pkp) which also showed maximum endothelial cell loss among all procedures. We also found no direct correlation between laser membranectomy procedure and loss of endothelial cell count.

5. Discussion

After successful penetrating keratoplasty, the transplanted corneal endothelial cells generally remain viable as a true chimera for years. As such, they remain in a hostile environment under the constant threat of rejection by a host on which they must depend for sustenance.

In this study it was noted that the Re-pkp (14.20%) involved the highest percentage loss of average endothelial cell count (ECC) after 6 months. This may be attributed to allogenic graft rejection in regraft cases. The studies by Nishimura et al,[5] Bourne et al,[6] and Patel et al[7] have shown that the greatest cell loss after penetrating keratoplasty occurs initially, with 30% to 50% loss in the first year compared to preoperative counts, and then declines gradually as the endothelium stabilizes. We found that the age of the donor paid a significant role in the postoperative loss of ECC at the end of the study. While donor age >50 showed a considerable loss of 12.44 % when compared to the first month, the donors younger than 50 years should a reduced cell loss ranging from 8.33-11.66%. Daniel Böhringer et al found donor age was to have a significant negative effect on the long-term endothelial prognosis.[8]

The study did not show a strong correlation between death to enucleation time and postoperative ECC loss. In this study we found that smaller graft size of 8.5 mm had a lower postoperative cell loss of 10.79% while larger graft size of 10.5 mm had a relatively higher cell loss of 12.43% at the end of the study. This is related to the fact that smaller grafts have low chances of allograft rejection due to less limbal vasculature proximity and retention of major portion of healthy endothelium.

6. Conclusion

1) There is a significant Endothelial Cell Count (ECC) loss post a major procedure like Penetrating keratoplasty ranging from 6.45% to 15.47% at the end of 6 months when compared to the annual 0.6% loss of ECC in normal eyes
2) Re grafts/re-penetrating keratoplasties show a larger margin of ECC loss compared to other modifications like pseudophakic pkp, phakic pkp, triple procedure and aphakic pkp.
3) Grafts of donors <45 years of age showed a lower ECC loss. So also, phakic donors had a lower ECC loss as compared to pseudophakic donors.
4) Smaller donor size of 8.5 mm was associated with lower ECC loss as compared to larger grafts
5) Keratoplasties which required additional per-op procedures like synechiolysis, corneal suture removal, anterior vitrectomy showed a lower ECC count as observed at the end of 6 months. Post-operative procedures like laser membranectomy had no significant role in the loss of ECC.

References

[8] Influencing factors on chronic endothelial cell loss characterised in a homogeneous group of patients

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