

# Are We Wasting Tissue for DSAEK? Viability Staining of Pre-Cut Tissue Deemed Unsuitable via Slit-Lamp and Specular Microscopy Exam

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**PURPOSE:** To utilize calcein AM to quantitatively assess endothelial cell damage to pre-cut corneal grafts deemed unsuitable by standard eye bank techniques of slit lamp and specular microscopy.

**RESULTS:** Cell viability staining demonstrated 13.4 % ECL at an avg of 5 days post-processing for tissues deemed unsuitable for transplantation, with a cell loss from 9 to 16%. Compared to 12.5 % ECL in the control group sample, this difference was not statistically significant (p=0.445)

**METHODS:** Specular and slit-lamp microscopies were utilized to determine suitability for EK preparation and subsequent potential transplantation according to Eye Bank Association of America standards. Evaluations were performed on corneas pre and post EK preparation with Moria microkeratome . During the time period of Oct. 2011 to March 2012 there were 7 DSAEK prepared tissues deemed unsuitable for transplantation due to difficulty visualizing cells and atypical appearance of cell morphology upon specular and slit-lamp exam. The unsuitable corneas (n=7) and a sampling of suitable corneas (n=6) were trephined and stained with calcein AM, images were captured with florescent microscope and endothelial cell loss (ECL) was calculated using Adobe Photoshop. Mean ECL was compared between groups using Mann Whitney U-test in SPSS v. 19.

	Pre-Resection Specular	Post -Resection Specular	Exclusion Criteria	Calcein AM Photomontage	Analysis % ECD
Tissue 1	 ECD = 2469		Atypical appearance and poor visualization of endothelium		 16%
Tissue 2	 ECD = 2994		Atypical appearance and poor visualization of endothelium		 14%
Tissue 3	 ECD = 3021		Atypical appearance and poor visualization of endothelium		 9%
Tissue 4	 ECD =2611		Atypical appearance and poor visualization of endothelium		 13%
Tissue 5	 ECD =2519		Atypical appearance and poor visualization of endothelium		 13%
Tissue 6	 ECD =2591		Atypical appearance and poor visualization of endothelium		 14%
Tissue 7	 ECD =2653		Severe endothelium drop-out by slit-lamp evaluation		 14%

**CONCLUSIONS:** We have demonstrated that some grafts deemed unsuitable by traditional methods of exclusion with specular and slit lamp evaluation may have minimal endothelial cell damage and could still be used for transplantation based on calcein am staining. Viability staining of the entire graft after processing could be helpful in determining suitability for transplantation and avoid unnecessary tissue wastage. Further investigations regarding long-term viability of this endothelium with atypical appearance upon slit-lamp and specular microscopy is warranted.

**DISCLOSURES:** None

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