Corneal Graft Rejection; Prevalence and Risk Factors

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Abstract

Background: Rejection is the most common cause of late corneal graft failure. The purpose of this study is determining the prevalence, and defining the role of those factors which may affect the risk for rejection.

Methods: The study was conducted on all patients who had penetrating keratoplasty without immunosuppressive therapy with at least 1 year follow up. Demographic status, cause of graft, and types of rejections were detemined. Contribution to graft rejection, persistence of transparency, and rate of failure due to rejection were considered for each factor. Statistical analysis was performed by multivariate regression and ANOVA tests.

Results: Two hundred ninety five penetrating keratoplasties were performed on 286 patients (61% male, 39% female). Mean age at keratoplasty was 38 ± 20 years. Mean follow-up period was 20 ± 10 months. Graft rejection occurred at 7.3 ± 6 months (20 days to 39 months) in 31.8% of cases. The most common type of rejection was endothelial (20.7%). Contributing factors were extent of corneal vascularization, regrafting, anterior synechiae, irritating sutures, active inflammation at the time of surgery, additional anterior segment surgery, history of trauma, uncontrolled glaucoma, history of prior graft rejection, recurrence of herpes infection on graft, and eccentric graft ($P<0.05$). Patients' age, size of donor cornea, and bilateral transplantation were not significant.

Conclusions: The extent and severity of corneal vascularization, anterior synechiae, irritating sutures, active inflammation, regrafting, additional surgery, trauma, uncontrolled intra-ocular pressure, history of graft rejection,

recurrence of herpes, eccentric grafting and corneal scarring on graft are significant risk factors for graft rejection.

INTRODUCTION

Penetrating keratoplasty (PKP) is the most common human organ transplantation, which is performed for various corneal disorders.[1] In 2000, more than 2100 corneas provided by the Central Eye Bank of Iran were used for corneal transplantations. In the recent two decades, due to improvements in surgical techniques, operating microscopes, suture materials, advanced methods of cornea preservation, use of steroids and immunosuppressive agents corneal transplant success rate has significantly increased.[1,2] Despite these advances, rejection of donor endothelium by the recipient immune system is the leading cause of corneal transplant failure.[2,3,4]

In 1948, Paufique et al, introduced the term "maladie du greffon" (graft disease) for graft opacification following an initial period of transparency.[5] In 1951, Maumenee et al. demonstrated that the basis for graft rejection is host sensitivity to donor cornea antigens.[6] Khodadoust et al. showed immunologic reactions can occur in each layer of the cornea (epithelium, stroma, and endothelium), possibly leading to rejection of a single layer or different layers in concert.[7] In the studies mainly by Sondo et al. it was shown that anterior chamber immune deviation (ACAID) is probably responsible for the preservation and survival of the graft, and graft rejection is known to be due to loss of equilibrium in the ACAID system.[8]

Reported prevalence of corneal graft rejection ranges from 2.3 to 68 percent in different studies.[9,10] At least one episode of corneal rejection may occur

in 30% of grafts, 12 percent with good prognoses and 40% with poor prognoses eventually leading to complete loss of corneal transparency.

Graft rejection most commonly occurs between 4 and 18 months following transplantation, but may occur any time after the surgery. In one report, 53.3% of rejections occurred during first year.[9]

Various risk factors for graft rejection have been studied including loose sutures, early suture removal,[11] extent and severity of recipient cornea vascularization,[2,11] increasing number of regrafts,[12,11] bilateral graft,[12,13] anterior synechia (recipient iris adhesion to graft tissue),[14] ocular inflammation at the time of transplantation, young age of recipient,[11] size of donor cornea,[13,14] graft eccentricity, uncontrolled glaucoma [15] , atopic dermatitis, and dry eye states.[16,17] Prompt diagnosis and treatment of rejection will frequently result in recovery and clearance of corneal graft. Eighty-seven percent of rejections are symptomatic.[9] About 50% - 70% of cases of corneal graft rejections can be controlled with topical steroids, but in some severe cases oral steroids might be needed.

Considering the importance of graft rejection and subsequent failure, it seems necessary to investigate the role of causative factors which increase the risk of graft rejection. In this study performed at Labbafinejhad Medical Center from 2003 to 2005 on patients who had corneal transplantations with at least 1 year of follow up, prevalence, clinical signs, and factors influencing rejection were evaluated.

MATERIALS & METHODS

In this cohort study, between June 2003 and September 2006 all patients having corneal transplantation were sequentially included. Only those with

follow-up visits longer than one year were included. Patients with immunosuppressive therapy due to systemic or ocular diseases were excluded. Preoperative complete ocular examinations including uncorrected and corrected visual acuities, slit-lamp microscopy, intra-ocular pressure measurement and funduscopy were performed. All patients' data before and after surgery were recorded. If graft rejection was diagnosed, relevant data including type of rejection, detectable risk factors, causative factors, method and result of treatment, and final corneal transparency were recorded.

All surgeries were performed under general anesthesia. The corneas harvested either from whole globes (preserved in cold wet chambers) or corneoscleral rims, preserved in media for 2 ± 1.9 days (Likoral, Chauvin, France. or Optisol GS, Bausch & Lomb, USA.). The size of trephination was chosen to remove most of the recipient corneal pathology, sparing at least 1-1.5mm peripheral corneal rim for suturing. Barron- Hessburg vaccum trephines (Katena, Denville, NJ, USA) with endothelial punches were applied to cut the recipient and donor corneas. Donor-recipient disparity was considered 0.25-0.5mm. Four cardinal interrupted stitches with a 10/0 nylon (CU-1, Alcon laboratories, Fort Worth, TX, USA) were followed by permanent suturing (interrupted, single running, double running, or mixed) of the graft according to surgeon judgment. Finally, the anterior chamber was formed using BSS (Balanced Salt Solution), and the wound was inspected for leakage and proper apposition. The day after the surgery, the eyes were examined in terms of visual acuity, graft clarity, size of epithelial defect, sutures, wound stability, and ocular surface/ anterior chamber inflammation. Topical 0.1%betamethasone drop was started every

6 hours, continued for 6-8 weeks and tapered according to anterior chamber and/or ocular surface inflammation. In case of transplantation due to aphakic or pseudophakic corneal edema, steroid drops were continued 1-2 drops per day along with intraocular pressure control. Oral prednisolone (1-2mg/kg for 7-10 days) was administered in those cases with severe corneal or anterior chamber inflammation. Regular follow-up visits were conducted on days 1, 2 and 3; then every week in the first month, every 2 weeks until 3 month, and subsequently every month in the first year. Afterwards, follow up visits were performed every 3-4 months.

Corneal transparency and absence of any sign of rejection during the first 10 days was a prerequisite for the diagnosis of corneal graft rejection. Clinical diagnosis of various types of graft rejection was made based on biomicroscopic findings. Epithelial rejection was defined as the presence of an epithelial line which may be moving on sequential close follow-up visits and stained with fluorescein. Subepithelial rejection was defined as the occurrence of multifocal infiltrative lesions with no other inflammatory signs in the superficial stroma (Fig 1). Local endothelial rejection was the presence of white keratic precipitates in a limited area of the donor corneal endothelium which may be associated with a mild anterior chamber reaction. Diffuse endothelial rejection was defined as the presence of endothelial rejection line (Khodadoust line), and/or a high number diffuse KPs which might be associated with corneal edema (Figs 2 & 3).

Epithelial or subepithelial graft rejections were treated with topical 0.1%betamethasone drops 4-6 times per day and were gradually tapered off. In milder forms of endothelial rejection betamethsone drops were initiated

on a more frequent basis (every 1 hr.). In severe cases, oral prednisolone 1-2mg/kg was added. Subtenon injection of methylprednisolone (40mg) was considered when despite frequent steroid treatment there was no sign of improvement after 3 days.

Based on biomicroscopic and retinoscopic findings, clarity of transplanted corneas were classified. If the cornea was completely transparent with no edema or opacity, the iris and anterior chamber details were clearly observed, and the eye was easily refractable, it was considered as clear. If corneal edema or opacity was present, iris and anterior chamber details were not clearly visible, and the eye was barely refractable, it was considered as hazy. If corneal edema was severe such that iris and anterior chamber details were barely visible, and refraction was impossible, it was considered as opaque. Those hazy and opaque corneas which were not responsive to medical therapy were considered as failures.

For each risk factor two groups of eyes with and without the factor categorized. For statistical analysis of data using SPSS (version 10) was used for applications Multi-variate regression analysis, and ANOVA tests.

Results

Two hundred ninety five corneal transplantations in 286 patients were included (61.4 % male, 38.6 % female). Mean age at surgery was 37 ± 20 years (40 days - 90 years). Mean follow-up period was 20 ± 10 months (24 - 43 months). Keratoconus (31.9%) was the most common indication for transplantation, followed by regraft (13.9%), infectious keratitis (12.6%) (perforated or non-perforated), trauma scars (7%) and sequelae of chemical burns (2%). Penetrating keratoplasty was optical in 79.7% and tectonic in 15.9% of cases, respectively.

Lamellar keratoplasty was performed in 4.4% of cases. Donor corneas provided either from whole globes (71.9%) or from corneoscleral rims (28.1%) of cases. There has been no correlation between duration of preservation and occurrence of rejection (P> 0.5).Diameter of donor cornea was ≤8mm in 62.8% of cases and >8mm in the remainder.

Clarity based on the cause of transplantation

At the end of follow-up, all cases of congenital hereditary endothelial dystrophy (CHED), 97.8% of keratoconic cases, 96.3% of other corneal dystrophies, 66.7% of pseudophakic bullous keratopathies, 61.5% of aphakic bullous keratopathies, and 60% of corneas with vascularized scars had clear corneas. Forty seven percent of corneal regrafts, 28% of corneas with traumatic scars, and 16.7% of corneas with scars due to chemical burns had clear grafts.

Corneal graft rejection

Rejection was seen in 94 grafts (31.8%). It occurred once in 20.8% of cases, twice in 7.8%, three times in 2%, and more than 3 times in 0.9% of cases. The mean interval between transplantation and first rejection reaction was 7.3 ± 6 months (20 days - 39 months). Sixty three percent of rejections occurred within the first 6 months after the surgery, and 87.2% within one year. The most common type was endothelial rejection (20.7%). In 6.1% of rejections all corneal layers were involved. Subepithelial infiltration was seen in 3.1% of cases. At last follow-up visit, 70.6% of corneas were clear. Graft failures were due to rejection (6.1%), early decompensation (7.1%), and late decompensation (11.6%) of cases. Most frequently, rejections occurred in April (4.1%), followed by July (3.4%), September (3.4%), and November, December, and January each (each 1.7%).

Risk factors for graft rejection

Age: Rejection occurred in 28.4% of patients under the age of 40 years versus 37.8% in older cases, but the difference was insignificant (table 1). There was a meaningful correlation between patient age and corneal transparency (P<0.001), i.e., older the patient higher the chance for graft remaining clear.

Corneal vascularization: Corneal vascularization was present in 29.1% of cases. Rejection was observed in 27.8% of patients without corneal vascularization. This raised to 38.9% in those with mild corneal vascularization, 48.3% in those with moderate, and 38.5% with severe corneal vascularization (P= 0.05). In the last follow up examination, 78.9% of corneas without vascularization were clear. This rate was 77.8 %, 62.1 %, and 30.85% in corneas with mild, moderate, and severe vascularization, respectively. In the non-vascularized group, failure was due to rejection in 4.8% of cases, but in the vascularized group it was 13.8%, which was proportionate to increased vascularizton (P<0.001).

Regraft: Rejection developed in 48.6% of regrafted cases in comparison to 29.5% of cases without regraft (P=0.025). In regraft group, 47% of corneas remained clear, while this rate was 73.8% in those without regraft. Failure rate in the regraft group was 18.8% while 4.6% in the other (P<0.001).

Corneal graft of the second eye

Bilateral graft was performed in 17.5% of all cases. Rejection rate was 27.5% and 32.8% in unilateral and bilateral groups, respetively (P<0.001). At the end of follow-up, 98% of bilateral cases and 65.2% of unilateral ones had clear grafts (P<0.001). Failure rate in unilateral group was 7.4%, while no cases of failure occurred in bilateral group (P<0.001). Considering only keratoconus patients, there was no statistical significant difference in terms of graft rejection between unilateral and bilateral grafts.

Size of donor cornea

Rejection rate was the same for donor corneal diameters smaller or larger than 8mm. At the end of follow-up, 73.7% of the larger than 8mm group and 69.5% of the smaller group had clear grafts. The rate of rejection was 6.0% and 6.3% in two groups, respectively, which was not statistically significant

Presence of anterior synechia

Anterior synechia was present in 11.5% of cases. The rejection rate was significantly different between groups with or without synechiae (table 1). At the end of follow-up period, 44.1% of those with anterior synechiae and 74.3% of those without had clear grafts ($p=0.005$). Failure rate due to rejection was 49.2% versus 20.6% in cases with and those without anterior synechiae, respectively ($p<0.005$).

Presence of irritating sutures

Loosening or breakage of at least one suture was seen in 7.8% of cases. Rejection rate was 78.3% versus 28.1% in groups with and those without these events ($p<0.001$) .

Presence of active inflammation

Active inflammation of the cornea or anterior segment of the eye was present in 10.1% of cases at the time of surgery. Compared to the group without inflammation, incidence of rejection was definitely more ($p=0.0035$) (table 1). Clear grafts were present in 75.5% versus 30% in groups with and those without inflammation. Also, the rate of failure due to rejection was 20% versus 4.5% in groups with and those without inflammation, respectively ($p<0.001$).

Additional surgical procedures

Intra-ocular anterior segment surgeries (other than penetrating keratoplasty) was performed in 10.2% of cases simultaneously or later. The rate of rejection was 50% in

this group (p=0.022). At the last follow up, clear cornea was seen in 72.3% of those without versus 57.1% of those with additional surgeries (p=0.002).

History of trauma

Corneal scar secondary to trauma was the cause of corneal graft in 11 cases (3.7%). Rate of rejection was 63.6% in this group (p=0.02). At the end of follow-up, 28.6% of these grafts remained clear (p=0.001). Failure rate due to rejection was 14.3% in this group (p<0.001) (table 1).

Uncontrolled intra-ocular pressure at the time of transplantation

Fifteen cases (5.1%) had uncontrolled intra-ocular pressure at the time of transplantation. Rejection occurred in 60% of this group (p=0.02). Only 15.4% of grafts remained clear and 7.7% had failures due to graft rejection which was not statistically significant (table 1).

Eccentric graft

Nine cases (3.1%) had eccentric grafts. Rejection rate was 66.7% in this group versus 30.7% in those with central graft (p=0.038).

History of graft rejection in the same or fellow eye

Thirteen cases (4.4%) had such a history. Of these, 69.2% had graft rejections (p=0.002). Rate of clear corneas was not significantly different between the two groups, but failure due to rejection was 15.4% compared to the group without a previous history of rejection (p=0.001).

Occurrence of ulcer in corneal graft

In 5 cases (1.7%), keratitis occurred on the corneal graft, 40% of them experienced graft rejection (p=0.02). Rate of clear cornea and failure due to rejection was 60% and

20%, respectively (p<001). Furthermore, 9 cases (3.1%) had recurrence of herpes on the graft, of them 66.7% had graft rejections (p=0.021).

Yag laser posterior capsulotomy

This procedure was performed only on 1 case, no rejection took place and cornea remained clear at the end of follow up.

DISCUSSION

Keratoconus is the most common cause of corneal transplantation in our center, but the rate of regraft has increased from 7.2% to 13.9%.[18] The reason for this could be more common usage of immunosuppressive drugs which enable the surgeons to do corneal transplantation (including regrafts) in high risk cases.[19,20] On the other hand, decrease in rate of corneal ulcers leading to corneal transplantation from 15.3 to 12.6% could be due to better medically controlled in these cases. In multiple studies primary indications for PKP are significantly similar to our study.[10,20,21,22,23] In Aiken-O'Neil report, the most common cause for corneal transplantation was bullous keratopathy (22.3%) followed by regrafts (21.1%), Fuch`s endothelial dystrophy and finally keratoconus.[24]

Endothelial rejection is the most common type of rejection in our study (20.7%). The incidence of endothelial rejection has been reported from 12% to 44% in different studies, [10, 22, 25] which might be due to differences in population type, indication for PKP, criteria for rejection, steroid and immunosuppressive usage. In our report, SEI was observed in 3.1% of cases. It's rate has been reported from 2.4% to 2.8%. [2,22] Subepithelial infiltrations may be diagnosed less often due to their temporary nature and low influence on visual acuity.

In our study, rejection rate was the same in cases under and above 40 years of age, which is opposite to the findings of Maguire[15] and Vail[26]. In an extensive report on

2800 cases, age over 40 was introduced as a risk factor for rejection and graft failure.[27] In another study, older age was considered as a risk factor for graft rejection.[14] It is postulated increasing age causes decrease in acute rejection due to both immunosenescence and decreased resistance to immunosuppressive drugs.[28] This marked difference could be the result of differences in patient population and primary indications for corneal transplantation. Knowing that keratoconic patients (the most common cause of corneal transplantation and graft rejection in our study) are transplanted in an earlier age compared to bullous keratopathy patients (the most common cause of corneal transplantation in other studies), findings different from those of the other studies can be explained. In Vail's study, in which donor and recipient age were nearly the same, they showed recipient factors such as primary diagnosis, prior transplantation, and glaucoma as risk factors, but they couldn't find the recipient age as a risk factor for rejection.[26] In Simon`s study risk of rejection has decreased when storage of donor cornea has increased more than seven days[29] but in our study rejection rate had no corelation with duration of preservation.

We found direct correlation between extent and severity of corneal vascularization with graft rejection. Alldredge et al. found that increased vascularization increases endothelial rejection incidence but not SEI and epithelial rejection.[10] In Yamagami [30] and Maguire [15] reports there was a positive correlation between rejection rate and extent of vascularization. Severe vascularization in all four quadrants of the recipient cornea increases the risk of graft failure by 1.7 times compared to a non-vascularized recipient cornea.[15] The primary cause of vascularization in the recipient cornea may itself induce rejection, and therefore, be effective in the survival of graft.[19]

More common usage of immunosuppressive drugs (e.g., cyclosporine) in high risk patients has improved survival of regrafts and led to perform more corneal

transplantations. In Maguire's study, the risk of graft rejection and failure after regrafts increased three times.[15] In none of the other studies the role of regrafts in increasing the risk of rejection has been denied.[10,26] This can be due to increased interaction between recipient and donor immune systems. After each regraft this immune system sensitization increases which accelerates immune reactions and graft rejection. Improvement of graft prognosis in penetrating normal - risk keratoplasty by HLA class 1 and 2 matching is suggested by some authors.[31]

If all cases of corneal transplantation are considered, second eye graft has a better prognosis. There were less rejection-induced graft failures and more durable clear grafts in second eye cases. Musch et al. reported the rate of rejection in second eye of bilateral grafts was the same as the first eye 1 and 5 years after the surgery.[12] However, Donshik reported a rejection rate of 27% in bilateral cases versus 13% in unilateral ones.[32] In keratoconus, rejection rate was statistically the same between unilateral and bilateral grafts.[18] Tuft demonstrated the irrelevance between bilateral graft and graft survival. [13] According to Musch report, when there is no rejection in the first eye, the risk for rejection in the second eye does not increase, but if rejection does occur in the first eye, the risk increases for the second.[12]

In our study, the size of donor cornea (≤8mm or >8mm) had no effect on graft rejection and subsequent failure. In some studies, donor corneas larger than 7.5mm had a greater risk of rejection.[13,17] However, in Vail's study the greater size of donor cornea had greater risk of rejection with a lower chance for failure. [26] Trigui showed increased rejection with corneal grafts ≥ 8 mm and with change in suturing technique.[14] In CCTS study, corneal size less than 8mm had a greater risk of rejection[15]. A larger donor cornea increases the risk of rejection due to transfer of a greater number of Langerhans cells (which are found more frequently in the periphery

of cornea which contain HLA-DR antigens), and their proximity to limbal vessels.[22,32] In CCTS study, surgeons were free to choose different sizes of grafts and suturing methods, and therefore smaller size grafts with interrupted sutures for high-risk grafts were "confounding by indication".[15] Thus, primary indication for penetrating keratoplasty (which had obliged a smaller donor cornea and interrupted suturing technique) is of greater relevance to the occurrence of rejection than is corneal graft size, which is actually a secondary issue. Therefore, for determinig the role of graft size in the occurrence of rejection, a study excluding primary corneal disease and other variables should be designed. On the other hand preservation of donor cornea more than seven days has decreased risk of rejection and prolonged graft survival.[29]

In our study, anterior synechiae between the iris and donor cornea increased the risk of rejection, and its consequent failure. Because of the easier access of blood vessel to donor endothelial cells, rejection reactions are accelerated.[15] Synechiae cause direct damage to adjacent donor endothelial cells and also raise the probability of elevations in intraocular pressure and subsequent glaucoma.[30] In all studies, anterior synechiae were shown to be as a risk factor for rejection, failure and glaucoma.[15, 22, 30, 26]

In current study, presence of irritating sutures and anterior segment inflammation was associated with increased risk of rejection. These factors trigger the immune system and cause presentation of Class 2 HLA antigens leading to graft rejection reactions. [22] Anterior segment surgeries at the time of transplantation was a risk factor for graft rejection. These factors also influenced the overall survival of the grafts. In the CCTS study, simultaneous anterior segment surgery at the time of penetrating corneal graft had strong correlation with graft results.[11,15] Effect of prior anterior segment surgery on graft rejection and failure has not been clearly determined,

although this factor has been introduced as a risk factor for corneal graft in Australia.[9,33]

In recent study, history of previous graft rejection was a risk factor for rejection and subsequent failure, but there was no correlation with overall graft failure. Maguire et al. found that with sequential corneal grafts, risk of rejection and subsequent failure, and overall failure increases.[15] Endothelial rejection is the most serious form of rejection, which is associated with local or generalized destruction of endothelial cells of donor cornea. Multiple attacks of rejection followed by incomplete treatment can potentially lead to graft failure. Due to sensitization of recipient immune system to especially HLA Class 2 antigens, corneal regrafts under these circumstances can cause new attacks of rejection.[11,14] Prolonged use of very low dose of topical steroid can cause suppression of local immune system which may prevent recurrence of rejection.[34]

Eccentric penetrating keratoplasty was another risk factor in our study. Although total number of cases was limited, 66% experienced graft rejection. Also, history of trauma and activation of herpetic keratitis on graft caused a meaningful increase in the risk of graft rejection. If chemical burn is considered as a type of corneal trauma, the rate of rejection and subsequent failure is significantly increased.[15] Corneal graft in cases with a positive history of chemical burn should be considered as a high-risk graft.[9,17,22] History of herpetic keratitis and its resultant corneal vascularization can act as a precipitating factors for graft rejection and subsequent graft failure, which has also been shown in present as well as previous studies.[15,22,26,34]

Our study show that increased intra-ocular pressure is a risk factor for graft rejection. Increased intraocular pressure has been considered as an important risk factor for graft rejection and failure. Compared to the normal pressure group, this factor either

as ocular hypertension or glaucoma has been associated with a larger number of rejections, but there was no statistical significant correlation with subsequent graft failure.[11] Although in CCTS report, elevated intra-ocular pressure has been considered as a risk factor for graft failure but not rejection.[15] On the other hand in Yamagami study coexistence of glaucoma and anterior synechiae are defined to be a major risk factor for graft rejection,[30] According to Naache et al. reversibility of graft rejection is the same between those with and without a history of glaucoma or ocular hypertension.[35] Medical and surgical control of intraocular pressure after graft surgery is difficult, therefore pre-operative control of glaucoma is of utmost importance.

CONCLUSION

Risk factors that are directly involved in the development of corneal rejection and its possible subsequent failure are recipient corneal vascularization, regrafts, anterior synechiae, graft irritating sutures, anterior segment inflammation at the time of transplantation, simultaneous anterior segment surgery, eccentric grafts, history of trauma (especially chemical burns) previous history of rejection, recurrence of herpes, and finally uncontrolled elevated intra-ocular pressures at the time of surgery. In current study, there was no significant correlation between recipient age and donor cornea size with corneal graft rejection.

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Table 1: Risk Factors for Graft Rejection

Risk factor		Relative frequency (%)	Graft rejection (%)	P value	Corneal transparency (%)	Failure rate due to rejection (%)	P value
Age	< 40 yrs	62	28.4	0.095			
	≥ 40 yrs	38	37.8				
Corneal vascularization							
	mild	6.1	38.9	0.015	77.8		
	moderate	9.8	48.3		62.1	13.8	<0.001
	severe	13.2	38.5		30.8		
	without	70.9	27.8		78.9	4.8	
Number of grafts							
	regraft	12.5	48.6	0.025	47	18.8	<0.001
	primary graft	87.5	29.5		73.8	4.6	
Graft in second eye		17.5	27.5	0.035	98		<0.001
Unilateral graft		82.5	32.8		65.2	7.4	
Size of donor cornea							
	< 8mm	29.8	30.5	0.3	73.7	6.3	0.11
	≥ 8mm	67.9	29.8		69.5	6	
Iridocorneal adhesion		6.5	64.7	<0.001	44.1	49.2	0.005
Irritating suture		7.8	78.3	<0.001			
Active intraocular inflammation		10.1	36.7	0.035	30	20	<0.001
Secondary intraocular surgery		10.2	50	0.025	57.1		
Eccentric graft		3.1	66.7	0.038			
Primary trauma		3.7	63.6	0.02	28.6	14.3	<0.001
Uncontrolled intraocular pressure		5.1	60	0.02	15.4	7.7	0.1
History of graft rejection		4.4	69.2	0.002	29.2	15.4	0.001
Ulcer in corneal graft		1.7	40	0.021	60	20	0.001
Recurrence of herpes simplex		3.1	66.7	0.021	-	-	-