Transplanting the Light Fantastic

By RICK WEISS

Cells from eye donors may someday restore vision in some blind individuals

Reading these words requires the cooperation of more than a quarter of a billion specialized, light-sensitive nerve cells neatly arranged along the backs of your eyes. There, upon an exquisitely organized neuronal array called the retina, these alphabet-shaped patterns of darkness and light get translated into patterns of electrical potentials. Thus encoded, the written words rush to the brain for processing and, ultimately, for decisions — such as "keep reading" or "I'm bored, turn the page."

But what about the estimated 5 million people in the United States who, through disease or injury, have lost some of their light-sensitive retinal cells? For these individuals, this eyeball-initiated biochemical and electrical cascade proceeds sloppily at best, leaving them visually compromised or completely blind. And because mammalian photoreceptor cells so far appear incapable of significant regeneration, visual recovery for these people remains impossible today.

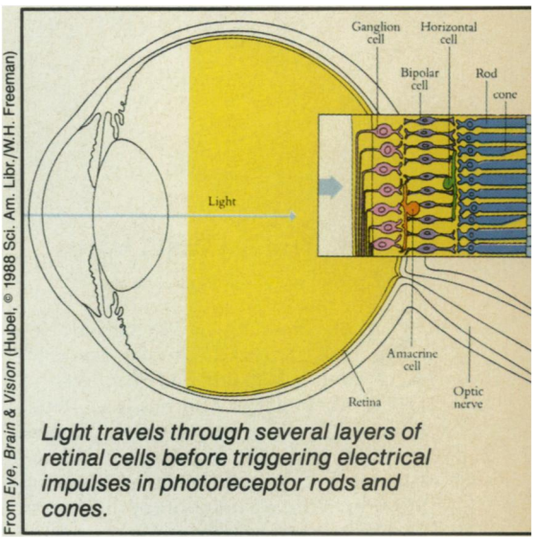
Researchers experimenting with photoreceptor transplants hope to change that prognosis. Recent progress in the ability to graft healthy, light-sensitive neurons inside the eyes of blind animals suggests partial restoration of vision for people with photoreceptor damage may someday become feasible.

"The mindset was that it would be impossible," says Martin S. Silverman, a neurobiologist affiliated with Washington University and the Central Institute for the Deaf, both in St. Louis. Now, he says, that view has begun to change. In the August INVESTIGATIVE OPHTHALMOLOGY & VISUAL SCIENCE, Silverman and Stephen E. Hughes describe the first successful transplants of gelatin patches containing organized arrays of photoreceptor cells. The grafts thrive in the eyes of blinded rats, and tests suggest the transplanted receptors respond properly to light.

Photoreceptor transplants performed in test animals have yet to yield evidence of visual improvement. Indeed, investigators remain uncertain whether the transplanted cells have made the neuronal connections necessary to send messages to the brain. But researchers experimenting with various transplant techniques express increasing optimism that grafted photoreceptors can perform the basic functions needed to tell other cells what they "see."

"It's not something you would necessarily expect to work," says University of Pittsburgh neuroscientist Raymond D. Lund, commenting on Silverman's unique method for splicing photoreceptor patches into damaged retinas. "But it looks like a very promising technique."

Indeed, says James E. Turner, a retinal transplant researcher at Wake Forest University's Bowman Gray School of Medicine in Winston-Salem, N.C., "if such a technique becomes practical, it certainly would be more than helpful for some major classes of retinal dystrophies." Retinal dystrophies refer to a class of diseases, including retinitis pigmentosa and macular degeneration, that result in the gradual deterioration of photoreceptor cells. "There's no known cure for any of these diseases," Turner says.



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Light travels through several layers of retinal cells before triggering electrical impulses in photoreceptor rods and cones.

Photoreceptor cells — known as rods and cones — form one of 10 cell layers in the human retina. Each photoreceptor cell contains a light-reactive protein called an opsin, which converts light into an electrical potential. These minute power surges must leap across tiny gaps, or synapses, to trigger similar potentials in bipolar cells and ganglion cells that reside in neighboring retinal layers. Ganglion cells then conduct the current via the optic nerve to the brain, which reconstructs a "map" of the visualized image in much the same way a television produces pictures from rows of colored dots.

Sight restoration in people with disrupted ganglion cells seems impossible for now because of difficulties in getting nerve cells to regenerate the long distances traversed by these cells. But many retinal diseases result in a loss of photoreceptors alone, while other parts of the retina remain healthy and intact. These are the diseases that researchers hope to treat with photoreceptor transplants.

Some researchers have attempted to restore light responsiveness in rats afflicted with retinal dystrophies by injecting solutions of fresh photoreceptors into the rats' eyes. Results have been largely disappointing — perhaps because the transplanted cells in these experiments become so disrupted in the process.

In an attempt to leave undisturbed the intrinsic, orderly arrangement of donor photoreceptors, Silverman takes a different approach. First, he immerses donor-rat retinas in gelatin. After chilling this biological gel, he uses a micromilling machine to shave consecutive horizontal slices until he gets to the retinal layer containing photoreceptors. Then, performing eye surgery on rats lacking photoreceptors, he slips this cell-laden gelatin slab between the appropriate layers of retinal cells. Hours later the gelatin dissolves and disappears, leaving the transplanted cells in position.

Tests indicate the cells remain alive for at least six weeks and produce large amounts of opsin. Moreover, the transplanted cells utilize increased quantities of glucose after exposure to light — providing indirect evidence that they are performing their intended electrical functions in response to light.

Those results look promising, but they fall short of demonstrating restoration of true neural function. Glucose uptake tests are fairly messy as a means of confirming specific neuronal activity, notes Lund. Light may simply accelerate the transplanted cells' metabolic rates — and hence their glucose consumption — without actually inducing within the cells an electrical potential. "We need very care-

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Neptune: A new page in the book of worlds

Scientists working with Voyager 2's closeup measurements of Neptune have barely begun their in-depth study of the data. This week, however, several presented their first reports of findings since the late August flyby, at a meeting of the American Astronomical Society's planetary division in Providence, R.I.

One striking result from the mission was the discovery on Neptune's big moon Triton of two towering plumes of gas, probably nitrogen, one leaping up about 8 kilometers from the surface (SN: 10/14/89, p.247). Scientists found photographic evidence of the plumes well after the Voyager flyby. Laurence A. Soderblom of the U.S. Geological Survey in Flagstaff, Ariz., suggested this week that the nitrogen may become heated by sunlight absorbed in dark material in Triton's surface ice and escape through vents or fissures in an ice layer 2 to 3 meters thick. Though the nitrogen has a pressure of only 1 millibar, he says, it would push up against a thin atmosphere with a surface pressure 100 times less than that.

A hallmark of any planetary encounter's early data analysis is the struggle to extract subtle details from photos of the surface, and Triton proved no exception. According to Voyager assistant project scientist Ellis Miner of Jet Propulsion Laboratory in Pasadena, Calif., many of the shapes visible on the surface are probably water ice, since ices of methane and nitrogen "would not retain the large vertical structures that we see," such as cliffs 100 to 300 meters high.

Triton and Nereid were Neptune's only known moons before the Voyager encounter, but the spacecraft's photos revealed six more. The number has not increased since, giving Neptune the fewest known moons of the four major outer planets. But Carolyn Porco of the University of Arizona in Tucson notes that photo interpreters are reanalyzing Saturn 2's pictures for signs of a small moon suspected outside Triton's orbit.

The researchers still seek to refine the length of Neptune's day. In August, scientists put it at 16 hours and 3 minutes. Now a Neptunian day appears a little longer, perhaps 16 hours and 6 to 7 minutes.

Since a deep atmosphere hides Neptune's surface, the key to fixing the length of a Neptunian day lies in analyzing the radio emissions produced by its magnetic field. Studying the emissions in detail, however, turns out to be tricky. Voyager 2 has detected both brief, or "bursty," emissions and longer-term smooth ones, says Michael L. Kaiser of NASA's Goddard Space Flight Center in Greenbelt, Md.

The bursty ones, he says, often appear in unusually narrow frequency bands, typically showing up in only one channel at a time of Voyager's planetary radio astronomy instrument, with each burst no more than 20 kilohertz wide. Uranus has some similarly narrow bursts but at much lower frequencies — below 0.1 megahertz at Uranus, compared with greater than 1 megahertz at Neptune.

One curious aspect of the radio signals, Kaiser says, is the intricate polarization pattern of the smooth ones, showing emissions with both left- and right-hand polarization even though they seem associated with just one of Neptune's magnetic poles — "the weak one, the north pole," according to Kaiser. Scientists would expect right and left polarization to go hand in hand with observations from two poles.

The planet displays a remarkably complex magnetic field. The simplest form of a planetary magnetic field is called a dipole, like that of a bar magnet. At Neptune, says Miner, the field is "not easily represented by a multipole model, or even by a dipole plus quadrupole plus octopole." — *J. Eberhart*

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ful electrophysiological studies [in these cells] to show that electrical responses can be generated in response to light," Lund says.

While Silverman has not yet provided that proof, other researchers using a different transplant technique recently reported evidence of electrical activity in their grafts. Robert J. Collier of the University of Rochester (N.Y.) and his colleagues looked at electroretinograms — tracings of retinal electrical activity that resemble the electrocardiograms used to measure heart functions — recorded from retinal tissue they had transplanted into rats.

Unlike Silverman, Collier's group grafted retinal cells into the front of rats' eyes — far from the retina where any functional transplant must ultimately reside but in a location that allowed the researchers to observe the transplanted cells more easily. At the annual meeting of the Association for Research in Vision and Ophthalmology in May, Collier showed electroretinograms indicating that the transplanted cells, like normal photoreceptors, conduct waves of electrical potentials in response to light.

Those results still don't show that transplanted cells, when placed in the rear of the eye, can grow the micron or so necessary to come within shouting distance of ganglion cells. "The retinogram tells you that these retinal cells are talking, but it does not yet tell you that they are talking to the brain," says Collier's co-worker Manuel del Cerro. Proof of that ultimate electrophysiological link, he says, will require measuring evoked potentials in the brain's visual cortex in response to illumination or to patterns shown on a screen.

No researcher has shown such responses in the brains of photoreceptor recipients, says Silverman, in part because rats have very poor visual acuity in the first place. Silverman says he's preparing to perform transplants in rabbits, cats and primates, which are easier to test for specific brain responses to visual stimuli.

Even if photoreceptor connections prove neurologically sound, researchers foresee other potential complications. For example, transplanted photoreceptors tend to grow abnormally into so-called rosette formations that create bumps on the normally smooth retinal layer. Rosettes are definitely a concern, says Silverman. "We'd expect any disruption to degrade the visual image."

Unfortunately, says del Cerro, "nobody is sure how photoreceptors orient themselves during development. We know the rosettes are linked to abnormal development, but since we do not know what normal is, it's very difficult to know what we should be doing differently in transplantation to avoid having rosettes."

Graft rejection remains another potential problem. Nervous tissue rarely triggers immune responses, making photoreceptors an ideal material for transplantation. But the presence of contaminating, non-neural retinal cells within a graft could trigger an immune response, warns del Cerro. In theory, that could lead not only to graft rejection but also to a sight-threatening antibody attack against the recipient's other eye.

So far, Silverman notes, no such complications have arisen. And even if immunosuppressant drugs become necessary in some cases — as they were in his recent successful transplants of human retinal cells into rats — they can be applied directly to the eye to prevent the side effects that go along with systemic use of such drugs.

Nobody knows to what extent photoreceptor transplants may restore vision in people with retinal diseases. However, del Cerro emphasizes, patients regard even poor vision as a vast improvement over no vision at all.

Before transplantation, "the rats we are working with cannot see anything at all; the transduction of light is totally demolished," he says. "So anything that can say to the retina, and eventually to the brain, 'Here there is light,' would be a considerable jump." □