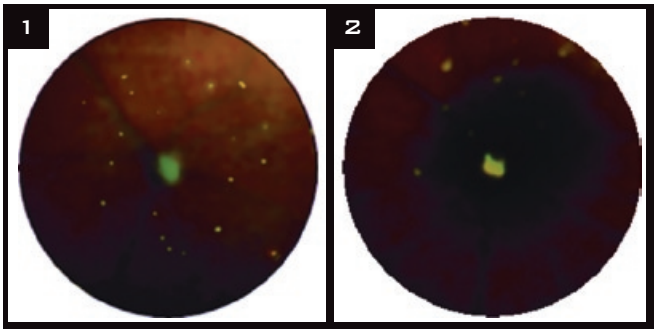
News in Review

A LOOK AT TODAY'S IDEAS AND TRENDS

A Window Into Alzheimer's Disease



SEEING GREEN. Retinal images of a living 14-month Alzheimer's mouse (1) compared with an aged, control living mouse (2). Many more retinal nerve cells are in the early phase of apoptosis (green spots) in the Alzheimer's mouse.

Researchers at the University College London's (UCL) Institute of Ophthalmology have developed a technique that makes it possible to directly and noninvasively monitor the death of single retinal nerve cells in the eyes of live animals in real time. The potential of this technique reaches far beyond that of retinal health—whether it provides a diagnostic "canary in the coal mine" by signaling early death of nerve cells in the brain or offers a tool for evaluating the efficacy of neuroprotectants for Alzheimer's disease.

The UCL research team previously added to the growing body of data linking a wide range of neurodegenerative diseases to common triggers and a convergence of final pathways by providing evidence that the protein beta amyloid—a major component of Alzheimer's brain plaques—is responsible for harm to the optic nerve. These and other connections prompted the team, led by M. Francesca Cordeiro, MD, PhD, and Stephen E. Moss, PhD, to broaden the focus of their current study beyond glaucoma.

"An increasing number of studies have come out showing that the retina is affected by Alzheimer's disease," said Dr. Cordeiro, professor of glaucoma and neurodegeneration studies at University College London and an attending physician at the Western Eye Hospital in London. "But these studies all were confined to postmortem eyes. Ours was the first showing retinal activity in vivo."

Reporting in *Cell Death & Disease,* the researchers delineated a series of steps to observe retinal cells and monitor the stage and type of cell death.[1] This involved the use of fluorescent cell-death markers that bind to specific cells on the retina of transgenic mice with aspects of Alzheimer's disease. To track individual live cells over hours, days, weeks and months, the researchers used a customized confocal scanning laser ophthalmoscope—not far afield from those used clinically—to detect emission wavelengths from three fluorescent labels: annexin V positive only (to visualize early apoptosis), propidium iodide only (to visualize necrosis) and both annexin V and PI positive (to visualize late-phase apoptosis).

"We combined the two markers [annexin V and PI positive] to give us an idea of the severity or activity of cell death and to differenti-

EyeNet *thanks Christopher J. Rapuano, MD, for his help with this issue's News in Review.*

ate between the different phases—whether complete death or an early stage with the capacity for reversal," said Dr. Cordeiro.

One important finding was that necrosis plays a key role in neurodegeneration, she said, meaning apoptosis is not an exclusive player in this process, as previously thought.

By making it possible to stage neurodegeneration in real time with a simple eye test, these studies open the possibility for Alzheimer's treatment during the narrow window of early apoptosis. They also help make the case for broadening the role of the general ophthalmologist, said Dr. Cordeiro. "Eventually, I think this will knock on the door of the neurologist a bit," she said.

Dr. Cordeiro and collaborators hope to extend these techniques to clinical trials in glaucoma patients later this year. —*Annie Stuart*

1 Cordeiro, M. F. et al. *Cell Death Dis.* Published online Jan. 14, 2010.

Dr. Cordeiro is a named inventor on a patent application covering the technology disclosed in the Cell Death & Disease *report.*

Cornea Report

Donor Cell Density Can't Predict PK Success

Cornea transplant success after penetrating keratoplasty (PK) cannot be predicted by donor endothelial cell density, but it can be predicted by cell density six months postoperatively,[1] according to the Cornea Donor Study Investigator Group, which previously found that donor age does not affect graft success.

In the prospective cohort study, both the grafts that failed and those that remained clear at five years started out with similar median cell counts, 2,670 cells/mm^2 and 2,687 cells/mm^2, respectively.

This finding should ease surgeons' concerns about obtaining corneas with the highest number of cells, said lead investigator Jonathan H. Lass, MD, professor and chairman of ophthalmology and visual sciences at Case Western Reserve University and director, University Hospitals Eye Institute in Cleveland, Ohio. "Minimum count at most eye banks is about 2,000 cells/mm^2 or above," he said. "What this study is saying is, as long as you're within the minimum, the baseline count didn't make a difference."

Flash-forward to six months after surgery: Now cell count matters. The six-month endothelial cell density, and the change from baseline, were predictive of graft failure at five years of follow-up.

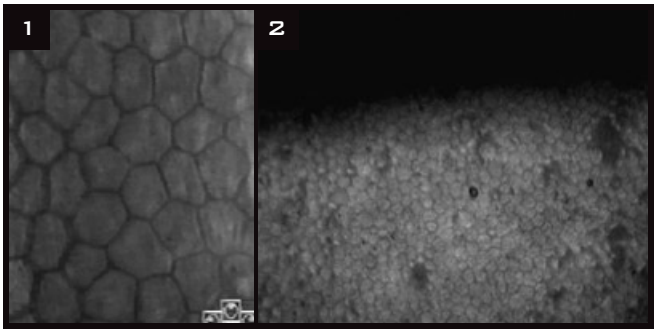
By six months, the median endothelial cell density in the failed group was 1,774 cells/mm^2. In the cases that did not fail, the median endothelial cell density was 2,514 cells/mm^2.

These findings have clear clinical implications. "If you have a low count at six months, watch those patients because they have a higher risk for failure," Dr. Lass said. Monitor them more frequently, and if there's a noticeable increase in corneal thickness between visits, get a cell count, he said.

Interestingly, the study found that a graft can remain clear with an endothelial cell density below 500 cells/mm^2. Success appeared related to the trajectory of cell loss, Dr. Lass said. "If your cornea has a stable population of cells that are functioning at a relatively low cell count, that graft can do well."

Now the NIH-funded study group is looking at factors that influence cell loss, such as length of time the cornea was in storage prior to surgery as well as recipient diagnosis.

"Our main goal is to try to change practice patterns in how surgeons approach use of donor corneas," Dr. Lass said. —*Miriam Karmel*



SEEING CLEARLY. A penetrating keratoplasty patient with a clear graft at five years (1). Endothelial cell density is 428 cells/mm^2, and there has been 87 percent cell loss from the original donor count of 3,207 cells/mm^2 (2).

JONATHAN H. LASS, MD

1 Lass, J. H. et al. *Arch Ophthalmol* 2010;128:63–69.

HISTORY : Egyptian Cosmetics

Makeup may have been a key to eye health in ancient Egypt. Electrochemical data obtained from 52 makeup samples taken from ancient containers housed at the Louvre museum revealed that two lead chlorides were used in the manufacture of eye makeups and lotions. Considering our current understanding of lead toxicity, these findings are somewhat surprising. However, it is presumed that the application of these compounds led to the production of nitrogen monoxide molecules—a catalyst in the immune response, which could have offered the Egyptians some protection from bacterial eye diseases and inflammations common to the areas surrounding the marshy Nile River. —*Leslie Burling-Phillips*

Contact Lens News

Antimicrobial Contact Lenses in the Pipeline

As early as this month, an Australian research group could begin a 250-patient trial of one of the leading passive strategies for protecting contact lens wearers from bacteria on their silicone hydrogel lenses: coating the lenses with selenium.

Unlike some other antibacterial add-ons being considered, the selenium does its work without being released into the eye, said microbiologist Mark Willcox, PhD, professor of optometry and vision science at the University of New South Wales in Sydney, Australia.

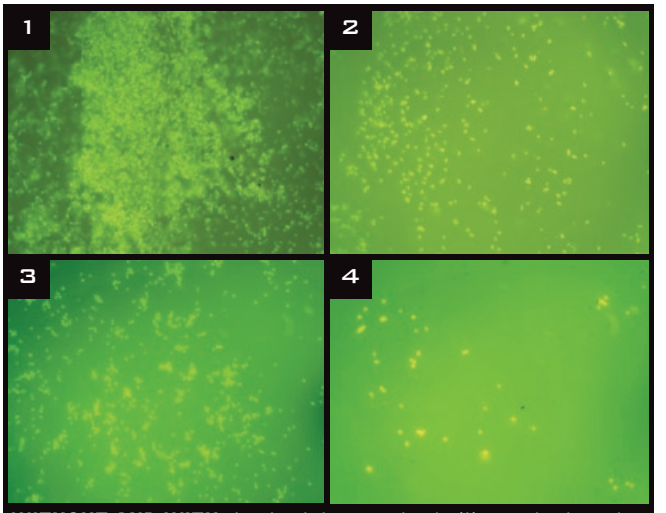
Instead, the selenium is incorporated into an organic compound and covalently bonded to the lens. There, it causes the localized generation of superoxide free radicals that injure any bacteria present, preventing them from growing and adhering to the lens in a persistent biofilm.[1]

"The selenium acts on a local, microscopic scale in a way similar to that of hydrogen peroxide," said Dr. Willcox, chief scientific officer at the Institute for Eye Research, where the selenium-coated contact lenses will be tested. "The smaller the cell, the nearer it can get to the surface of the lens and the more it is affected by the free radicals. Human cells of the cornea and conjunctiva aren't affected."

So far, the research findings on selenium coating of contact lenses include:

- In the lab, silicone hydrogel lenses that were covalently coated with selenium grew 1,000 times less *Staphylococcus aureus* than uncoated versions did. They also resisted colonization by *Pseudomonas aeruginosa*.
- After 48 days of continuous wear by rabbits, eyes that wore the test lens showed no significant differences compared with control eyes in clinical signs, epithelial and corneal total thickness, corneal morphology and corneal histology.
- In a controlled, randomized, contralateral human trial, 20 subjects wore selenium-coated and uncoated silicone hydrogel contact lenses for 24 hours. The researchers found no differences between the eyes in the patients' subjective evaluations, bulbar and limbal redness, and corneal and conjunctival staining.[2]
- Despite 24 hours of continuous wear, the coated lenses retained their antibacterial properties.

Dr. Willcox said that lab tests indicate that selenium might also make silicone hydrogel contact lenses resist colonization by the fungus *Fusarium*, one of the organisms behind the 2004 to 2007 outbreaks of microbial keratitis among contact lens wearers. It has not been tested yet against *Acanthamoeba*, another common cause of outbreaks.



WITHOUT AND WITH. In the lab, standard silicone hydrogel lenses showed greater susceptibility to contamination by *S. aureus* (1) and *P. aeruginosa* (3) than did contact lenses coated with selenium (2,4).

Researchers at the Sydney institute and elsewhere around the world have also been exploring other molecules that might be added to contact lenses to inhibit microbes. These include silver; polymeric quaternary ammonium compounds, to act as disinfectants; polymeric pyridinium compounds, to break bacterial cell walls; quorum-sensing compounds that interfere with bacterial signaling systems; nitric-oxide releasing polymers, which produce free radicals; and natural or synthetic peptides that cause microbial cell walls to leak.

Although the goal is to prevent infectious keratitis, the initial trial of selenium-coated contact lenses will not give a definitive answer on this issue. The study's size will limit researchers to tracking incidence of adverse events associated with infection risk, such as conjunctival inflammation and infiltrative keratitis.

Nonetheless, as the first large clinical trial of antibacterial contact lenses, the trial will be an important milestone in an ongoing effort. Institute for Eye Research scientists began hunting for ways to make antimicrobial contact lenses in 1999, Dr. Willcox noted. "It's taken us a long time to understand what causes these adverse events and what you can do to prevent them. So this is a big step for us," he said. —*Linda Roach*

1 Mathews, S. M. et al. *Cornea* 2006;25(7):806–814.
2 Ozkan, J. et al. Poster #5632/D941, Efficacy and clinical performance of selenium antibacterial silicone hydrogel contact lenses. Presented at ARVO, Thursday, May 7, 2009. Abstract available online at www.arvo.org. Choose "Meetings & Abstracts," then "Search 2009 Annual Meeting Abstracts." Search under "Program" for 5632.

Dr. Willcox has consulted for and received research and travel grants from several ophthalmic companies, including Abbott Medical Optics, Alcon, Allergan and Ciba Vision.

INSTITUTE FOR EYE RESEARCH, SYDNEY, AUSTRALIA