Adaptive Optics Ophthalmoscopy

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ABSTRACT

Retinal images in the human eye are normally degraded because we are forced to use the optical system of the human eye—which is fraught with aberrations—as the objective lens. The recent application of adaptive optics technology to measure and compensate for these aberrations has produced retinal images in human eyes with unprecedented resolution. The adaptive optics ophthalmoscope is used to take pictures of photoreceptors and capillaries and to study spectral and angular tuning properties of individual photoreceptors. Application of adaptive optics technology for ophthalmoscopy promises continued progress toward understanding the basic properties of the living human retina and also for clinical applications. [*J Refract Surg* 2000;16:S602-S607]

RETINAL IMAGING

When the first photographs of the living human retina were obtained in 1886 by Jackman and Webster, the camera had to be attached to the patient's head to avoid motion, and exposure times were over 2 minutes in length.[1] Only the largest retinal features, such as the optic disc were visible. Since that time, fundus imaging has evolved and improved with advances in light detection, optics, and optical design.

In spite of these improvements, the inside of the eye is still difficult to see. The retina is relatively transparent and the retinal pigment epithelium absorbs most of the visible light making the fundus a weak reflector. You can only expect about 1 to 100 photons returned for every 1,000,000 photons used to illuminate the eye.[2] Simply increasing retinal illumination for a photograph is not a feasible option since there are limits on how much energy can be safely delivered to the retina.[3] The use of fluorescent dyes injected in the blood stream can increase contrast in the retinal vasculature but, on the whole, the limited retinal reflectance imposes a real and uncorrectable constraint to retinal imaging.

Another limit for retinal imaging is imposed by the optical system itself. Although a good microscope employs a precision high-numerical-aperture objective lens to image its sample, ophthalmoscopes rely on the use of the eye's optical system as that objective lens. Not only does the eye, as an objective lens, limit the numerical aperture but its optics are fraught with aberrations that blur the retinal image. Until the last decade, these aberrations prevented ophthalmoscopes from getting the maximum image quality out of what little numerical aperture that the human pupil offers. The potential benefits of correcting the aberrations to diffraction-limited can be easily quantified in a simple formula, used to calculate the cut-off frequency through a diffraction limited optical system (the maximum spatial frequency that can be imaged by the optical system):

$$f_{cutoff} = d/57.3\lambda \tag{1}$$

where f_{cutoff} is the maximum spatial frequency in cycles per degree, λ is the wavelength of light, and d is the pupil size. When the eye's aberrations have been corrected, the modulation transfer drops off nearly linearly from 100% at 0 cycles per degree to 0% at this cut-off point. For a 3 mm pupil, which is considered to provide optimal optical quality in a typical aberrated human eye[4,5], the cut-off for 550 nm light is 95 cycles/degree By comparison, the spatial frequency of foveal cones is 120 cycles/deg so a 3 mm pupil would not be sufficient for photoreceptor imaging.

Different methods have been used to extract high frequency information from images of the retina[6-9] but none could reproduce a faithful representation

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of the retina on a microscopic scale of any appreciable size or without significant amounts of analysis after image capture. No attempts at imaging small features in the retina were successful until adaptive optics were used to compensate for the aberrations in the eye.

ADAPTIVE OPTICS

The possibilities of adaptive optics were realized long before the technology was available to accomplish it.[10] The original problem was of image degradation arising in ground-based telescopes due to turbulence in the earth's atmosphere. The military appreciated the benefits of this and funded programs during the 1970s and 1980s to develop the technology for imaging foreign satellites with ground-based telescopes.[11] Much of the military's information was declassified in 1992, a move that accelerated progress for all adaptive optics applications. At the present time, adaptive optics technology is still maturing and developing and there is much scope for improvement. Today, virtually every major telescope manufacturer has or is planning to establish an adaptive optics program. For a comprehensive set of links to websites from groups that are posting adaptive optics results, connect to the Center for Adaptive Optics website at www.ucolick.org/~cfao/.

ADAPTIVE OPTICS FOR THE HUMAN EYE

The history of adaptive optics for ophthalmic imaging is just over 10 years old. Adaptive optics was first used in a scanning laser ophthalmoscope by Dreher and colleagues.[12,13] The wavefront corrector was a 13-element segmented mirror and the correction was limited to only the astigmatism and defocus of the eye. The images they obtained were moderately better than without correction—not a surprising result given the low-order of the correction. Nonetheless, they are credited with the

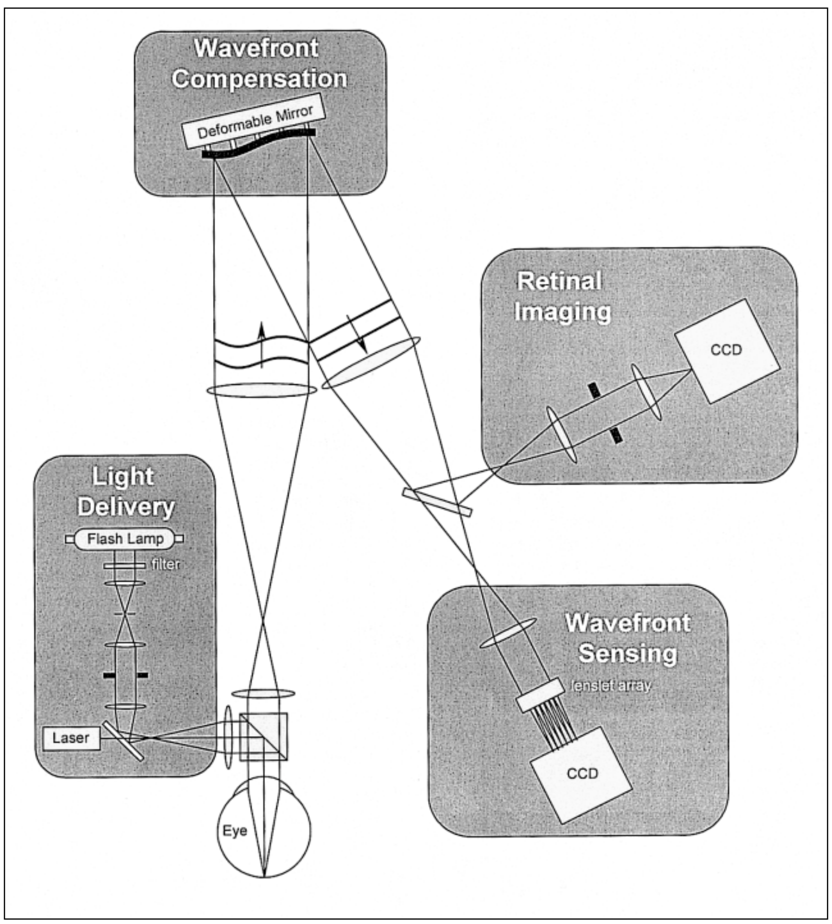

Figure 1. Optical system for wavefront sensing and correction. The eye focuses a collimated beam from a laser onto the retina. The light reflected from the retina forms an aberrated wavefront at the pupil which is measured by the Hartmann-Shack wavefront sensor. A deformable mirror, conjugate with the pupil, is used to compensate for the eye's wave aberration. After compensation is achieved, the retina is imaged by sliding a mirror into place to open the retinal imaging arm. For imaging the retina, a krypton flash lamp delivered a 4-ms flash, illuminating a 1 degree diameter retinal patch.

concept and the potential optical benefits of correcting the high-order aberrations were well described. It wasn't until 1996 that adaptive optics were successfully applied to high resolution imaging in the human eye.[14]

Methods

A typical adaptive optics system for the human eye is shown in Figure 1.[14] The adaptive optics part of the imaging system comprises two main components, the wavefront sensor and the wavefront corrector. The wavefront sensor is used to measure the aberrations of the optical system that are to be corrected. Different wavefront sensing techniques might be used, but the technique that is integrated into the Rochester adaptive optics ophthalmoscope is the Hartmann-Shack wavefront sensor.[14,15] The wavefront corrector in the Rochester ophthalmoscope is a deformable mirror (Xinetics, Inc.), which is a 2-mm-thick reflecting mirror mounted at the back onto a grid of 37 piezoelectric actuators that can push or pull the mirror over a 12-bit range spanning ±2 µm, thereby allowing for wavefront compensation of up to 8 µm. Alternative techniques to compensate for wavefront aberrations are being explored but as yet, none have been demonstrated successfully. Nonetheless, an alternative choice is attractive given the high cost and large size of the deformable mirror system.

In the adaptive optics ophthalmoscope, the wavefront sensor and the deformable mirror are integrated into the same instrument and are carefully aligned. The patient sits at the instrument and their eye is aligned with the instrument axis. With the deformable mirror in an initially flat state, the first aberration measurement of the eye is made. The wavefront sensor reconstructs the wavefront aberration and prescribes the appropriate voltages on the pistons of the deformable mirror to compensate for the aberration. The wavefront is corrected to 40% of the total wavefront aberration per cycle. Each subsequent wavefront measurement measures the eye plus the partial correction by the mirror and repeated iterations are used to converge toward a final and complete wavefront correction. Since there are a limited number of actuators in the deformable mirror and the eye has a small fraction of very high order aberrations, a perfect correction is not possible. In practice, one continues the iteration until the reductions in wavefront correction asymptote to a minimum value. Once the wavefront is flattened, a mirror is placed in the path to redirect light from the eye to the retinal camera, which looks at the retina through the compensating mirror. For imaging, the retina is illuminated with light from a krypton flash lamp. Narrow-bandwidth interference filters are placed in the illumination path to control the wavelength of light that is used for imaging.

Adaptive Optics Imaging Results

Photoreceptor Images—Using the system developed at the University of Rochester, we obtained the best pictures ever of the cone mosaic in the living human eye.[14] Figure 2 shows three images of the same retinal location (1 degree from the center of the fovea) in a living human eye. The image on the left is a single snapshot taken without any compensation, but corrected for defocus and astigmatism. The middle image was taken with adaptive compensation. After aberration compensation the retina is better resolved and has higher contrast. Each bright spot is a single cone photoreceptor, which at this retinal location are about 5 µm in diameter. Nearly all photoreceptors are resolved in a single image. Registration of multiple images, as shown on the rightmost image, improves the signal:noise in the image to a point where virtually all photoreceptors are resolved.

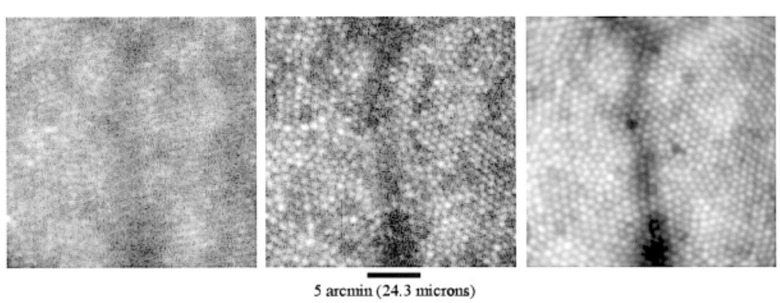

Figure 2. Images before and after adaptive compensation for the right eye of a living human subject. All three images are of the same retinal area located 1 degree from the central fovea. Images were taken with 550-nm light (25 nm bandwidth) through a 6 mm pupil. The dark vertical band down the center of each image is an out-of-focus shadow of a blood vessel. The leftmost image shows a single snapshot taken after defocus and astigmatism have been corrected. The middle image is a snapshot after additional aberrations have been corrected with adaptive optics. The rightmost image shows the benefits in image quality obtained by registering and averaging multiple frames, 61 in this example.

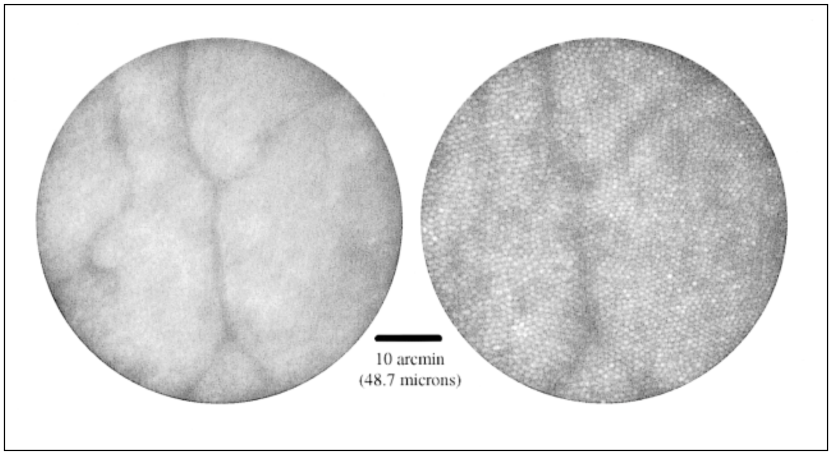

Figure 3. 1 degree images of same retinal location at two different focal planes in the right eye of a living human subject. In the left image, capillaries as small as 6 µm are resolved. By focusing deeper into the retina, the underlying photoreceptors are resolved and the capillaries appear as faint shadows. These images are from a location 1 degree from the central fovea, which is located to the right in the direction of decreasing photoreceptor diameter.

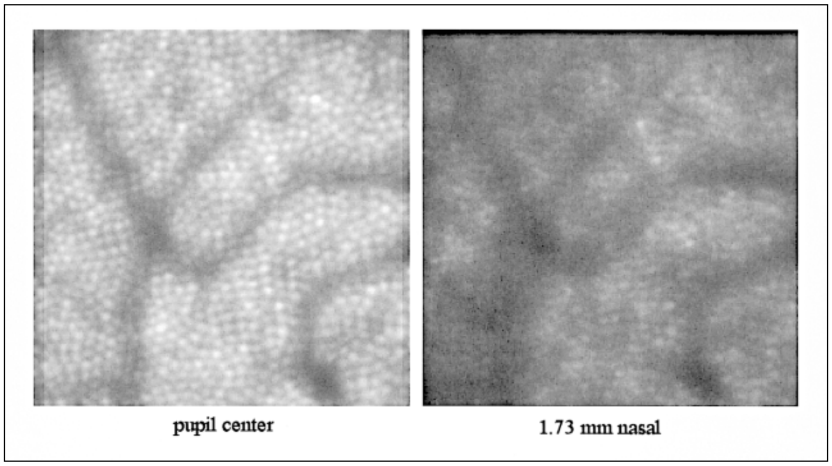

Figure 4. Photoreceptor images taken with two different illumination angles. Both images were taken through a 6-mm pupil with the same energy of 550-nm light delivered through a 2-mm diameter entrance pupil. For the left image, the 2-mm pupil was located so that the photoreceptors were illuminated along the axis of the photoreceptors. In the right image, the same photoreceptors were illuminated with the entrance pupil displaced 1.73 mm in the nasal direction. Both intensity and contrast is reduced in the image taken with the offset illumination angle.

Capillary Images—The retina is a thick multilayered structure. In Figure 2, the dark vertical band down the center of the image was the shadow of a capillary. Figure 3 shows that by adjusting the focus to a higher plane the capillaries can be resolved.[16] In this image, the capillaries are resolved as distinct shadows that are back-illuminated by the underlying photoreceptor layer. This occurs because at this retinal location and with a wavelength of 550 nm, the photoreceptors are the source of most of the scattered light from the retinal image. The capillaries in Figure 3 delineate the edge of the avascular zone. From these pictures, one can not only detect the presence of a capillary but can accurately measure its diameter. We measured capillaries as small as 6 µm in diameter.

Angular Tuning—Cone photoreceptors act as fiber optic waveguides, a property that gives rise to the well known Stiles-Crawford effect. The entry point in the pupil that has the maximum luminous sensitivity is called the Stiles-Crawford peak and this peak typically lies slightly nasal from the pupil center.[17] We can measure the directional properties by determining how light is coupled into the cones as a function of illumination angle. If less light gets into a cone, the reflected light from that cone will also be less. We tested this hypothesis by taking images of the same cone mosaic under identical conditions except that we varied the illumination angle. The illumination angle was controlled by adjusting the location of a 2 mm diameter illumination beam to different locations in the pupil. In all cases we took images though a 6 mm exit pupil, which was necessary to obtain sharp retinal images. The pair of pictures shown in Figure 4 show striking differences in the amount of reflected light with a less than 2 mm shift in the illumination beam location in the pupil. By resolving individual photoreceptors, we have the first opportunity to measure the tuning properties of individual cones.

Arrangement of S, M, and L Cones—Although it was proposed over 200 years ago that the human retina was comprised of three cone types, it wasn't possible to determine the spatial arrangement of

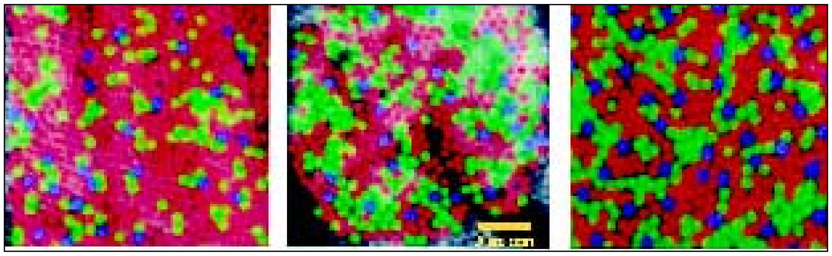

Figure 5. Pseudocolor images of the trichromatic cone mosaic in two human eyes (left, center) and (right) a macaque monkey. Blue, green and red colors represent the short (S), middle (M), and long (L) wavelength sensitive cones respectively. The two human subjects have a more than threefold difference in the number of L vs. M cones.[19] In all cases, the arrangement of the M and L cones is essentially random.

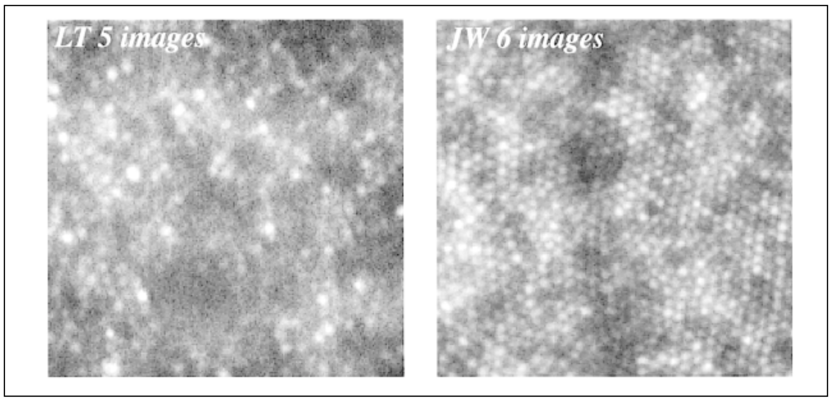

Figure 6. The left slide shows a sum of 5 images of a patient with retinal dystrophy. The photoreceptor array is patchy compared with a set of 6 images from a healthy eye.

those cones prior to the development of high-resolution imaging with adaptive optics. Retinal densitometry has been used for years to measure the pigment concentration in cone photoreceptors[18] but high resolution imaging makes it possible to perform densitometry on individual photoreceptors and identify their subtype. Here we present the results of such an experiment that was performed on the eyes of two humans and one macaque. Full details of this experiment are described elsewhere.[19] A pseudocolor image of the arrangement of the cones is shown on Figure 5.

Patient with Cone Dystrophy—Retinal imaging was performed on a patient in a collaborative effort with Dr. George Bresnick (then Chair of Ophthalmology Department, Strong Memorial Hospital, Univ. of Rochester, Rochester, NY). The patient had bilateral central field loss that was determined, through multi-focal ERG measurements, to be retinal in origin, although little was known about the cause or the particular nature of the disorder. Vision was 20/200 in both eyes over the central 5 degrees. Images obtained in a single imaging session exhibited an irregular patchiness of the cone mosaic in the affected region. By comparison, no similar traits were observed in any of the normal subjects for which we obtained images of comparable resolution. A comparison between the cone dystrophy patient and a normal subject is shown on Figure 6.

THE FUTURE OF ADAPTIVE OPTICS IMAGING

Basic Science Applications

High-resolution ophthalmoscopy with adaptive optics has already been used to produce the first images of the S, M, and L cone mosaics in living human eyes. This opens many possibilities, including the study of a large population of eyes, studying eyes with various color deficiencies, studying the development of the S, M, and L cone mosaic, or measuring the changes in S, M, and L cones toward the periphery. The complementary study of the color vision in the same eyes also opens interesting possibilities.[20] Other properties of the cone photoreceptor mosaic that are currently being studied are the angular tuning of individual cones and the packing arrangement of the cones in the mosaic.

Clinical Applications

Early detection and appropriate treatment of retinal disease is known to be the best way to maintain good vision in the population. It follows that one of the most important future directions for adaptive optics imaging will be in the field of clinical science. The microscopic view of the retina obtained using adaptive optics offers an unprecedented sensitivity to the microscopic changes that occur in the retina during the earliest stages of a retinal disorder. In addition, the improved ability to image will open the possibility of testing the effectiveness of treatment interventions and learning more about the mechanisms of the retinal disease.

Alternate Technology: Reduced Size and Cost

Developing and expanding the scope of the use of adaptive optics for basic and clinical investigations will require that adaptive optics technology become simpler to use, more compact, and less expensive. The deformable mirror is the most effective technology for adaptive optics but these devices are large and expensive and a cheaper, smaller version of deformable mirror technology is not likely. However, technologies such as micro-electro-machined (MEMs) micro-mirror devices, liquid crystal spatial light modulators, and other types of deformable mirrors like membrane and bimorph mirrors[21-23] are possible alternatives and vision scientists are currently cooperating with industry to develop such devices.

Beyond Imaging

Direct imaging is not the only use for adaptive optics. Since light can be imaged with high resolution, it follows that light can be delivered to the retina with the same precision. This opens a number of possibilities ranging from studying the perception of aberration-free retinal images to realizing the potential for pinpoint laser treatment of the retina.

Using adaptive optics, aberration-free images can be projected on the retina. These methods are already being used to test the potential benefits of aberration-reducing refractive surgical techniques.[24] Although there are some obvious benefits to vision, such as an improvement in contrast sensitivity, it remains to be seen whether or not hyperacuity tasks might be compromised. Delivering small spots of color to the single photoreceptor cells might also be used to learn about the early stages of color processing the human retina.[14]

For clinical applications, laser systems can be equipped with adaptive optics to pinpoint the treatment to features as small as individual capillaries. Using this technology, laser treatments can be more localized to the damaged areas of the retina and spare the healthy areas.

Adaptive optics in ophthalmoscopy is still a young field. Both the technology and the ideas of how to apply the technology are still developing. With new technologies on the horizon and a host of new scientists and companies developing their own adaptive optics programs, the future promises to be exciting and productive for years to come.

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