When Is the LASIK Cornea Healed?

Paul M. Larson, MMSc, MBA, COMT, COE
Associate in Ophthalmology
Program Director, MMSc in Ophthalmic Technology
Emory University School of Medicine
Atlanta, GA

Support and Thanks

- Research to Prevent Blindness
- Emory University Ophthalmic Research Department
  - Henry F. Edelhauser, PhD, Chairman
  - Daniel G. Dawson, MD
  - Hans E. Grossniklaus, MD

Corneal Clarity

- Corneas clear for a number of reasons
  - Lattice orientation of corneal fibers
  - Refractive index fluctuations over small distances
  - Structures within corneas are smaller than ½ wavelengths of light or few in number
- Any variation to above might make for loss of clarity and decreased acuity

Corneal Healing – 1st 6 Months

- Initial injury (surgery)
  - Epithelial
  - Stromal
  - Corneal Nerves
    - Never recover full sensitivity even if density normal range
- Apoptosis *(cell death)* around Wound areas
  - Keratocytes
  - Epithelial Cells
- Proliferation & Migration
  - Surviving keratocytes
  - Epithelial cells
  - Re-population of acellular wound areas
- Keratocyte differentiation
  - 3 types
    - Migratory, Activated, and Myofibroblast
    - Some help heal, some do not since keratocyte healing is so primitive

Healing after 6 months

- Wound remodeling
  - Up to 3-4 years PO
- "Completion" of responses
  - Epithelial
  - Anterior stromal (between flap and epi)
  - Flap Interface
  - Mid and deep stromal
  - Corneal Nerves
  - Endothelium
- Compared to animal models, humans heal:
  - Less aggressively
  - More slowly
  - Not as completely
- *Theoretically:*
  - All complete by 4 years post-operatively
  - Now thought to be **much longer!**

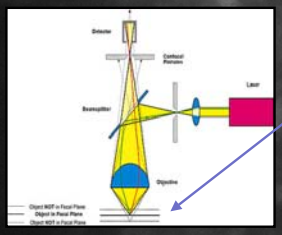
What Happens?

- Epithelial "circle" heals quickly
  - Pt. quickly regains comfort and VA
  - WOW factor
- Corneal Nerves
  - HUGE decrease immediately after surgery in density, slow recovery if at all
  - Reason we watch dry eye (anesthetic effect on blinking)
- Endothelium
  - Permanent damage if w/in 200 mμ
- Stromal Changes
  - All levels see keratocyte decreases
  - Migration and re-population occurs very slowly
  - Increases or aberrant density variations may give post-op haze (general or localized)
    - Increase IOP (or decreased ECC with age) might cause fluid at interface because of weakness & wound healing type
    - Be aware of future implications of ECC density decreases!

Corneal Confocal Microscopy

- Allows views into clear corneas and corneas otherwise "too cloudy to see into" using optical magnification at the keratocyte level
  - Corneas clear for a number of reasons
    - Lattice orientation of corneal fibers
    - Refractive index fluctuations over small distances
    - Structures within corneas are smaller than ½ wavelengths of light or few in number
  - Injured corneas normally have increased keratocytes which might add haze in addition to edema, scars, or other factors noted w/ Slit-Lamp

Confocal Microscopy



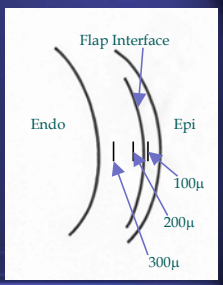
- Only objects in the focal plane are "assembled" for the image because the pinhole is **con**jugate to the **focal** point of the objective, and this allows the "optically thin" slice to seen clearly (*since the out-of-focus portions are eliminated*)

Confoscan 3 – Nidek Technologies

- Genteal Gel added to tip
- Optically couples objective to tear film and cornea (same index of refraction)
- Gel is tixotropic (breaks down in presence of endogenous salts in the patient's tears)

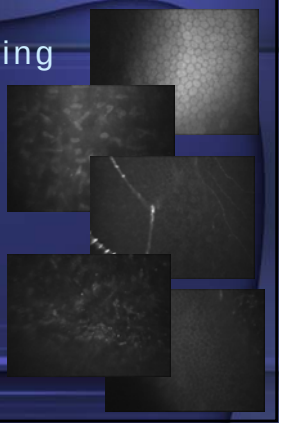
Recording Locations in Cornea

- Images taken from:
  - Endothelium (cell density)
  - Stroma
    - 300µ depth
    - 200µ depth
    - 100µ depth
  - Flap Interface (LASIK group only)
  - Epithelium



Confocal Imaging

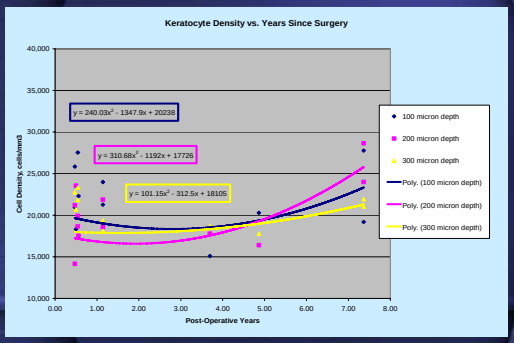
- What do we see?
  - Representative images shown at R
    - Endothelium
    - Stroma
      - Keratocytes
    - Corneal Nerves
    - Flap Interface
    - Epi (Columnar cells)



Results – Corneal Nerves

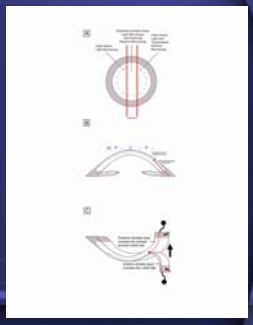
- Decreased reflectivity and number noted in nearly all LASIK patients post-op

Keratocyte Density



Wound Strength

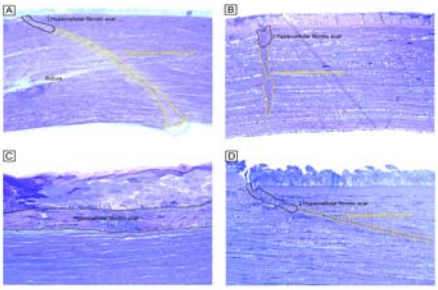
- Study done by Dawson, et al
- Post-mortem LASIK corneas used
- Histopathology and tensile strength looked at in nearly similar locations



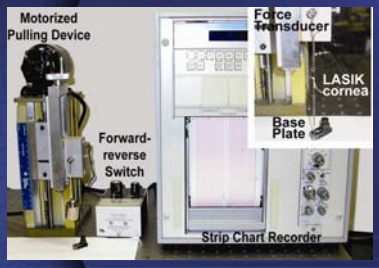
Scar formation differences

- At epithelium-stroma interfaces (peripherally):
  - Much stronger wound
  - ***Hypercellular fibrous scar***
  - 50 to 75 mμ thick
  - Epithelial ingrowth noted in 53% as a focal or diffuse finding
- Away from edge (& where no epithelial ingrowth was noted):
  - MUCH weaker
  - ***Hypocellular & primitive scar***
  - Average 5 mμ thick

Histopathology



Wound Tensile Strength



- Hooks used through corneal edges and tracing of tensile strength needed to pull cornea apart was graphed in real time

Wound Tensile Strength

- Findings:
  - No change in tensile strength over time
  - Normal cornea 22-36 g/mm cohesive tensile strength
  - Normal corneas greatest strength in peripheral regions and in the anterior 1/3 of stroma
  - Epith. Scar measured 30 g/mm average
  - Central area of flap **ONLY 0.7 g/mm** average
    - This is 2.4% of expected (and NO CHANGE OVER TIME!!!!)

Young's Modulus

- Elastic strength of Cornea
- Merely cutting a flap most likely weakens entire cornea
  - Currently only animal models studied
    - Remember human healing!
    - Slower, less aggressive, not as complete
  - May be one contributing reason for ectasia when thickness seems adequate
  - Modulus currently not measured BUT newer instruments:
    - Reichert Ocular Response Analyzer measures corneal hysteresis
    - Might explain why LASIK corneas measure lower than thickness change alone might suggest (remember OHTS data re.: IOP/thickness relationship?)

Conclusions

- Healing may not be complete for 6-7 yrs
  - **If ever!**
- Keratocyte and nerve density may recover eventually
- LASIK findings do not match PRK evidence to date in any of these studies
- Wound strength of LASIK flap only good at edge
- Very poor central flap adhesion
  - **Probably indefinitely!**
- Popularity of procedure has exceeded research at actual mechanisms of "*what happens??*"

Thank You!

- Questions??
- My email/phone:
  - plarson@emory.edu
  - 404.778.4305

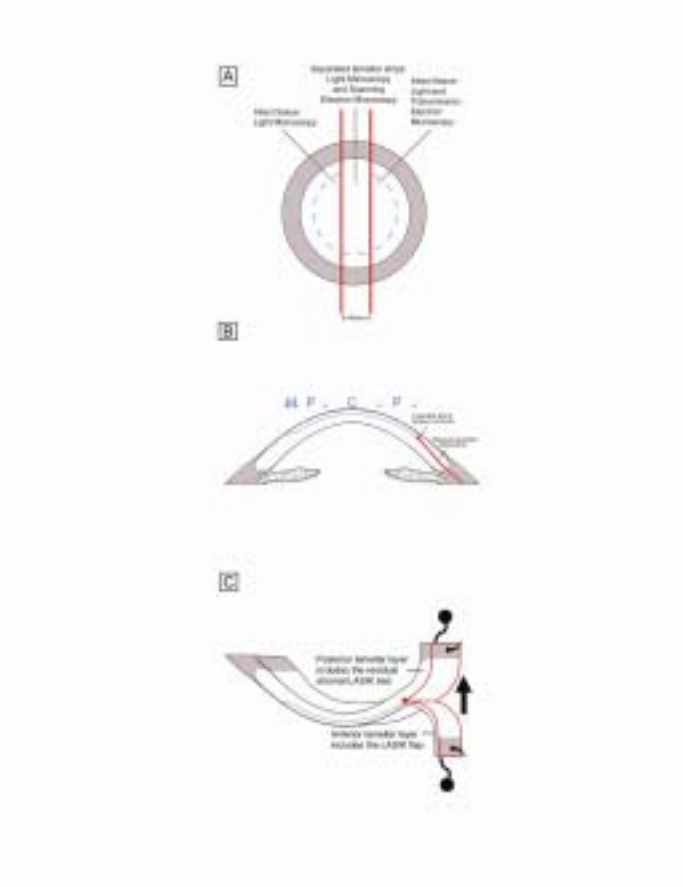
When is the "LASIK Cornea" Truly Healed?

Presented October, 2005 to the Association of Technical Personnel in Ophthalmology
Twentieth Scientific Session, Chicago, Illinois

Paul M. Larson, MMSc, MBA, COMT, COE
Associate in Ophthalmology, Program Director, MMSc in Ophthalmic Technology
Department of Ophthalmology, School of Medicine
Emory University, Atlanta, GA

I. Corneal Healing in General
   A. Active wound healing (*up to 6 months post-operatively*)
      1. Initial injury (epithelial and stromal) leads to apoptosis of keratocytes and epithelial cells in and around wound; extracellular tissue disruption
      2. Proliferation and migration of surviving keratocytes and epithelial cells to re-populate acellular wound areas
      3. Differentiation of keratocytes into three distinct types:
         a. Migratory keratocyte
         b. Activated keratocyte
         c. Myofibroblast
   B. Wound re-modeling (*up to 3-4 years post-operatively*)
   C. Completion of healing responses
   D. General Comments on Human corneal healing
      1. Compared to animals, humans <u>heal less aggressively, more slowly, and not as completely</u>
      2. Theoretically, human post-operative LASIK corneas should be healed completely by 4 years post-operatively (but sub-clinical re-modeling may occur indefinitely)

II. Flap-Stroma Interface
   A. Wound strength:
      1. Varies based on scar characteristics (hypo- vs. hyper-cellular)
      2. Where epithelium-stroma interactions occur, a much stronger scar exists
   B. Centrally vs. Peripherally
      1. Since most epithelial-stromal apposition is located peripherally at the location of the flap edge, this would tend to be the strongest area of the wound.
      2. Epithelium usually only enters slightly into the wound edge, and central to this the only repair mechanism available is that from the keratocyte injury and is considerably weaker.
   C. Method: illustrated in below figure: ***donor corneas used***

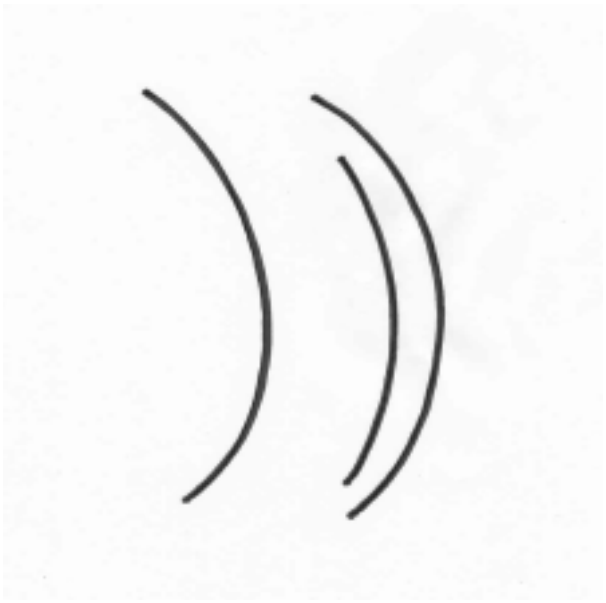


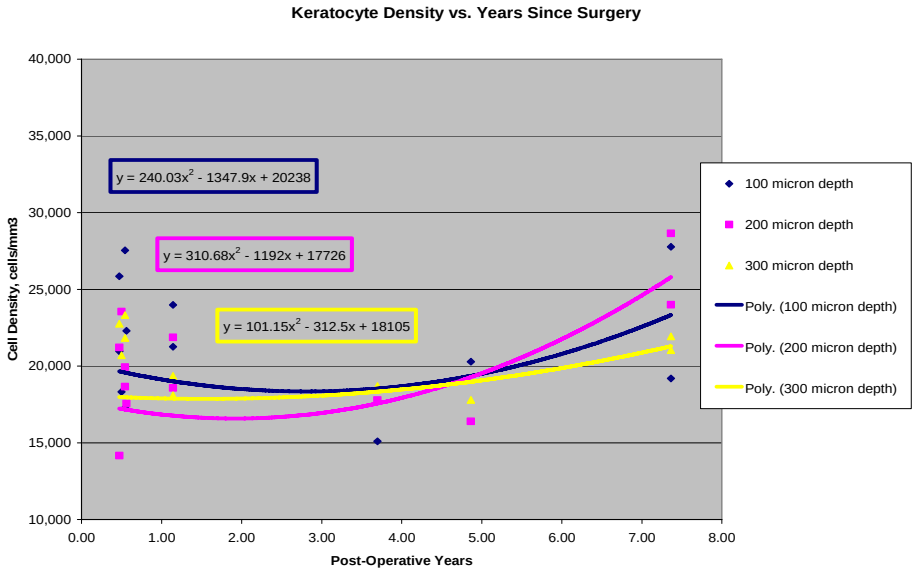
III. Stroma
- A. Clarity factors
- B. Haze factors
  1. Keratocytes
  2. Macrophages
  3. Fluid Pockets
  4. DLK
  5. Epithelium and Bowman's problems
- C. Stromal folds
  1. Ectasia
  2. Flap folds/edema

IV. Corneal Nerves and Keratocytes over Time

A. Most literature has shown losses of nerves and keratocyte density tends to stabilize at around 4 years. I found that there may be a trend to re-populate the keratocytes after this 4 year period with levels returning to near pre-operative levels at each level, with anterior levels being higher. This would usually be accompanied by some nerve regeneration as well.

B. Confocal Microscopy uses





V. Conclusions

A. Some wound healing may be noted for over 7 years post-operatively although the cornea may remain clear and refraction remain stable.

B. Because of the long healing period, patients should be monitored at least annually for development of epithelial ingrowth, keratectasia, as well as topographic, refractive, and pachymetric changes.

C. The wound tensile strength remains “indefinitely” weak in the center where the hypocellular primitive stromal scar is located.

VI. References

Anderson NJ, Edelhauser HF, *et. al*. Histologic and Ultrastructural Findings in Human Corneas after Successful Laser in situ Keratomileusis. Arch Ophthalmol. 2002; 120:288-293.

Bühren J and Kohnen T. Stromal Haze after Laser in situ Keratomileusis. Clinical and Confocal Microscopy Findings. J Cataract Refract Surg. 2003; 29:1718-1726.

_____. Corneal Wound Healing after Laser in situ Keratomileusis Flap Lift and Epithelial Abrasion. J Cataract Refract Surg. 2003; 29:2007-2012.

Calvillo MP, McLaren JW, *et.al.* Corneal Reinnervation after LASIK: Prospective 3-Year Longitudinal Study. Invest. Ophthalmol. 2004; 45(11):3991-3996.

Dawson DG, Holley GP, *et. al. Ex vivo* Confocal Microscopy of Human LASIK Corneas with Histologic and Ultrastructural Correlation. Ophthalmology. 2005 (in press).

_____, Edelhauser HF, Grossniklaus HE. Long-term Histopathologic Findings in Human Corneal Wounds After Refractive Surgical Procedures. Am J Ophthalmol. 2005; 139:168-178.

Erie JC, Nau CB, *et. al*. Long-term Keratocyte Deficits in the Corneal Stroma after LASIK, Ophthalmology. 2004; 111:1356-1361.

Geggel HS and Talley AR. Delayed Onset Keratectasia following Laser in situ Keratomileusis. J Cataract Refract Surg. 1999; 25:582-586.

Geroski DH, Dawson DG, *et. al*. (NEI grant R01 EY 00933, P30 06360, and T32 EY 07092). Histopathologic and Ultrastructural Correlation of LASIK Interface Opacities using Confocal Microscopy. Poster presented at Association for Vision Research in Ophthalmology 2004 annual meeting.

Jester JV, Petroll WM, Cavanaugh HD. Corneal Stromal Wound Healing in Refractive Surgery: the Role of Myofibroblasts. Progress in Retinal and Eye Research. 1999; 18(3):311-356.

Kramer TR, Chuckwaipong V, *et. al*. Pathologic Findings in Postmortem Corneas After Successful Laser In Situ Keratomileusis. Cornea. 2005; 24:92-102.

Mastropasqua L and Nubile M. Confocal Microscopy of the Cornea. 2000. SLACK. Thorofare.

McCally RL and Farrell RA. Structural Implications of Small-angle Light Scattering from Cornea. Exp. Eye Res. 1982; 34:69-81.

Mitooka K, Ramirez M, *et.al.* Keratocyte Density of Central Human Cornea After Laser in situ Keratomileusis. Am J Ophthalmol. 2002; 133:307-314.

Mohan RR, Hutcheon AEK, *et. al.* Apoptosis, Necrosis, Proliferation, and Myofibroblast Generation in the Stroma following LASIK and PRK. Exp. Eye Res. 2003; 76:71-87.

Møller-Pedersen T. Keratocyte Reflectivity and Corneal Haze. Exp. Eye Res. 2004; 78:553-560.

Mustonen RK, McDonald MB, *et. al*. Normal Human Corneal Cell Populations Evaluated by *In Vivo* Scanning Slit Confocal Microscopy. Cornea. 1998;17(5):485-492.

Vesaluoma M, Perez-Santonja J, *et. al.* Corneal Stromal Changes Induced by Myopic LASIK. Invest Ophthalmol Vis Sci. 2000; 41:369-376.

_____, Petroll WM, *et. al.* Laser in situ Keratomileusis Flap Margin: Wound Healing and Complications Imaged by In Vivo Confocal Microscopy. Am J Ophthalmol. 2000; 130:564-573.

Wilson SE. Role of Apoptosis in Wound Healing in the Cornea. Cornea. 2000; 19(Suppl. 1):S7-S12.