



UNIVERSITY
OF BRESCIA

Vito Roman

DMEK
eye bank prepared

ENDOTHELIAL CELL VIABILITY

Preoperative surgeon evaluation of corneal endothelial status: the Viability Control of Human Endothelial Cells before Keratoplasty (V-CHECK) study protocol

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 Stephen B Kaye^{12,13}, Luigi Fontana¹⁴, Kunal A Gadhvi¹⁵,
 Antonello Moramarco¹⁶, Marina Rodriguez Calvo de Mora^{17,18,19},
 Carlos Rocha de Lossada^{20,21,22,23}, Vincenzo Scorcía²⁴, Pietro Viola²⁵,
 Stefano Calza²⁶, Hannah J Lewis²⁷, Mohit Parekh²⁸,
 Alessandro Ruzza²⁹, Stefano Ferrari³⁰, Diego Ponzin³¹,
 Francesco Semeraro³², Vito Romano^{33,34}



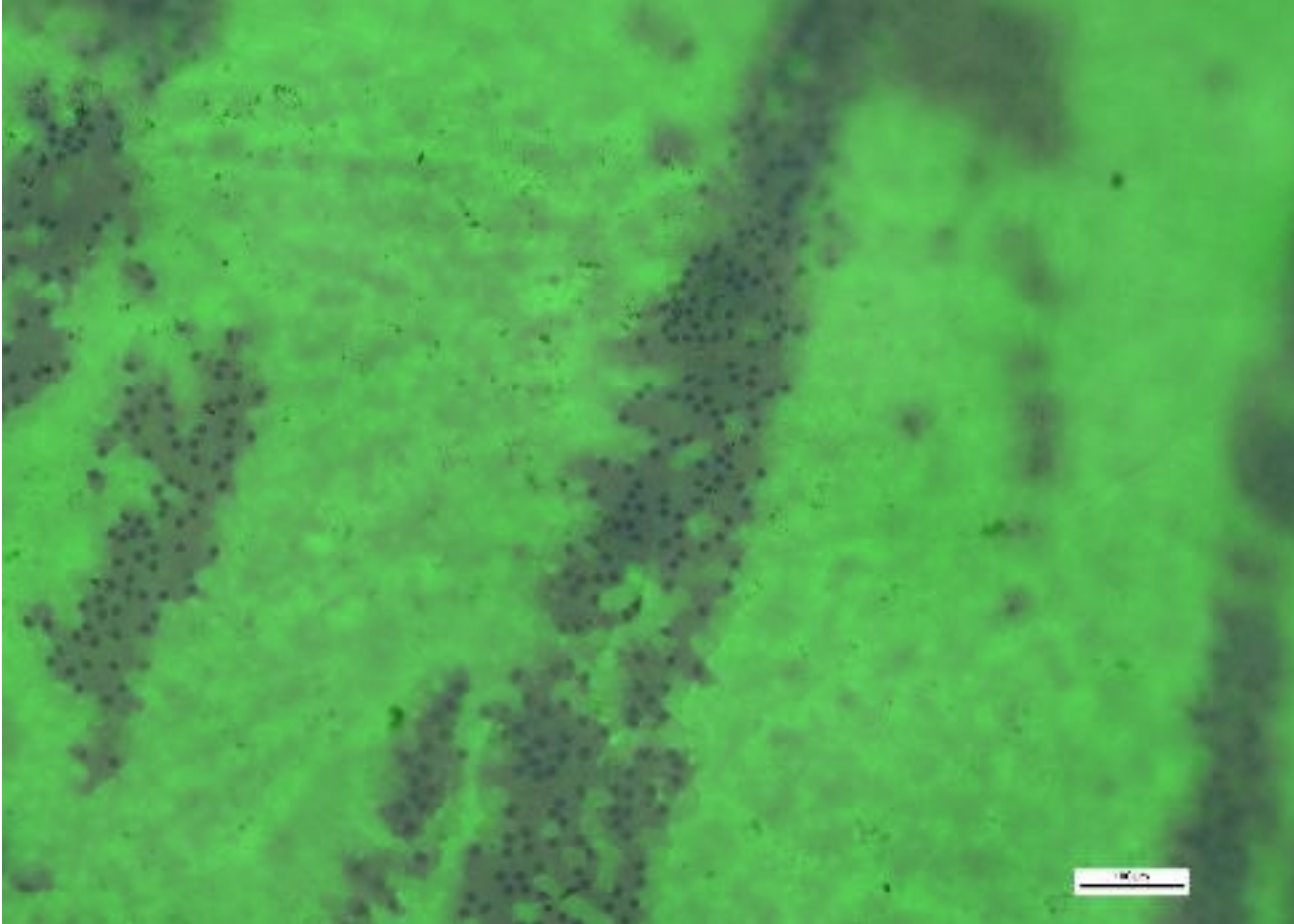
The V-CHECK method enables reproducible estimation of endothelial cell viability in donor corneas

tvst

Cornea & External Disease

Validation of a Deep Learning–Assisted Evaluation of Total Corneal Endothelial Cells Viability

Matteo Airaldi^{1–3}, Filippo Airaldi⁴, Zhuangzhi Gao³, Alessandro Ruzza⁵, Mohit Parekh⁶,
 Diego Ponzin⁵, Stephen Kaye^{2,3}, Francesco Semeraro^{7,8}, Stefano Ferrari⁵,
 Yalin Zheng³, and Vito Romano^{3,7,8}



-Live/dead endothelium staining shows near perfect agreement:

-**Trypan blue 0.05%** – stains dead endothelial nuclei and/or denuded Descemet

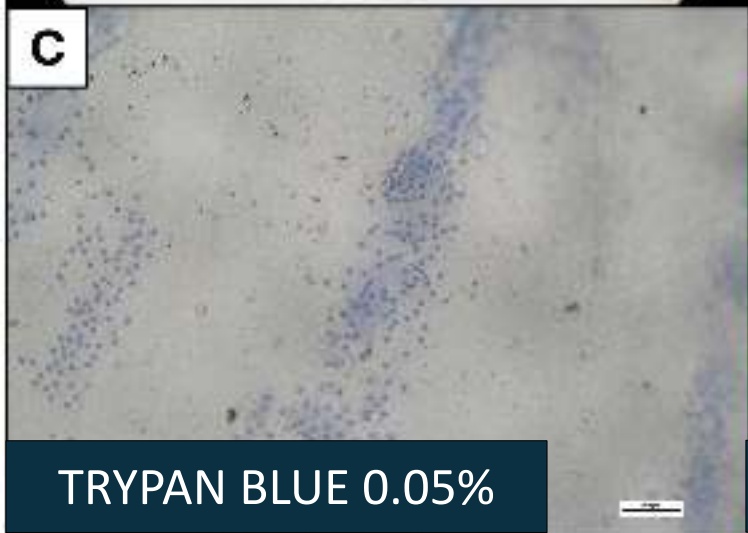
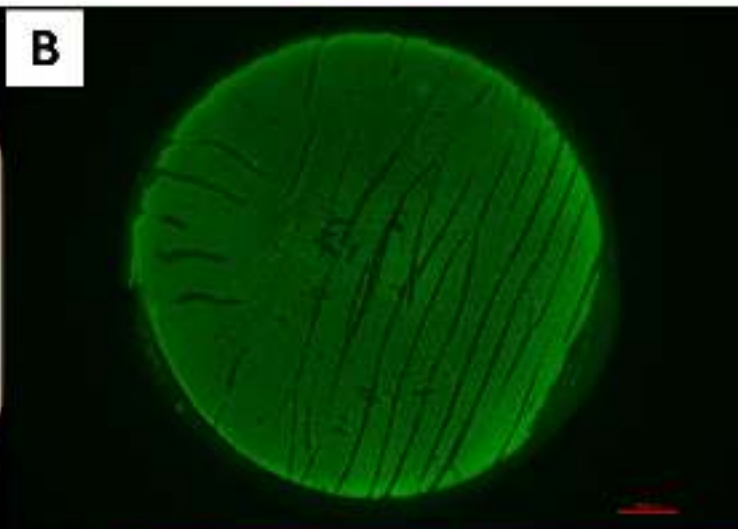
-**Calcein AM** – stains live endothelial cells

Surgeons and eye bank technicians need an **independent** and **reliable** evaluation of the **global** endothelial viability of the graft

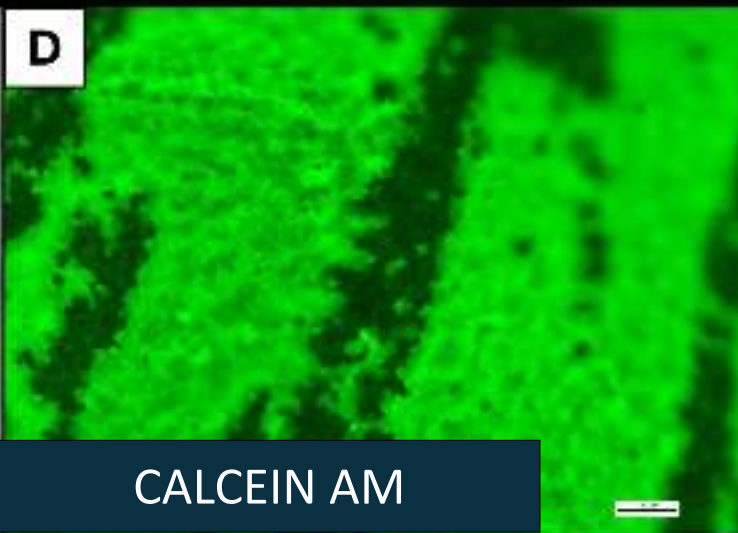
INTERVENTION: Develop an automatic algorithm to evaluate the extension of dead/denuded areas

Viability Control of Human Endothelial Cells before Keratoplasty (V-CHECK) method:

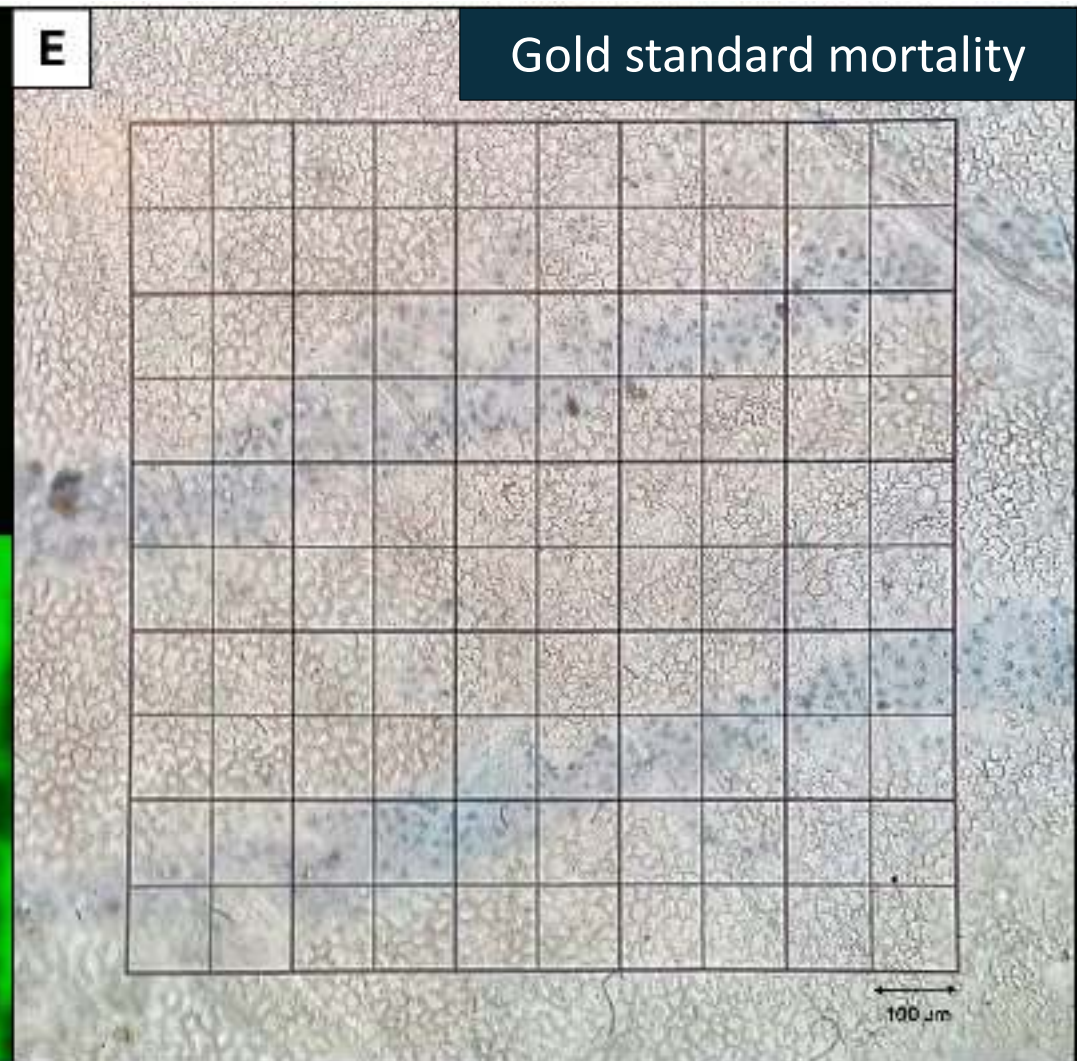
- Image acquisition
- Image analysis
- Validation



TRYPAN BLUE 0.05%



CALCEIN AM



INTRA-OP

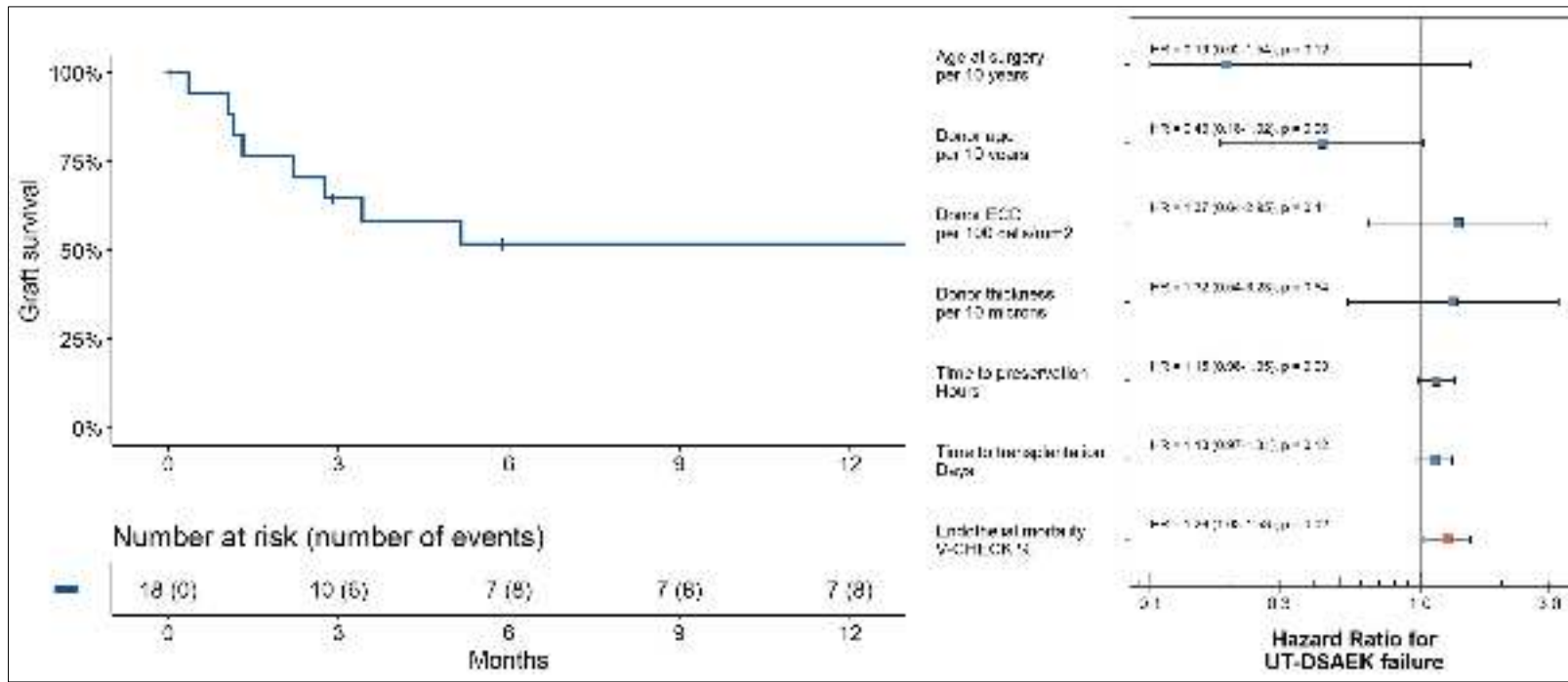
Donor check



V-CHECK – Image acquisition

INTRA-OP

Donor check



- V-CHECK: Multicenter cohort study (NCT05847387) – ongoing
- Preliminary results from a subgroup of UT-DSAEK patients (n = 18)
- Average donor lenticule ECD: 2592.3 (103.8) cells/mm²
- Average V-CHECK mortality: 13.9% (8.5%)
- High incidence of **Early Graft Failure** (<6 months) in grafts exhibiting endothelial staining (44.4%)

V-CHECK – Clinical relevance

Conclusions

CORNEA EVALUATION

Endothelial Cell Density: 2538 Clear Zone: 8 mm Lens Type: Phakic
of Fields: -----

Epithelium: MILD DIFFUSE EXPOSURE; DIFFUSE SLOUGHING
Stroma: CLEAR AND COMPACT; MILD ARCUS
Descemet's: NO FOLDS; NO DEFECTS
Endothelium: FEW DIFFUSE STRESS LINES; NO DEFECTS; MILD DIFFUSE CELL DROPOUT
Additional: REGULAR SCLERAL RIM; SCLERAL RIM 2MM OR GREATER CIRCUMFERENTIALLY
Comments: -----

Specular Microscopy
Image



Conclusions

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Comments: -----

Specular Microscopy
Image



**V-CHECK
MORTALIT
Y:
22%**

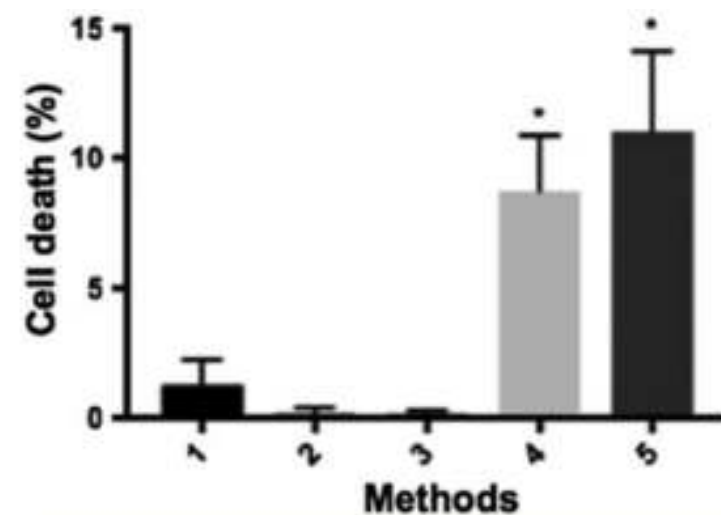
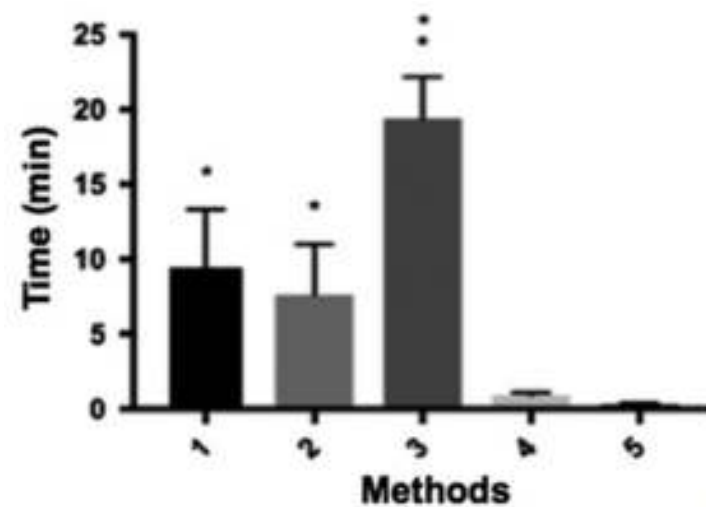
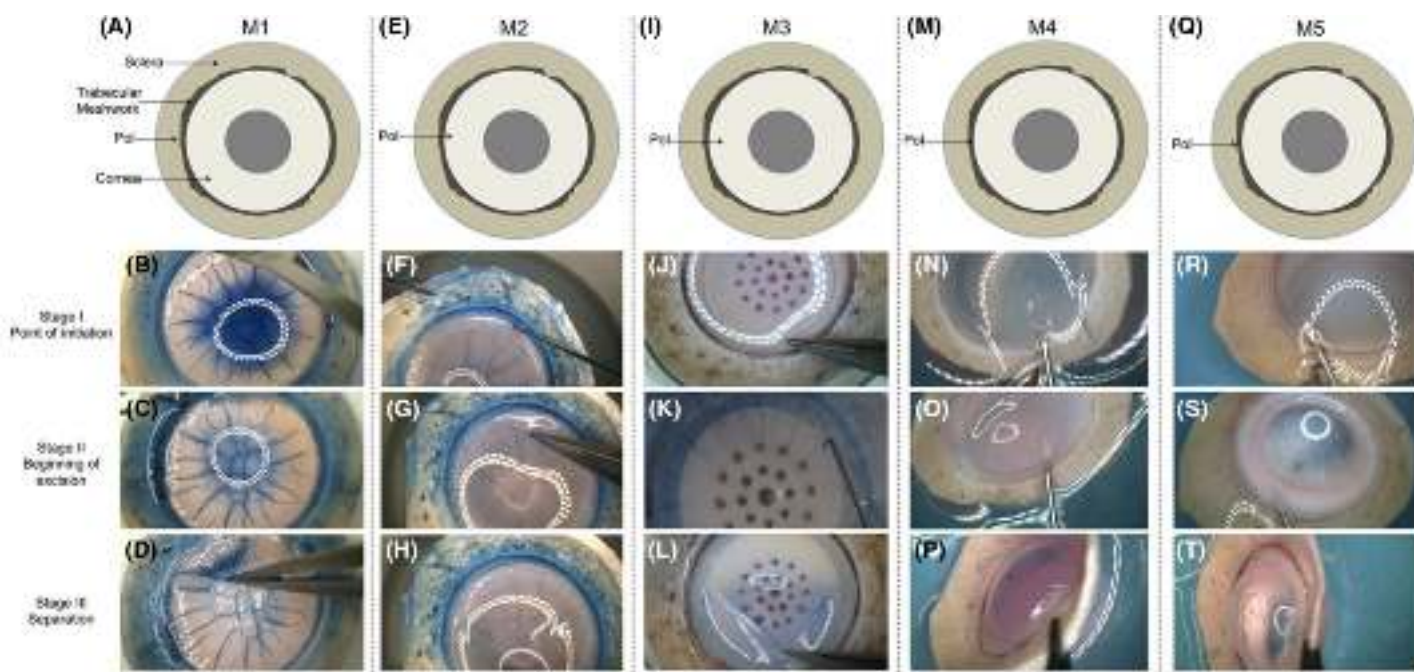
Conclusions

- Developed a **reproducible** evaluation of **pan-corneal** endothelial viability
- Estimated mortality is **consistently higher** than eye bank declared (agreement with literature)
- The **V-CHECK method** can be implemented:
 - in the **Eye Bank**: evaluation of tissues after processing and before shipment – **Change in reporting policy?**
 - in the **Operating theatre**: final check before transplant, evaluation of surgical damage
- The **V-CHECK method** can help standardize keratoplasty outcomes and survival
- Currently being implemented in a multicentric cohort study (IT, ES, NL, UK) – aim n = 135 patients
- Correlation of preoperative endothelial viability with keratoplasty success

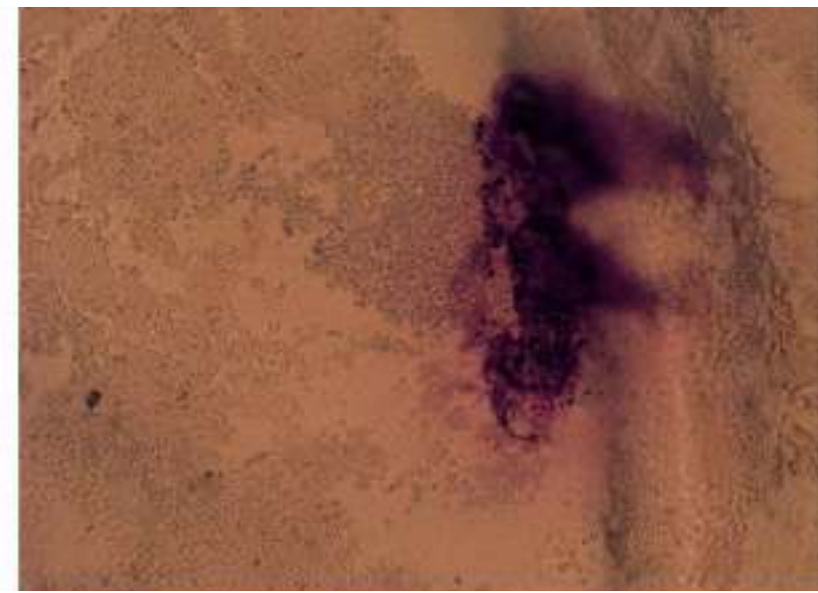
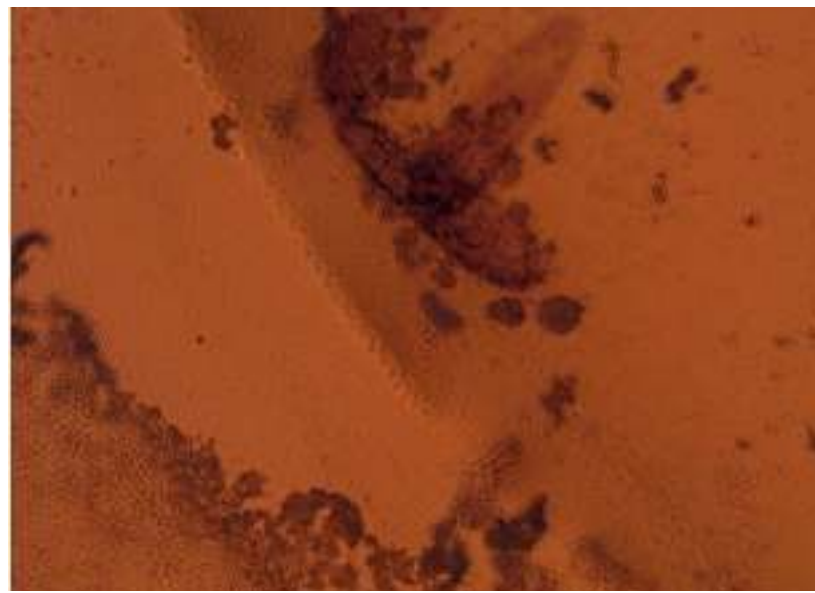
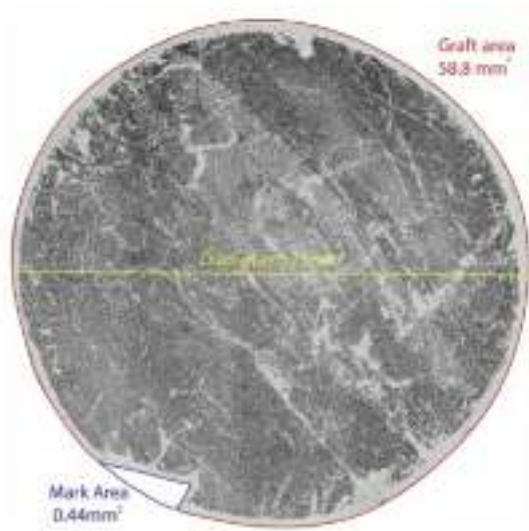
GRAFT PREPARATION

A comparative study on different Descemet membrane endothelial keratoplasty graft preparation techniques

Mohit Parekh,¹ Davide Borroni,^{2,3} Alessandro Ruzza,¹ Hannah J Levis,⁴ Stefano Ferrari,¹ Diego Ponzin¹ and Vito Romano⁵



marking



Evidence of cell loss surrounding marks Romano, BJO 2018



DMEK preparation



TABLE 2. Success Rates (With Percentages in Parentheses), Preparation Time (in Minutes), and ECL (%) After DMEK Graft Preparation

	Total	Senior Surgeon	Independent Surgeon	Corneal Fellow	P
Absolute success	15/18 (83.3)	5/6 (83.3)	5/6 (83.3)	5/6 (83.3)	0.56*
Relative success	1/18 (5.6)	1/6 (16.7)	0/6 (0)	0/6 (0)	
Failure	2/18 (11.1)	0/6 (0)	1/6 (16.7)	1/6 (16.7)	
Preparation time	6.21 ± 1.45	5.53 ± 1.45	6.58 ± 1.24	6.51 ± 1.62	0.39†
ECL	2.67 ± 1.98	2.49 ± 1.81	2.74 ± 2.31	2.78 ± 2.19	0.92†

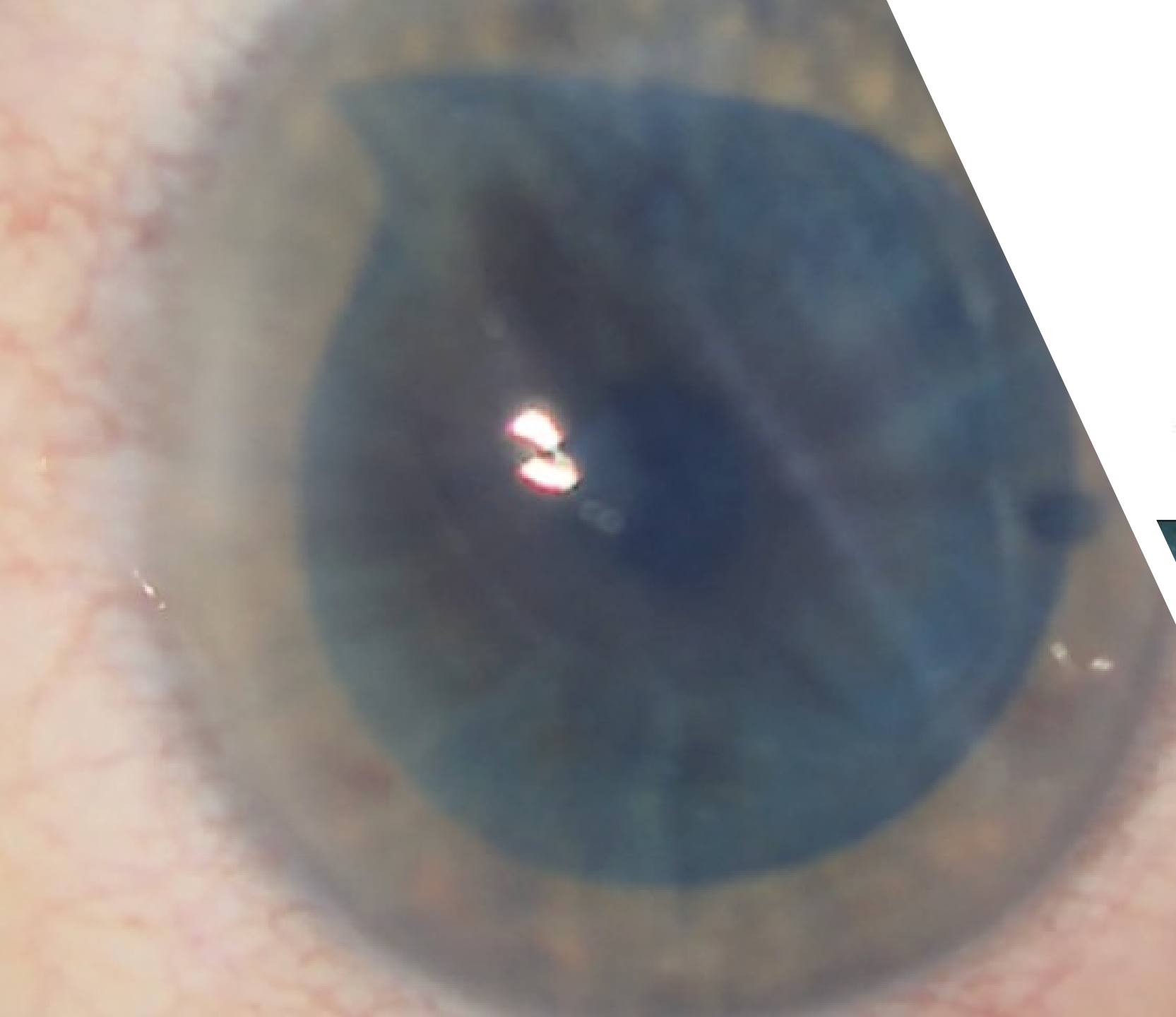


SHARK

DMEK



Shark DMEK



SUGICAL TECHNIQUE

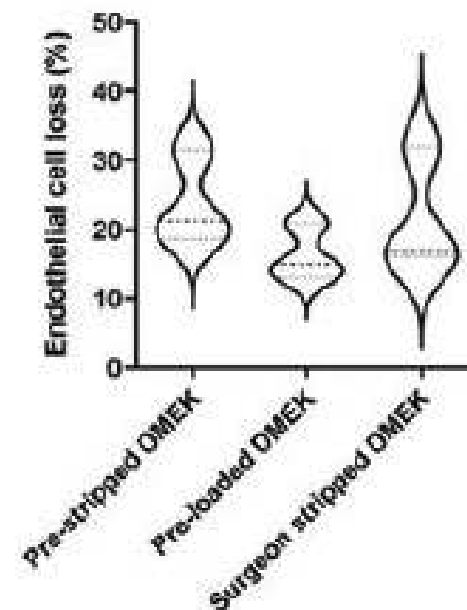
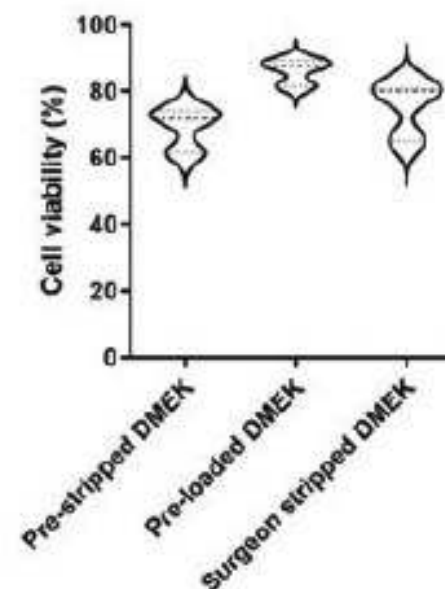
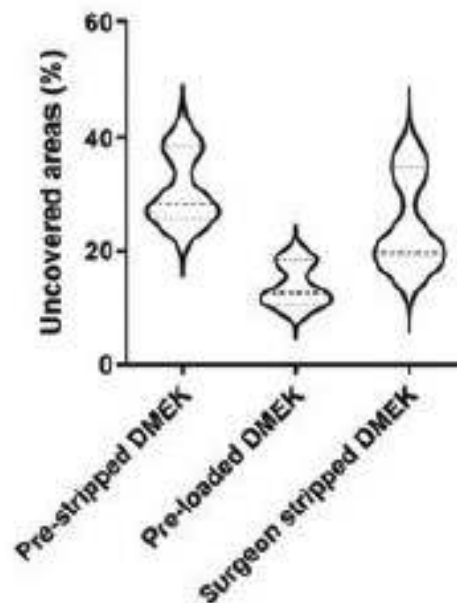
DMEK EYE BANK PREPARED



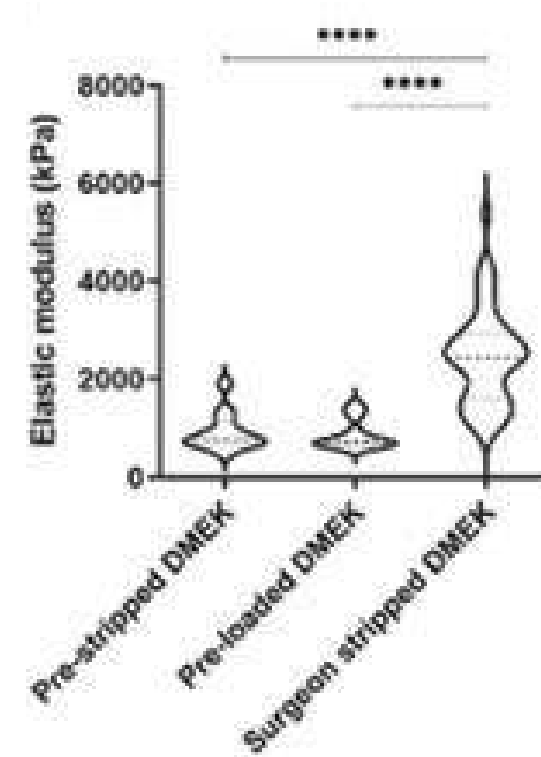
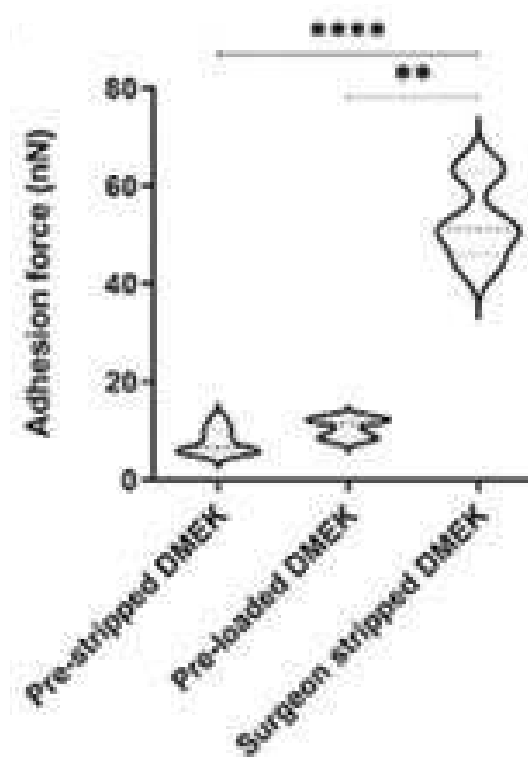
Eye bank versus surgeon prepared DMEK tissues: influence on adhesion and re-bubbling rate

Vito Romano ^{1,2,3} Ahmed Kazaili,^{4,5,6} Luca Pagano,⁷ Kunal Ajit Gadhvi,² Mitchell Titley,³ Bernhard Steger ⁷ Luis Fernández-Vega-Cueto,¹ Alvaro Meana,¹ Jesus Merayo-Llves,¹ Ponzin Diego,⁸ Riaz Akhtar,⁶ Hannah J Lewis ³, Stefano Ferrari,⁶ Stephen B Kaye,^{2,3} Monit Parekh⁹

BJO



	ps-DMEK	pl-DMEK	ss-DMEK	P value
Patient details				
N	20	31	40	
Gender (M/F)	10/10	10/21	19/21	0.3
Age (years)	73.8±9.5	77.1±9.2	72.4±8.6	0.09
Outcome analysis				
Rebubbling	40%	48.40%	15.40%	0.009
Combined cataract surgery	80%	61.30%	47.50%	0.06
Preoperative BCVA (LogMAR)	0.59±0.46	0.86±0.65	0.68±0.56	0.23
Postoperative BCVA (LogMAR)	0.29±0.34	0.41±0.52	0.35±0.48	0.65



Factors Affecting the Success Rate of Preloaded Descemet Membrane Endothelial Keratoplasty With Endothelium-Inward Technique: A Multicenter Clinical Study

MOHIT PAREKH, EMILIO PEDROTTI, PIETRO VIOLA, PIA LEON, ENRICO NERI, LORENZO BOSIO,
ERIK BONACCI, ALESSANDRO RUZZA, STEPHEN R. KAYE, DIEGO PONZIN, STEFANO FERRARI, AND
VITO ROMANO

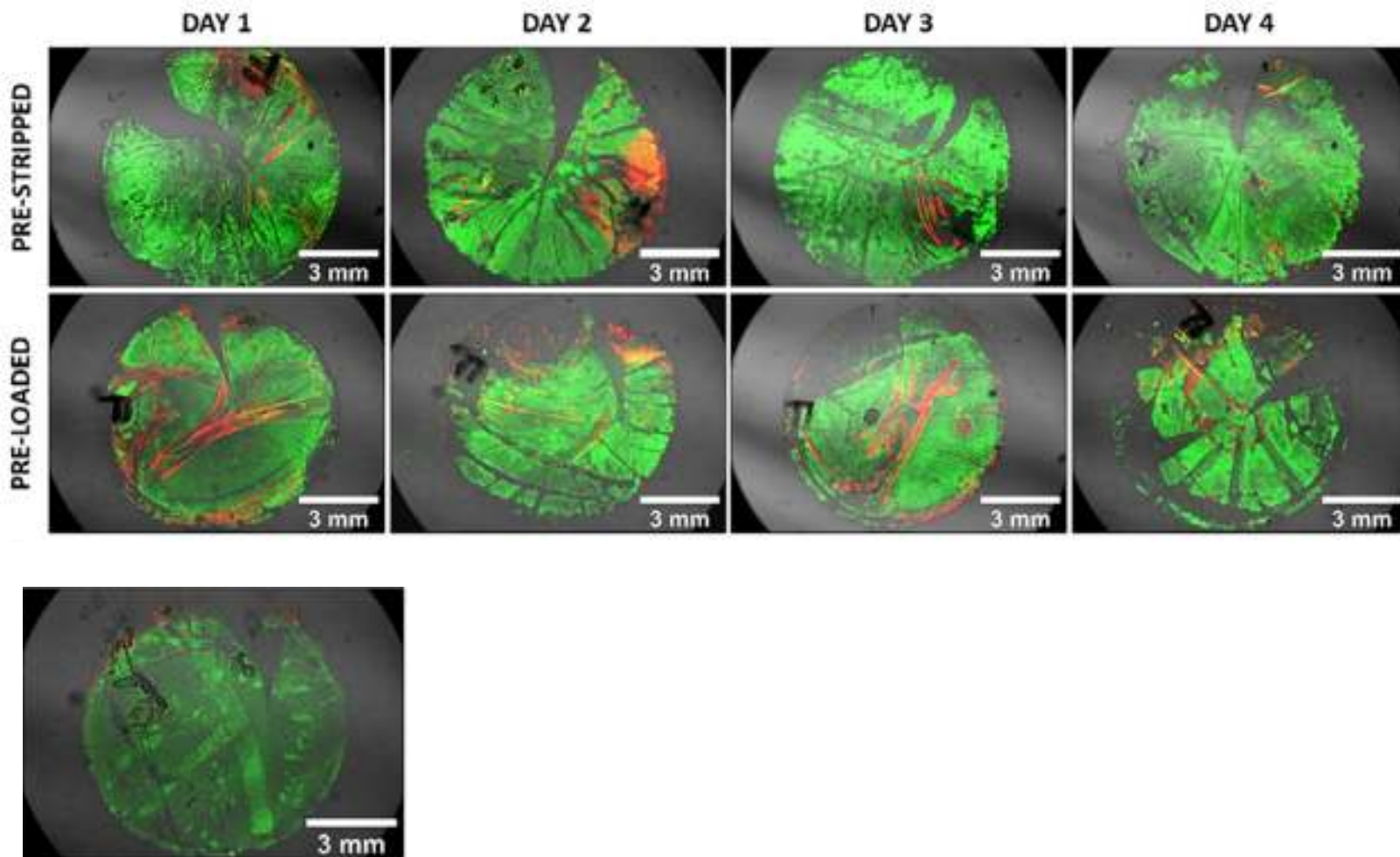
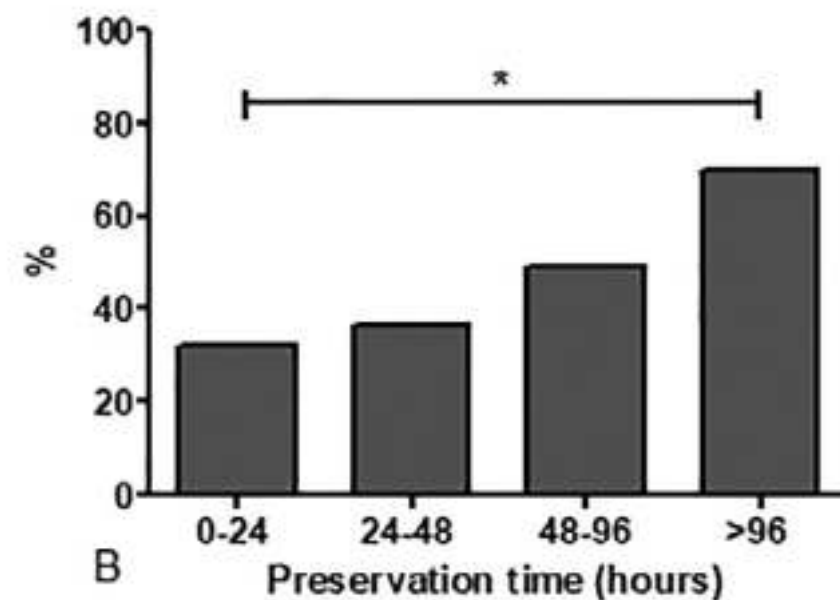


TABLE 1. Viability of Control at Day 0, Prestripped and pl-DMEK After Preserving the Tissues for 1, 2, 3, and 4 Days

Time Point	Control Prestripped DMEK (%)	
Day 0 (n = 3)	72.8±7.5	
	Prestripped DMEK (%)	PI-DMEK (%)
Day 1 (n = 4)	56.5±14.7	58.2±9.6
Day 2 (n = 4)	71.3±15.1	63.4±2.1
Day 3 (n = 4)	65.1±9.7	60.2±14.9
Day 4 (n = 4)	58.3±15.2	43.7±3.3

Preservation time Vs Rebubbling





Multicenter Early Experience of Preloaded Descemet Membrane Endothelial Keratoplasty With Endothelium-Inwards Technique With Dextran-Free Preservation Media

Alexander Wallace ^{1 2}, Mohit Parekh ³, Matteo Airoldi ², Vito Romano ^{4 5 6}, Alfredo Borgia ², Enrico Neri ⁷, Pietro Viola ⁷, Alessandro Ruzza ⁶, Stefano Ferrari ⁶, Diego Ponzin ⁶, Kunal A Gadhvi ^{2 4}

TABLE 2. Demographics

Demographic	Liverpool	Vicenza	Pooled
Surgery, number	47	18	65
Age (mean $\pm$ SD), years	75 $\pm$ 7	74 $\pm$ 4	74 $\pm$ 9
Gender, number	Male, 16	Male, 3	Male, 19
	Female, 31	Female, 15	Female, 46
Indication, number	FECD, 35	FECD, 15	FECD, 50
	BK, 12	BK, 3	BK, 15
Preoperation BCVA, LogMAR	0.69 $\pm$ 0.45	0.88 $\pm$ 0.20	0.84 $\pm$ 0.55
CCT, μ m	670.2	628.8	664.5
Single DMEK, number	23	8	31
Triple DMEK, number	24	10	34

33%
rebubbling rate

Thank you

