EFFECT OF DEATH-TO-PRESERVATION TIME ON DONOR CORNEAL EPITHELIUM

BY **Woodford S. Van Meter MD,*** Douglas G. Katz MD, Harrison White BS, AND Robert Gayheart BS

ABSTRACT

Purpose: Surface disease is one of multiple variables affecting the quality of the postkeratoplasty donor cornea. Trauma to Bowman's layer before and during harvesting can denude the donor epithelium and result in epithelial defects in the donor following penetrating keratoplasty. Eye banks use death-to-preservation (DP) time intervals as long as 18 hours. This study evaluates the effects of higher DP time on the donor epithelium in storage medium and immediately following keratoplasty.

Methods: Eighty-one consecutive corneas were procured by the University of Kentucky Eye Bank, rated by one technician (H.W.), and used by one surgeon (W.S.V.) for elective penetrating keratoplasty. Donor records were retrospectively reviewed for age, DP time, and epithelial condition. All corneas were harvested and evaluated according to Eye Bank Association of America standards. Donor charts were reviewed for DP time and for condition of the epithelium in storage. Recipient charts were reviewed for epithelial defects following keratoplasty.

Results: Average DP time of all 81 donor corneas was 6:18 hours (ie, 6 hours, 18 minutes). Average DP time of 13 corneas with epithelial sloughing was 7:02 (range, 2:01 to 12:25) hours, and nine (69%) had DP time longer than 6 hours. Average DP time of 68 corneas with no sloughing was 6:09 (range, 1:59 to 11:03) hours ($P < .32$). Average DP of 28 recipients with epithelial defect on day 1 was 8:01 (range, 3:41 to 12:49), and average DP in 53 patients with an intact epithelium on day 1 was 5:23 (range, 1:59 to 9:46) ($P < .001$). The percentage of postoperative patients with epithelial defects in the graft on day 1 rose from 14% when DP was less than 4 hours to 100% when DP was greater than 10 hours. Average DP in 13 donors under age 30 was 8.3 hours.

Conclusion: DP time longer than 6 hours was more likely to result in sloughing of the donor epithelium. Care of donor epithelium prior to harvesting becomes increasingly important with DP times longer than 6 hours. Higher-than-average DP times occurred in donors under 30 years of age. Higher DP time results in an increasing likelihood of epithelial defects in the graft. Donor corneas with lower DP time may be important in penetrating keratoplasty ocular surface disease.

Trans Am Ophthalmol Soc 2005;103:209-224

INTRODUCTION

Penetrating keratoplasty is the most common transplant operation in the United States.[1] In 2004, 46,841 corneal transplants using Eye Bank Association of America (EBAA) donor corneas were performed in the United States.[2] The quality of vision of the corneal transplant recipient is determined by the shape of the cornea and graft clarity. A clear graft is expected following keratoplasty, but multiple factors, such as surface disease, endothelial failure, and rejection, can adversely impact graft clarity. Corneal surface disease has been shown to cause significant morbidity, delay visual rehabilitation, and reduce postkeratoplasty acuity.[3] A poor corneal surface can result in decreased visual acuity due to an irregular tear film interface (resulting in a poor refractive surface), discomfort, infectious keratitis, permanent damage to Bowman's layer, and scarring of the anterior stroma.

Corneal surface disease has been observed following keratoplasty since the operation was first performed. Postkeratoplasty epitheliopathy can range from near-normal with a completely clear and intact corneal epithelium to the complete absence of the corneal epithelium with basement membrane damage and exposure. Bron[4] in 1973 noted whirl patterns in the epithelium of postkeratoplasty patients. The vortex patterns of the corneal epithelium[5] and hurricane keratopathy[6] also have been described following penetrating keratoplasty. Stulting and colleagues[7] showed in 1988 that the overall failure rate in patients with the epithelium removed at the time of surgery was higher than that in patients with an intact epithelium, although the absence of the corneal epithelium did not affect graft rejection. Feiz and associates[3] in 2001 reviewed multiple factors that affect graft clarity following keratoplasty and reported that death-to-preservation (DP) time had minimal effect on punctate epithelial keratopathy, graft clarity, or postoperative vision. However, Price and colleagues[8] estimated that as many as 25% of grafts may fail on account of surface problems.

The intact corneal epithelium protects Bowman's layer from mechanical and chemical trauma. However, the intact donor epithelium on the transplanted cornea is ultimately replaced by the recipient's epithelium. Reepithelialization of the donor occurs by transformation of the host stem cell population into a new donor epithelium with mitosis, migration, and hemidesmosome attachments.[9] This process may occur within several weeks in an otherwise healthy denuded cornea, but postoperative transplant patients suffer the additional insults of denervation, topical medications, altered topography, and mechanical trauma to Bowman's layer.[10]

From the Department of Ophthalmology, University of Kentucky, Lexington, Kentucky.
*Presenter.
Bold type indicates AOS member.

Mannis and associates[9] in 1997 evaluated risk factors for surface keratopathy following keratoplasty and found that surface keratopathy occurring months after keratoplasty was not related to DP time or donor epithelial status. However, they noted that corneal surface abnormalities after transplant can be a source of delayed visual rehabilitation for the patient and a genuine threat to the success of the graft. Because other patient factors, such as lid malposition, abnormal blink, and ocular surface disease, can be outside the control of the transplant surgeon, many surgeons recognize that an intact epithelium on day 1 suggests a smoother postoperative course for the graft, especially in high-risk patients with dry eyes, ocular surface disease, inflammatory eye disease, or exposure.

The purpose of this study was to evaluate the effect of DP time on the donor epithelium prior to transplant and subsequently in the recipient patient immediately following transplant. Corneal epithelial status on day 1 can vary from a pristine intact corneal epithelium to a complete epithelial defect with exposure or damage to Bowman's layer. Measures that promote a healthy epithelium immediately following keratoplasty may reduce the likelihood of postoperative epitheliopathy and potentially improve the visual performance and longevity for corneal grafts.

METHODS

Eighty-one consecutive corneal donors procured by one eye bank over a 2-year period and used by a single surgeon were included in the study. Procurement, surgical technique, and postoperative care were consistent regimens. Donor tissue was distributed by the eye bank according to EBAA medical standards or rejected because of medical or social history factors thought to adversely affect the donor cornea, infectious or structural contraindications, opacification or foreign material on slit-lamp examination, and, rarely, by serologic testing.[1] We retrospectively evaluated the effect of DP time on the status of the corneal epithelium, specifically noting whether the epithelium in preservation medium was intact or sloughed (Figure 1). We evaluated the epithelial status in patients following keratoplasty, noting epithelium and the clarity of the graft at day 1 and subsequently until epithelialized (Figure 2). The intent of the study was to see what effect DP time had on the quality of the donor epithelium and what effect the donor epithelium had on recipient surface following keratoplasty.

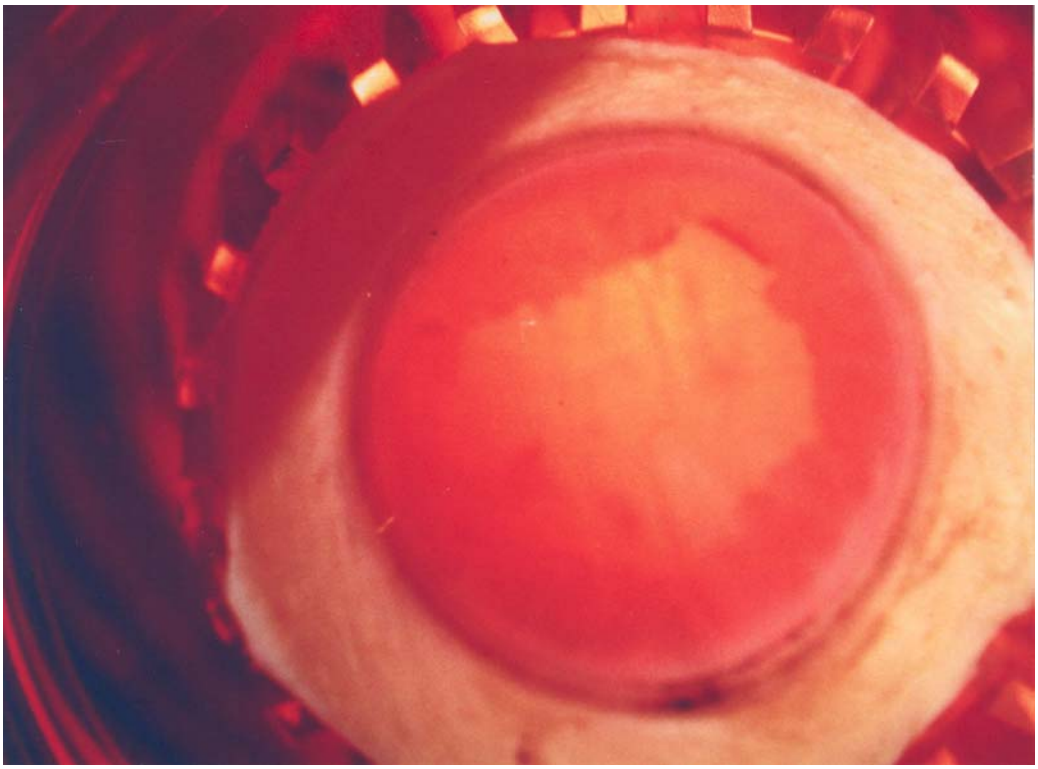

FIGURE 1

Central epithelial defect in donor resulting from exposure prior to placement in storage medium.

Between January 1, 2002, and December 31, 2003, 81 donor corneas were used for penetrating keratoplasty for elective keratoplasty by one surgeon (W.S.V.) (Table 1). Patients underwent penetrating keratoplasty for a variety of indications: pseudophakic corneal edema (40 patients), Fuchs' dystrophy (19), corneal opacification (7), keratoconus (5), and other (10) (Table 2). Corneas were matched for patients by the eye bank utilizing the recipient age and diagnosis according to standard EBAA protocol

TABLE 1. DEATH-TO-PRESERVATION TIME (DP), EPITHELIAL STATUS IN PRESERVATION MEDIUM, AND EPITHELIAL STATUS ON DAY 1 POSTOPERATIVELY IN 81 PATIENTS

PATIENT NO.	DONOR NO.	DONOR AGE	DP	EPITHELIAL RATING*	RECIP INITIALS	RECIP DX	1 DAY FOLLOW-UP	1 WEEK FOLLOW-UP	6 MONTH FOLLOW-UP	RECIP NOTES
1	134-02	59	4:01	E	JP	Edema	Clear	Clear	Clear	
2	142-02	72	5:26	E	AD	Fuchs	Clear	Clear	Clear	
3	144-02R	75	4:40	E	VC	Other	Intact	Clear	Clear	Irregular astigmatism
4	144-02L	75	4:40	E	EP	Fuchs	Intact	40% Defect	Clear	
5	148-02	55	3:03	E	VB	Edema	Intact	Intact/edema	Clear	
6	162-02	55	2:25	H	CK	Edema	Intact	Intact	Intact	
7	172-02	41	3:41	H	MR	Fuchs	30% Epithelial defect	Clear	Clear	
8	176-02	65	3:55	E	LF	Edema	Intact	Clear	Clear	
9	177-02	29	6:51	Sloughing	AH	Other	Defect	Clear	Clear	
10	183-02R	71	6:10	H	LB	Edema	98% Epithelial defect	Clear	Clear	
11	183-02L	71	6:10	H	BM	Opacity	Clear	Clear	Clear	
12	198-02	61	2:56	E	VF	Opacity	Clear	Clear	Clear	
13	203-02R	15	11:03	E	GB	Edema	35% Epithelial defect	60% Epithelial defect	Clear	Fungal keratitis

TABLE 1. (CONTINUED) DEATH-TO-PRESERVATION TIME (DP), EPITHELIAL STATUS IN PRESERVATION MEDIUM, AND EPITHELIAL STATUS ON DAY 1 POSTOPERATIVELY IN 81 PATIENTS

PATIENT NO.	DONOR NO.	DONOR AGE	DP	EPITHELIAL RATING*	RECIP INITIALS	RECIP DX	1 DAY FOLLOW-UP	1 WEEK FOLLOW-UP	6 MONTH FOLLOW-UP	RECIP NOTES
14	203-02L	15	11:03	E	KL	Edema	100% Epithelial defect	95% Epithelial defect	Clear	
15	205-02	61	8:55	E	KM	Edema	10% Epithelial defect	Intact	Intact	
16	211-02	56	7:10	E	MB	Other	Intact	Intact	Intact	
17	216-02R	67	5:20	E	MG	Edema	100% Defect	Intact	Intact	
18	216-02L	67	5:20	E	MK	Edema	Clear	Clear	Clear	Regraft
19	219-02	51	4:30	E	ES	Edema	Intact	Intact	Clear	
20	221-02	63	7:07	H	MW	Edema	Intact	Intact	Intact	
21	234-02	74	6:00	E	JF	Edema	70% Epithelial defect	Intact	Intact	
22	237-02	16	3:55	E	RB	Edema	Intact	Clear	Clear	Regraft
23	239-02	58	8:20	E	GL	Opacity	25 % Epithelial defect	Clear	Clear	
24	241-02	40	4:32	H	MB	Opacity	Clear	Clear	Clear	
25	242-02	56	6:46	Sloughing	DT	Other	25% Epithelial defect	Clear	Clear	Regraft
26	251-02R	50	8:45	E	MH	Fuchs	95% Epithelial defect	Clear	Clear	
27	251-02L	50	8:45	E	GA	Other	Clear	Clear	Clear	Lattice dystrophy
28	253-02	57	5:32	H	AS	Edema	Clear	Clear	Clear	
29	259-02	17	12:25	Sloughing	ML	Fuchs	20% Epithelial defect	Clear	Clear	

TABLE 1. (CONTINUED) DEATH-TO-PRESERVATION TIME (DP), EPITHELIAL STATUS IN PRESERVATION MEDIUM, AND EPITHELIAL STATUS ON DAY 1 POSTOPERATIVELY IN 81 PATIENTS

PATIENT NO.	DONOR NO.	DONOR AGE	DP	EPITHELIAL RATING*	RECIP INITIALS	RECIP DX	1 DAY FOLLOW-UP	1 WEEK FOLLOW-UP	6 MONTH FOLLOW-UP	RECIP NOTES
30	265-02	15	8:42	E	DC	KC	60% Epithelial defect	Clear	Clear	
31	271-02	71	3:50	E	NP	Edema	Clear	Clear	Clear	
32	284-02R	51	5:36	E	NG	Edema	Clear	Clear	Clear	
33	284-02L	51	5:36	E	ZS	Edema	Clear	Clear	Clear	
34	287-02R	20	9:05	E	CH	KC	90% Epithelial defect	Clear	Clear	
35	287-02L	20	9:05	E	PJ	Fuchs	25% Epithelial defect	Clear	Clear	
36	293-02	17	4:57	E	GG	Fuchs	Clear	Clear	Clear	
37	294-02	66	5:23	CL	GB	Edema	Central defect	10% defect	—	Regraft 3 months
38	296-02	54	6:00	Sloughing	LW	Edema	Defect	Central opacity	Central opacity	See Figure 5
39	300-02R	58	9:46	E	VF	Opacity	Intact	Clear	Clear	Regraft
40	300-02L	58	9:46	E	JG	Fuchs	Intact	Clear	Clear	
41	302-02R	60	9:37	E	EH	Fuchs	Intact	Clear	Clear	
42	302-02L	60	9:37	E	JC	Edema	Defect	Clear	Clear	
43	309-02	21	8:49	E	RL	KC	100% Epithelial defect	Clear	Clear	
44	312-02	68	7:00	E	SP	Fuchs	75% Epithelial defect	Clear	Clear	
45	315-02	34	7:20	E	CA	Fuchs	Defect	Clear	Clear	
46	324-02	62	4:59	CL	IW	Edema	Intact	Clear	Clear	
47	329-02	19	8:20	E	JP	Fuchs	Intact	Clear	Clear	
48	332-02	71	4:13	CL	LP	Opacity	Intact	Clear	Clear	

TABLE 1. (CONTINUED) DEATH-TO-PRESERVATION TIME (DP), EPITHELIAL STATUS IN PRESERVATION MEDIUM, AND EPITHELIAL STATUS ON DAY 1 POSTOPERATIVELY IN 81 PATIENTS

PATIENT NO.	DONOR NO.	DONOR AGE	DP	EPITHELIAL RATING*	RECIP INITIALS	RECIP DX	1 DAY FOLLOW-UP	1 WEEK FOLLOW-UP	6 MONTH FOLLOW-UP	RECIP NOTES
49	334-02	55	8:06	Sloughing	MB	KC	75% Epithelial defect	Clear	Clear	
50	335-02	60	4:17	E	WA	Edema	Intact	Clear	Clear	
51	337-02	14	2:32	E	IR	Edema	Intact	Clear	Clear	
52	08-03	73	8:03	E	GK	Fuchs	Clear	Clear	Clear	
53	13-03R	68	4:38	Central sloughing	AB	Opacity	Clear	Clear	Clear	
54	13-03L	68	4:38	E	EP	Fuchs	Intact	Intact	Intact	Fuchs
55	19-03R	66	6:44	Epithelial tears at limbus	AW	Edema	Intact	Clear	Clear	Epithelial tears from recovery
56	19-03L	66	6:44	E	LW	Edema	Intact	Clear	Clear	
57	27-03	55	4:20	E	JD	Other	Intact	Clear	Clear	Perforated ulcer
58	32-03	11	7:19	Patchy sloughing	AS	Edema	20% Epithelial defect	Clear	Clear	
59	33-03	38	5:43	Patchy sloughing	HL	Other	Intact	Clear	Clear	Perforated ulcer
60	34-03	42	7:40	E	DW	Edema	75% Epithelial defect	Clear	Clear	
61	36-03	63	2:01	Sloughing	SQ	Other	100% Defect	Haze	Failed graft	Regraft
62	37-03	44	5:10	Sloughing	RD	Edema	Intact	Clear	Clear	
63	39-03	64	7:14	CL	BA	Edema	Intact	Clear	Clear	Epithelial defect from sutures

TABLE 1. (CONTINUED) DEATH-TO-PRESERVATION TIME (DP), EPITHELIAL STATUS IN PRESERVATION MEDIUM, AND EPITHELIAL STATUS ON DAY 1 POSTOPERATIVELY IN 81 PATIENTS

PATIENT NO.	DONOR NO.	DONOR AGE	DP	EPITHELIAL RATING*	RECIP INITIALS	RECIP DX	1 DAY FOLLOW-UP	1 WEEK FOLLOW-UP	6 MONTH FOLLOW-UP	RECIP NOTES
64	44-03R	48	6:04	E	SW	Fuchs	5% Epithelial defect	Clear	Clear	
65	44-03L	48	6:04	E	CM	Edema	Intact	Clear	Clear	
66	46-03R	73	6:50	Patchy sloughing	MB	Other	Defect	Clear	Clear	
67	46-03L	73	6:50	Sloughing	LT	Edema	Intact	Clear	Clear	
68	56-03R	46	8:55	E	BD	KC	90% Epithelial defect	Defect	Failed, rejection	Poor compliance
69	56-03L	46	8:55	E	FS	Edema	20% Epithelial defect	Clear	Clear	
70	58-03	64	6:15	E	HW	Edema	90% Epithelial defect	2-mm defect	Clear	
71	74-03	41	6:48	E	AC	Edema	Intact	Clear	Clear	
72	87-03	62	3:25	CL	BA	Edema	Intact	Clear	Clear	Regraft
73	99-03	26	12:49	Sloughing	KR	Other	100% Epithelial defect	Clear	Clear	
74	101-03	58	7:05	E	PM	Fuchs	Intact	Clear	Clear	Fuchs
75	106-03R	68	3:01	CL	LS	Edema	Intact	Clear	Clear	
76	106-03L	68	3:01	CL	RD	Fuchs	Intact	Clear	Clear	
77	125-03	52	5:52	CL	WW	Fuchs	Intact	Clear	Clear	
78	128-03	42	1:59	CL	RB	Edema	Intact	Clear	Clear	
79	135-03	53	7:18	CL	JW	Edema	Intact	Clear	Clear	
80	144-03	60	2:31	E	MC	Fuchs	Intact	Clear	Clear	
81	151-03	64	4:43	E	BB	Edema	Defect	Defect	Haze	

CL = clear and intact; Dx = diagnosis; E = exposure; H = haze; KC = keratoconus; Recip = recipient.
In Optisol-G.

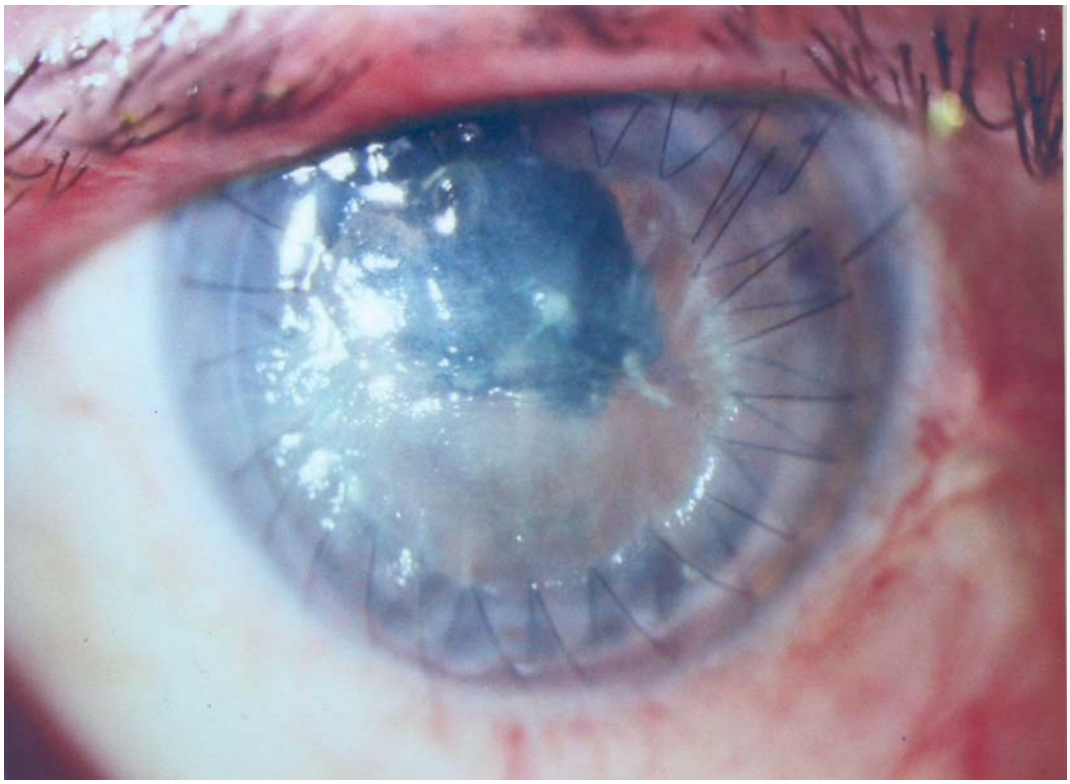

FIGURE 2

Epithelial defect in graft on day 1 resulting from donor tissue with epithelial defect.

using a patient-based distribution system. The cornea surgeon was aware of the tissue rating prior to surgery. The epithelial status was graded by using standard EBAA criteria (eg, intact, superficial punctate keratitis [SPK], sloughing). All corneas were procured by one eye bank, rated by one technician (H.W.), and harvested according to consistent standards of the EBAA and the University of Kentucky Eye Bank. All donor corneas were supplied as cornea sclera preparations in Optisol GS corneal storage medium (Baush & Lomb, St Louis, Missouri).

Penetrating keratoplasty was performed by using an Iowa punch (Jim's Instrument Manufacturing, Iowa City, Iowa) for the donor, a Hessburg-Barron vacuum trephine (Jed Med, St Louis, Missouri) for the host, and a 24-bite 10.0-nylon continuous running suture for closure. All grafts were oversized by 0.5 mm with an 8.0 mm trephine blade for the donor and a 7.5 mm trephine blade for the host. All patients in the operating room received an antibiotic-soaked collagen shield and subconjunctival injection of methylprednisolone, 40 mg. A lateral frost suture tarsorrhaphy was placed for 1 week. All patients were seen at 1 day, 1 week, day 8, and 1 month following surgery by the surgeon for follow-up care. The status of the donor epithelium and graft clarity were noted from the patient's charts at these intervals. Epithelial status on day 1 was recorded in the chart, with central defects larger than 10% included. Epithelial defects less than 10% or defects over the graft-host junction in the suture line were not considered related to the donor surface.

TABLE 2. INDICATIONS FOR KERATOPLASTY IN 81 STUDY PATIENTS

INDICATION	NO. OF PATIENTS (%)
Corneal edema (pseudophakic or aphakic)	40 (49.3)
Fuchs' dystrophy	19 (23.4)
Corneal opacity	7 (8.6%)
Keratoconus	5 (6.1)
Other	10 (12.3)

RESULTS

The average DP time of all 81 donor corneas was 6:18 hours (ie, 6 hours, 18 minutes) (range, 1:59 to 12:25) (Table 3). Thirteen corneas with epithelium sloughing in storage medium (Table 4) had an average DP time of 7:02 (range, 2:01 to 12:25) hours, and nine (69%) had DP time of longer than 6 hours. The average DP time of 68 corneas with no epithelium sloughing in storage medium was 6:09 (range, 1:59 to 11:03) hours ($P < .32$) (Table 5). Stratified by DP interval, the percentage of donor corneas with an intact epithelium decreases from 92.8% when DP time is less than 4 hours to 0% when it is 12 hours and over; the *P* value of .32 suggests limited causation or low numbers (Figure 3).

TABLE 3. DEMOGRAPHICS FOR ALL 81 DONORS DEMONSTRATING RANGE OF DEATH-TO-PRESERVATION TIMES

CHARACTERISTIC	VALUE
Age range (years)	11 to 75
Average age (years)	51.81481
Age, standard deviation	18.14601
Death to preservation, range (hours)	1:59 to 12:25
Death to preservation, average (hours)	6:18
Death to preservation, standard deviation	0.099959

Twenty-eight (35%) of 81 patients had central epithelial defects on day 1 (Table 6). Stratified by DP time, the percentage of patients with epithelial defects in the graft on day 1 decreased from 86% when DP time is under 4 hours to 0% when it is 12 hours and over (Figure 4). In contrast, the percentage of patients with epitheliopathy in the graft increased with increasing DP time, from 14% when DP time was under 14 hours to 100% when it was 12 hours and over. Average DP time in 53 corneas with an intact epithelium on day 1 postoperatively was 5.23 hours (Table 7). Average DP time in 28 patients with an epithelial defect on day 1 postoperatively was 8.01 hours. ($P = .000027$) (Table 6).

TABLE 4. DEMOGRAPHICS FOR 13 DONOR CORNEAS WITH SLOUGHING IN PRESERVATION MEDIUM DEMONSTRATING AVERAGE DEATH-TO-PRESERVATION TIME

CHARACTERISTIC	VALUE
Age range (years)	11 to 73
Average age (years)	46.69231
Age, standard deviation	21.00153
Death to preservation, range (hours)	2:01 to 12:25
Death to preservation, average (hours)	7:02
DEATH TO PRESERVATION, INTERVALS (HOURS)	
0 to 3:59	1 (7.7%)
4 to 5:59	3 (23.1%)
6 to 7:59	6 (46.1%)
8 to 9:59	1 (7.7%)
10 to 11:59	0 (0%)
12 and over	2 (15.4%)

Ten of the 13 recipients (77%) with donor corneas that had epithelial sloughing noted in storage medium had epithelial defects at slit-lamp examination on day 1 postoperatively. Twenty-one (31%) of 68 patients with donors that had an intact epithelium in preservation medium had an epithelial defect in the graft at day 1. Multiple host factors account for the variability of the corneal epithelium immediately after transplant.

Nine of 25 corneas (36%) with DP time longer than 8 hours were under 30 years of age, and the average DP time in 13 donors under age 30 was 8.3 hours. The increased DP time in younger donors is due to several factors in the retrieval process and is discussed subsequently.

TABLE 5. DEMOGRAPHICS FOR 68 DONOR CORNEAS WITHOUT SLOUGHING IN PRESERVATION MEDIUM DEMONSTRATING AVERAGE DEATH-TO-PRESERVATION TIME

CHARACTERISTIC	VALUE
Age range (years)	14 to 75
Average age (years)	53
Age, standard deviation	17.5529551
Death to preservation, range (hours)	1:59 to 11:03
Death to preservation, average (hours)	6:09
Death to preservation, standard deviation	0.09538163
DEATH TO PRESERVATION, INTERVALS (HOURS)	
0 to 3:59	13 (92.8%)
4 to 5:59	20 (86.9%)
6 to 7:59	17 (73.9%)
8 to 9:59	16 (94.1%)
10 to 11:59	2 (100%)
12 and over	0 (0%)
P value .322521332	

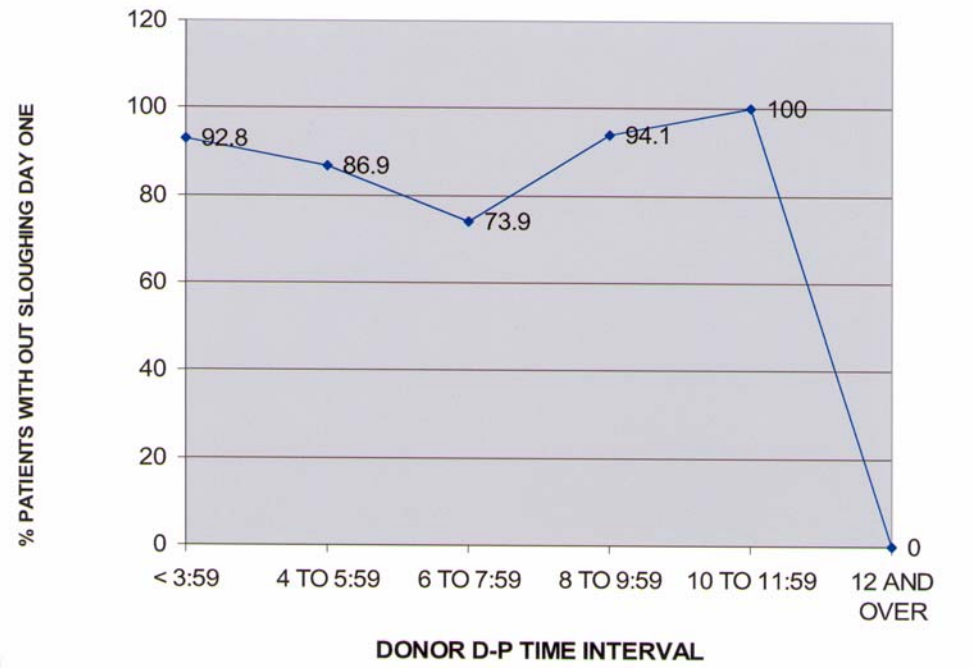

FIGURE 3

The percentage of patients without sloughing of the donor epithelium decreases as donor death-to-preservation time increases.

TABLE 6. DEMOGRAPHICS FOR 28 CORNEAS WITH EPITHELIAL DEFECTS ON DAY 1 POSTOPERATIVELY DEMONSTRATING AVERAGE DEATH-TO-PRESERVATION TIME

CHARACTERISTIC	VALUE
Age range (years)	11 to 74
Average age (years)	44.6785714
Age, standard deviation	21.49945
Death to preservation, range (hours)	3:41 to 12:49
Death to preservation, average (hours)	8:01
Death to preservation, standard deviation	0.091499
DEATH TO PRESERVATION, INTERVALS	
0 to 3:59	1 (7.1%)
4 to 5:59	3 (13%)
6 to 7:59	9 (39.1%)
8 to 9:59	11 (64.7%)
10 to 11:59	2 (100%)
12 and over	2 (100%)

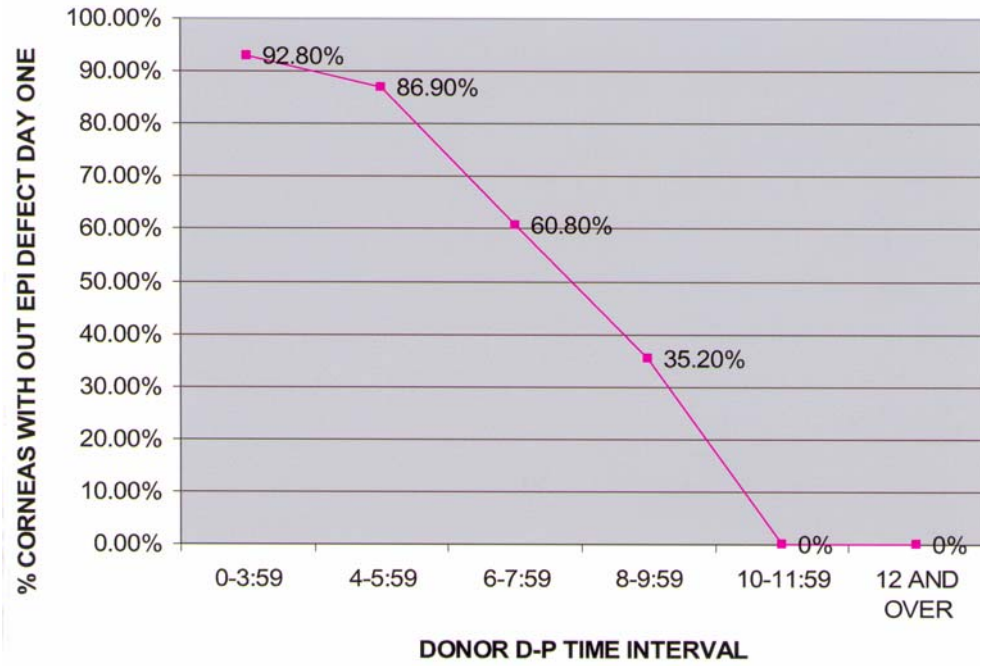

FIGURE 4

The percentage of corneas without an epithelial defect on day 1 decreases as donor death-to-preservation time increases.

TABLE 7. DEMOGRAPHICS FOR 53 CORNEAS WITHOUT EPITHELIAL DEFECT ON DAY 1 POSTOPERATIVELY DEMONSTRATING AVERAGE DEATH-TO-PRESERVATION TIME

CHARACTERISTIC	VALUE
Age range (years)	11 to 75
Average age (years)	56.4716981
Age, standard deviation	14.02142
Death to preservation, range (hours)	1:59 to 9:46
Death to preservation, average (hours)	5:23
Death to preservation, standard deviation	0.082673
DEATH TO PRESERVATION, INTERVALS (HOURS)	
0 to 3:59	13 (92.8%)
4 to 5:59	20 (86.9%)
6 to 7:59	14 (60.8%)
8 to 9:59	6 (35.2%)
10 to 11:59	0 (0%)
12 and over	0 (0%)

DISCUSSION

Because manpower and serologic testing costs exist for all tissue harvested, regardless of whether or not the tissue is utilized, the economic ramifications of increased tissue quality and utilization are not small. Our study suggested that higher DP times increase the chance of epithelial sloughing from the donor tissue. An absent or partially absent epithelium increases the potential for trauma from exposure, medications used in preparation of the donor, or preservation medium.

Maintenance of the donor corneal epithelium prior to harvesting is an important variable in determining the quality of the donor epithelium in preservation medium. EBAA medical standards encourage the use of lubricants, refrigeration, lid closure, and cleanliness to help protect the donor epithelium. Epithelial exposure, when noted, frequently is in the interpalpebral fissure. Attending to keeping the donor cornea lubricated and the lids closed helps to maintain a pristine epithelium. Epithelial sloughing was the fourth most common reason for not using donor cornea tissue, ranking behind medical social history, donor pathology, and opacity.

Corneal tissue was procured by EBAA eye banks in 2004 from over 80,000 donors. Traumatic epithelial defects such as those from motor vehicle accidents or blunt trauma usually were cause for the tissue to be declared unsuitable for transplant. Of most interest to the eye banking community would be the patient who initially has a pristine donor epithelium at the time of death but a disrupted or absent epithelium at the time of preservation. This particular cohort of patients would benefit most from meticulous attention to strict methods to preserve the donor epithelium, during the DP interval. Many variables, such as the preparation of the donor, antibiotics used, skill of the technician involved in the recovery, and the location of the body (eg, funeral home, hospital), cannot be controlled and are outside the scope of this study. Increased DP time increases the incidence of donor epithelial sloughing in storage medium, which may or may not rule out using the tissue. From a practical standpoint, protection of the epithelium should increase the clarity (rating) and usefulness (acceptability) of the tissue. Increased DP also increases the incidence of epithelial defects on the donor cornea postoperatively, which can adversely affect the survival of the graft.[10]

There was a stronger correlation when stratified by 2-hour time intervals between the DP time and epithelial defects in the graft on day 1 ($P < .001$) than between DP time and epithelial rating in storage medium ($P = .32$). Many variables that contribute to epithelial defects prior to tissue harvesting, such as refrigeration of the body, care of lids and eyes after death, and use of medications for preexisting ocular disease, are known. Other variables, such as eye care during extensive hospitalization or trauma around the time of death, are less well known. Consequently, insults to the epithelium may not show immediately in preservation medium and manifest themselves after the stress of surgery (Figure 5).

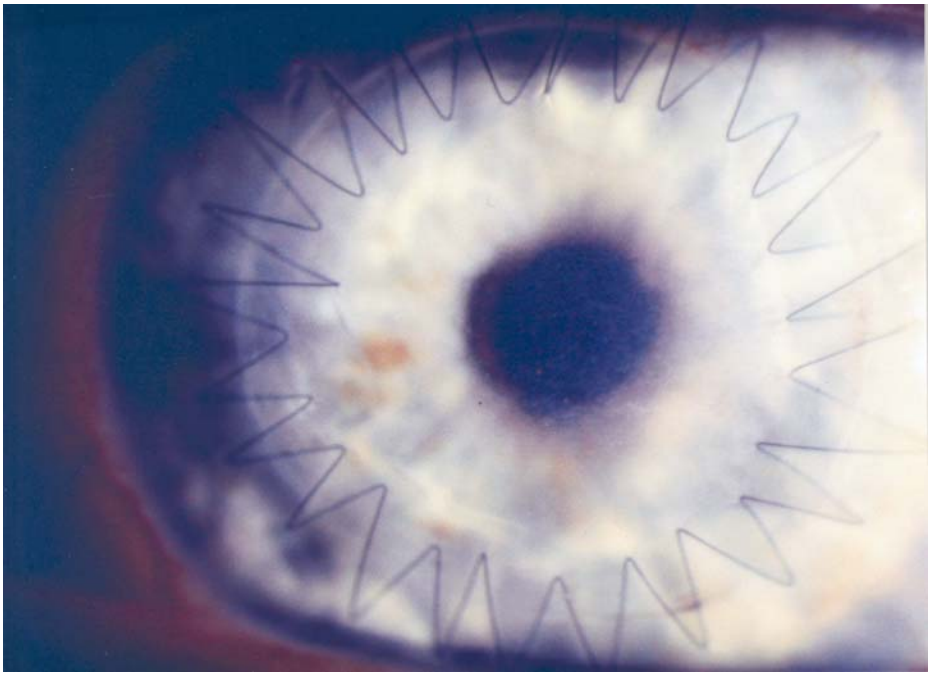

FIGURE 5A

Anterior stromal haze in graft resulting from a persistent central epithelial defect in the graft.

The significantly longer DP time noted in younger patients is related to the fact that death of younger patients is frequently more unexpected than death of older patients. Older patients are more likely to succumb to known preexisting diseases or die in a hospital, where it is easier to get consent from relatives and do a thorough evaluation of the medical and social histories. Younger patients are more likely to die of trauma, and a longer time is needed for medical and social history review when records have to be gathered from multiple sources. Consent frequently falls behind other family issues in order of importance and is discussed later or not all.

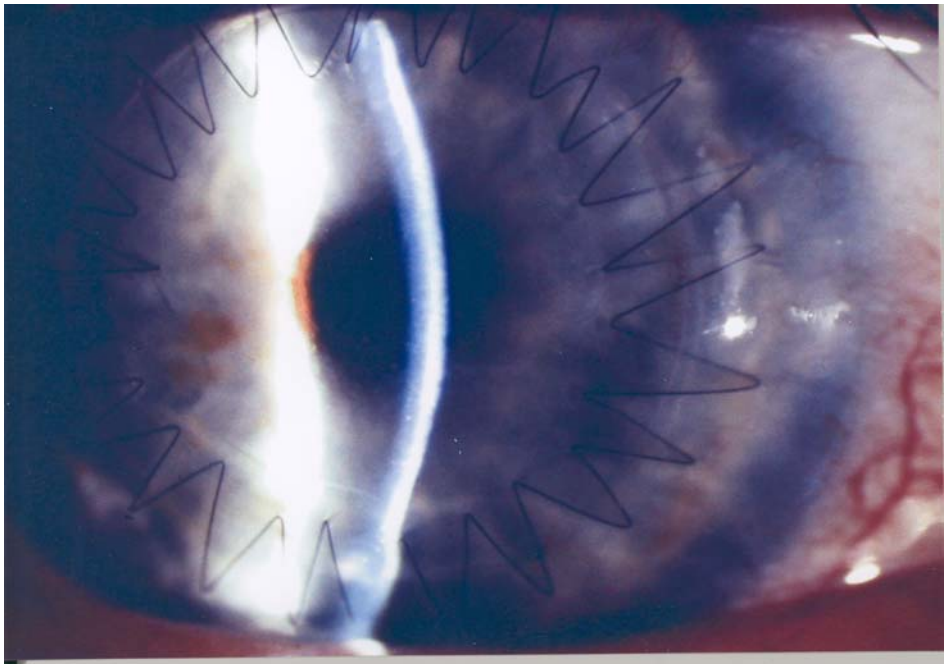

FIGURE 5B

A slit beam of light demonstrating subepithelial scarring in the graft.

Epitheliopathy on the recipient cornea is only one of the plethora of factors that influence graft clarity. Other features, such as recipient age, lid condition, blepharitis, tear film quality, ocular medications, and the immune status of the host, may be outside the

control of the surgeon or unrecognized by the surgeon. The eye banking community can do little about most recipient variables and can control only those variables under the jurisdiction of the procurement process. The operating surgeon has some control over the postoperative epithelium in dictating the use of medications, maintenance of the epithelium with lubricants, and tarsorrhaphy. Epithelial irregularities, such as hurricane keratopathy, specific epithelial defects, filamentary keratitis, or foreign bodies managed early and appropriately, are less likely to cause additional problems than those issues that are diagnosed late or managed ineffectively. We believe that an intact epithelium on day 1 makes surface maintenance easier and improves the chances of a clear graft, especially in patients with pre-existing ocular surface disease, but we acknowledge that the status of the recipient's surface for the long term is determined by multiple host factors in addition to the status of the epithelium on day 1.

Although our data show that increased DP time results in a poorer-quality donor epithelium, the effect of poor donor epithelium on the graft clarity after several months is difficult to show. Postoperative graft clarity depends on a number of variables, such as tear film, lid morphology, topical and systemic medications, and environment (eg, smoking, humidity). For example, one donor with DP time of 11 hours developed fungal keratitis at 2 months, but the mate remained clear without complications. Mannis and associates[9] reported a high incidence of SPK in older patients, suggesting that age of the recipient may be a very important determinant of the postgraft surface as well.

Some form of surface keratopathy is ubiquitous following keratoplasty. However, there is an advantage to an intact epithelium on the donor graft immediately following surgery. Many surgeons anecdotally report that patients develop subepithelial scarring when a donor epithelial defect fails to resolve spontaneously. Because epithelial defects can produce adverse effects for postkeratoplasty patients, the quality of the donor corneal epithelium should be maintained where possible. The DP time may be a more important variable than the epithelial rating by the eye bank for predicting epithelial defects on day 1 following keratoplasty. Additional long-term evaluation of the effects of early surface disease on long-term quality of the graft surface and graft clarity will shed new light on the importance of an intact epithelium on the donor cornea.

REFERENCES

1. United Network for Organ Sharing. 2004 Annual Report. Richmond, Virginia: United Network for Organ Sharing; 1996.
2. Eye Bank Association of America (EBBA). 2004 Eye Banking Statistical Report Washington, DC: Eye Bank Association of America; 2004.
3. Feiz V, Mannis MJ, Kandavel G, et al. Surface keratoplasty after penetrating keratoplasty. *Trans Am Ophthalmol Soc* 2001;99:159-170.
4. Bron AJ. Vortez patterns of the corneal epithelium. *Trans Ophthalmol Soc U K* 1973;93:455-472.
5. Lemp MA, Mathers WD. Vortex keratopathy of the corneal graft. *Cornea* 1991;10:93-99.
6. Mackman GS, Polack FM, Sydrys L. Hurricane keratitis in penetrating keratoplasty. *Cornea* 1983;2:31-34.
7. Stulting RD, Waring GO III, Bridges WZ, et al. Effect of donor epithelium on corneal transplant survival. *Ophthalmology* 1988;1988;95:803-812.
8. Price FW, Whitson WE, Collins KS, et al. Five-year corneal graft survival: a large, single-center patient cohort. *Arch Ophthalmol* 1993;111:799-805.
9. Mannis MJ, Zaknik K, Miller MR, et al. Pre-operative risk factors for surface disease after penetrating keratoplasty. *Cornea* 1997;16:7-11.
10. Spencer WH. Ophthalmic pathology. Vol 1. 4th ed. Philadelphia: Saunders; 1996:224.

PEER DISCUSSION

DR ALAN SUGAR. I agree with Dr Van Meter that many variables affect the quality of donor corneas used for transplantation. The status of the donor epithelium is only one of them. However, since the realization about 50 years ago that the endothelium was a critical tissue for corneal health, and especially since the development of clinical specular microscopy about 30 years ago, we have been more concerned about endothelial than epithelial cells. Dr Van Meter's concern about donor epithelium reflects renewed interest in corneal surface disease and currently limited information on donor epithelium.

There is no question that epithelial healing affects corneal graft clarity, causing up to 25% of graft failures[1]; but most failures from surface problems do not occur immediately. They occur at three months or later post-op. Machado and colleaguesl[2] have suggested that first post-operative day epithelial status is not predictive of later epithelial health or graft clarity. Nonetheless, the interactions between corneal epithelium and stroma, particularly following epithelial wounding, are complex. Wilson[3] and others have demonstrated apoptosis of anterior stromal keratocytes in the presence of epithelial defects, and Erie[4] has shown that these effects may be prolonged.

Dr Van Meter's study used a consistent group of patients from one experienced surgeon, and showed that the presence of an epithelial defect on the first day post keratoplasty significantly correlates with donor death to preservation time. Those with first day defects had mean death to preservation time of just over eight hours, while those without defects had mean death to preservation time of about 5 1/2 hours. 14% of donors preserved within four hours or less and 100% of those preserved after more than ten hours post death had day 1 defects. Six eyes had defects at one week, and the relationship to death-preservation time appeared to hold. There is no evidence, however, that in this relatively small series, the long-term graft success was affected. It is interesting that Kim and colleagues[5], showed an association of day one epithelial defects with increased preservation to surgery rather than death to preservation time. Data on that variable were not reported in the study we are considering here.

It happens that the Cornea Donor Study (CDS), a national collaborative cohort study designed to evaluate the effect of donor age on long-term keratoplasty outcomes in 1101 eyes, is currently approaching five-year outcome analysis. As part of that study, extensive donor data have been collected and recently reported[6]. Donors with defects of greater than 50% of the epithelium were excluded. In the CDS epithelial defects increased and quality declined and stromal edema and Descemets folds increased with increased death to preservation time. Donor body refrigeration or icing of eyes prior to corneal excision or enucleation was associated with a decrease in epithelial defects, even when we controlled for death to preservation time. The outcomes of the CDS and specific studies aimed at better understanding of the role of the epithelium in corneal transplantation, as Dr Van Meter's, will create a base of evidence for improved eye banking and corneal transplant outcomes. It is likely, however, that the epithelial results described today, will have only small effects on long-term outcomes. Before they are better understood, and before we can analyze the associated costs, I would caution against using these data to severely restrict death to preservation times and thus limit the availability of useful donor corneas, especially in recipients without surface disease.

REFERENCES

1. Price FW, Whitson WE, Collins KS, Marks RG. Five-year corneal graft survival: A large, single-center patient cohort. *Arch Ophthalmol* 1993; 111:799-805.
2. Machado RA, Mannis MJ, Mandel HA et al. The relationship between first postoperative day epithelial status and eventual health of the ocular surface in penetrating keratoplasty. *Cornea* 2002; 21:574-577.
3. Wilson SE, Mohan RR, Ambrosio R et al. The corneal wound healing response: cytokine-mediated interaction of the epithelium, stroma, and inflammatory cells. *Prog Retin Eye Res* 2001; 20:625-637
4. Erie JC. Corneal wound healing after photorefractive keratectomy: a 3-year confocal microscopy study. *Trans Am Ophthalmol Soc* 2003; 101:293-333
5. Kim T, Palay DA, Lynn M. Donor factors associated with epithelial defects after penetrating keratoplasty. *Cornea* 1996; 15:451-456.
6. Cornea Donor Study Group. Baseline donor characteristics in the Cornea Donor Study. *Cornea* 2005; 24:389-396

DR R. LINSY FARRIS. I would like to compliment Dr Van Meter for drawing our attention to the course of the ocular surface following corneal transplantation. So much of our attention has been given to the endothelium. How has the use of bandages such as extended wear contact lenses been worked into the algorithm of care of these corneas that have a sloughed epithelium?

DR VERINDER S. NIRANKARI. We know that corneal storage media is very good for preserving endothelium but, more important than death to preservation time, is a time from death to surgery time. These corneas can be in preservation media sometimes a week or 10 days because we have found that the endothelium is preserved for up to two weeks. I find that the longer the eye has been in preservation media before we use it results in more epithelial loss. Therefore it is perhaps more important than death to preservation time. Have your looked at that as well?

DR CHRISTOPHER J. RAPUANO. I agree with Dr Nirankari and advise our fellows, "if it looks bad pre-op, then there's going to be a big epithelial defect post-op; but if it looks good, and there's a long death-to-use time, then all bets are off. It could be good, or it may be bad." About 10 years ago we published a study on donor diabetes (Chou L, Cohen EJ, Laibson PR, Rapuano CJ: Factors Associated with Epithelial Defects after Penetrating Keratoplasty. *Ophthalmic Surgery* 1994; 25: 700-703) being a risk factor for epithelial defects in grafts. Did you look at any pre-op donor characteristics to see whether that correlated with epithelial defects?

DR JAY H. KRACHMER. Common sense tells us that when someone dies and their deceased tissue is sitting there, even cooled, it probably gets worse the longer it sits there. It is better to take it out of that poor environment, with decaying tissue and bad aqueous, and place it in a good preservative material. I agree with Dr Alan Sugar, that we have not yet really proven bad long-term results, but common sense tells us that we should really try to reduce that time.

DR RICHARD P. MILLS. Many of these donors had two eyes that were used for transplantation. Did you look at those paired results to see if the epithelium behaved similarly in the two donor eyes?

DR WOODFORD S. VAN METER. I appreciate all of these insightful comments. Let me answer Dr Mills question first. This study involved the donor corneas used by one surgeon for transplant, so we didn't look at paired donors unless both corneas in a pair happened to be assigned to the surgeon. There were thirteen paired donor corneas in our study: seven were the same on day one postoperatively, four pair with an intact epithelium in each cornea and three with epithelial sloughing on day one. Six of thirteen pairs were different on day one, one cornea having an intact epithelium and the mate with an epithelial defect. One donor with twelve hours death-to-preservation had epithelial sloughing on day 1 but did well, and the mate to it also had epithelial sloughing and developed fungal keratitis at two months. Clearly, multiple variables influence the postoperative donor epithelium.

I agree with Dr Rapuano that diabetes is detrimental to the epithelial surface. I did not study that factor in this present study but will research this question as soon as I get home.

I appreciate Dr Nirankari's comments about death to surgery time. I looked at the epithelial grading by the eye bank since that is a standard procedure of the eye bank as soon as the tissue goes into corneal storage medium. Many surgeons utilize the epithelial grade when evaluating tissue suitability. The time of death to surgery is equally important. All of the cases in this study were transplanted within four to seven days. Rarely can you get a cornea to the operating room in less than four days. I usually try not to use tissue after seven days in preservation medium.

Regarding use of a bandage contact lens, I agree that some protection of the new donor epithelium is helpful. Bandage contact lenses have been associated with an increased risk of infectious keratitis. I routinely do a lateral Frost suture tarsorrhaphy temporary lid closure on almost every patient at the time of transplant, regardless of what the cornea looks like at the time of surgery. I remove it on day one if the epithelium is pristine, but is nice to have it there in case the epithelium has SPK or is not perfect. In 90 percent of cases, I'll leave the tarsorrhaphy for a week. The tarsorrhaphy protects the epithelium so the patient can administer frequent topical corticosteroid eye drops with less fear of retarding reepithelialization.

I appreciate the comments of Dr Alan Sugar and agree that multiple variables affect the survival of a newly transplanted cornea. It is not my intent to discourage the use of corneas with long death-to-preservation times. I encourage those of you who are medical directors of your eye bank to encourage your eye bank to facilitate the preservation and recovery process utilizing the shortest possible of death-to-preservation time. Part of this oversight means taking care of the body, protecting the cornea, making sure the lids of the donor are closed, and working with your eye bank protocol to facilitate the recovery process. There is a financial incentive to improve the recovery process, since recently harvested tissue usually is more attractive to surgeons. We can use more corneas if we get a shorter death-to-preservation time. Ultimately, we hope the Cornea Donor Study will shed more light on how graft survival is affected by death to preservation time. While death-to-preservation time is but one variable affecting the donor cornea, our study suggests death-to-preservation time bears some impact on the quality of the epithelium in the early postoperative period.