Correcting for miniature eye movements in high resolution scanning laser ophthalmoscopy.

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ABSTRACT
The newest generation of confocal scanning laser ophthalmoscopes with adaptive optics correction of ocular aberrations provides retinal images of unprecedented resolution, allowing for real-time imaging of photoreceptors in the living human eye. Natural fixational eye movements made by the subject/patient during recording produce distortions that are unique in each frame. Correction for these distortions is necessary before multiple frames can be added together to achieve noise reduction or to build a mosaic image from different retinal areas. Here we describe the characteristics of fixational eye movements and the distortions they produce during retinal imaging, we show examples of images with particular distortions, and show eye movement records obtained during the correction of these distortions.

Keywords: Retinal Imaging; Scanning Laser Ophthalmoscope; Eye movements

1. INTRODUCTION
The human eye is constantly in motion, even during attempts to fixate steadily. These motions include rapid jerks or saccades, slower drifts, and high frequency tremor, and are collectively referred to as “physiological nystagmus” or fixational eye movements. Fixational eye movements typically produce gaze instability of 10 or 15 arc minutes during sustained periods of attempted steady gaze. These movements are too small to be seen with the naked eye or with mid-level eye movement monitors, but they are a significant source of image artifact when imaging the retina with high resolution systems such as Optical Coherence Tomography or Adaptive Optics Scanning Laser Ophthalmoscopy (AOSLO)\(^2\). Here we discuss the characteristics of these image artifacts as they relate to the AOSLO and we describe a method for recovering eye motions from the distortions they cause. This recovery is the first step in the process of removing the distortions, and it illustrates the power of retinal imaging as a method for studying the eye movements themselves.

Historically, the most precise measurements of fixational eye movements relied on the “Optical Lever” technique, in which a plane mirror was affixed to the eye and a beam of light was reflected from this mirror to a recording system. Rotations of the eye produced deflections of the beam, and these were typically recorded on a moving photographic film or plate. With sufficient distance between the eye and the recording system, beam deflections caused by translations of the eye become insignificant relative to those caused by rotations. Adler and Fliegelman (1934)\(^3\) successfully recorded fixation movements with a mirror stuck directly on the sclera, while later investigators (notably Riggs and colleagues\(^4,5\)) developed a tight fitting contact lens mount for the mirror. The rotations thereby recorded revealed an eye in constant motion. Typically, a fixating subject’s gaze would exhibit slow, apparently random drifts, interrupted periodically by rapid micro-saccadic movements with amplitudes in the range of 10 arc minutes. Additionally, a tremor component was found with an amplitude of 5 to 15 arc seconds and a frequency of 40 to 100 Hz\(^1,6\). Binocular recordings showed that the micro-saccades were simultaneous and nearly identical in the two eyes, but that drifts and tremor were apparently independently generated\(^7\). Subsequent studies have shown that the micro-saccadic component of fixation can be suppressed by some subjects\(^8\).

These early studies concentrated on horizontal eye rotations, but in fact the eye rotates in three dimensions when fixating and the amplitude of rotations are not equal for different rotation axes. (The coordinate systems used to describe rotational kinematics vary from one research group to another: one can consider the instantaneous rotational velocity of the eye either in terms of a single axis of rotation whose orientation changes over time, or in terms of three component rotations around cardinal axes such as Horizontal, Vertical and Torsional. Here we take the latter approach.) Using magnetic scleral search coils, van Rijn et al.\(^8\) found that torsional eye rotations were considerably larger than horizontal or vertical axis rotations in all four of their fixating subjects. A more recent study of ten subjects using a video technique found that the stability of gaze during fixation of a target was comparable on the three axes\(^8\). It is clear,
however, that accurate registration of images must include correction for torsion as well as horizontal and vertical rotations.

These parameters of fixational eye movement from the literature represent a best-case scenario for retinal imaging. The subjects were typically highly practiced observers with extensive knowledge of eye movement behavior, and one may assume they exercised great effort to maintain steady gaze. In clinical retinal imaging, one may encounter patients who exhibit varying amplitudes of nystagmus, square wave jerks, frequent eye blinks, or otherwise unstable gaze. In our own measurements of fixation stability we have found a surprising number of normal subjects with subclinical jerk nystagmus, typically upbeating.

1.2. Ophthalmoscopic eye trackers

The eye tracking systems discussed above are based on measurements of globe rotation, generally by attachment of an apparatus to the sclera. Non-contact, optical methods such as the dual-Purkinje image (dPi) eye tracker can also provide very precise measures of eye rotation, but neither optical lever nor dPi methods are perfectly suited to correction of retinal imaging distortions because the retinal image motion is not perfectly correlated with globe motion. For example, the forces applied during saccadic eye movements produce shifts in the optical elements of the eye (“lens wobble”), causing deflections of the retinal image that are not accurately represented by measurements of globe rotation. The optimal eye movement measure to apply must be based on ophthalmic imaging, so that any changes to the optics of the eye will effect both the eye tracking system and the retinal imaging system identically. This might be an analysis of the imaging data itself, or a separate, stand-alone system that tracks features of the retina during the imaging session.

The earliest implementation of an ophthalmoscopic tracking system was by Cornsweet, who reported a resolution of ten arc seconds in a system that tracked a blood vessel using an oscilloscope and photomultiplier tube. Although Cornsweet himself went on to develop the dPi system as a practical method for eye tracking, others have continued to build on the basic idea of tracking a single fundus feature in real time, in order to stabilize ophthalmic imaging against eye movement.

The development of the Scanning Laser Ophthalmoscope (SLO) led a number of research groups to extract eye motion from the recorded retinal images for various applications. Early approaches generally involved offline, manual marking of image features, such as blood vessels, in order to extract eye motion. A method of capturing horizontal movements at high temporal resolution was described by Stetter et al. and an elaborated method for capturing both horizontal and vertical movements was more recently described by Mulligan.

![Figure 1: Illustration of the distortions produced in scanned images by a) horizontal, b) vertical, and c) torsional rotations of the target during a single scan.](image-url)

1.3. AOSLO

The imaging distortions associated with eye movements have become more significant as the resolution of imaging systems has improved and the magnification has increased. SLO resolution has increased dramatically with the addition of adaptive optics technology to correct for the aberrations of the eye over the full diameter of a dilated pupil. These systems allow for non-invasive imaging of individual photoreceptors in the living eye. However, because of the low illumination levels used (to maintain safe retinal exposures) the signal in each frame is quite low. S:N in a still image is normally improved by registering and adding multiple frames. But in an SLO, the images are significantly distorted by fixational eye movements because the image is captured over time from a scanning focused beam. therefore, registration
is only possible after the image distortions due to eye movements are removed. Registration of successive frames taken with a moving eye can also allow for the assembly of a larger retinal image mosaic.

In its current form, the resolution of the AOSLO is about 0.5 arc minutes and can be sampled as high as 1/8 of an arc minute. With subpixel registration, it is possible to extract horizontal and vertical eye motion with subpixel resolution, or within a fraction of an arc minute. Resolution on the torsional axis (defined here to be the imaging axis) will be substantially lower. The temporal resolution is essentially the line scan rate of about 16 kHz. The AOSLO therefore has the potential to provide extremely fine measurements of fixational eye movements, rivaling any system reported to date. This presupposes, of course, that the image motion can be recovered accurately from the video.

1.4. Distortions produced by fixation movements in progressively scanned ophthalmic images.

In order to clarify the problem, we have generated test images to illustrate the distortions produced by horizontal, vertical, and torsional rotations of the eye during a scan. Figure 1 is an image of a standard piece of graph paper, scanned on a flat bed scanner, with motion applied to the graph paper during the scan. The velocity of motion was scaled to the scan rate to produce a reasonable simulation of how saccadic eye movement distorts images collected at the 60Hz frame rate of the AOSLO. Horizontal motions, parallel to the fast scan direction, produce a shearing of the image. Vertical motions, parallel to the slow scan direction, produce either compression or expansion of the image. Torsional rotations, around the imaging axis itself, produce a complex distortion involving both compression and expansion in different parts of the image. The simulation of torsion here (about 45 degrees) is exaggerated considerably to clarify the effect, but torsional saccades of 5 or 10 degrees may occasionally occur even during steady fixation.

Most previous work on the recovery of eye movements from SLO images has made use of particular features in the image, such as blood vessel bifurcations, to disambiguate the motion. The small field size of the AOSLO is such that many images of interest lack such large, distinct features. The approach taken here, and by others working on the problem, is to use image analysis techniques to recover motion over the whole frame. Additionally, we have recorded retinal images with the AOSLO system while at the same time recording eye rotation with a dual-Purkinje image tracker, so that the fixational eye movements recorded with the two techniques can be compared.

![Figure 2](image.png)

Figure 2. Successive frames from an AOSLO video recording of the foveal region of a human retina. The field size was one degree square. The image on the left is typical of most frames collected, with relatively little distortion due to eye movement. The frame on the right has a pronounced shear distortion due to a rapid horizontal rotation of the eye.

2. Methods

2.1. AOSLO imaging.
Subjects were healthy adult males from the research group with many years of experience as experimental subjects. All procedures were approved by the University of Houston Institutional Review Board and subjects gave informed consent prior to measurements. Subjects were dilated with 2.5% Phenylephrine and 1% Tropicamide before imaging. The head was stabilized by a bite board. The scanning field was set to either 1.5 or 2.0 degrees square in different recording sessions, and imaging was centered on the fovea. Aberrations of the eye were corrected over a 5.9 mm pupil, using data from a Shack-Hartman wavefront sensor built into the AOSLO system. The scanning beam produced a near diffraction-limited focused spot with a diameter of approximately 2.5 microns at the retina, somewhat larger than the smallest photoreceptors, but sufficient to image individual parafoveal receptors and to produce high contrast image data from all parts of the retina. The scanning rate was 30 Hz vertical and 16kHz horizontal. The horizontal scan was produced with a resonant mirror, resulting in a sinusoidal waveform. Video was captured during the relatively linear portion of the scan in one direction, and the remaining distortion due to the velocity variation over the scan was corrected for in software. Video sequences were digitized and stored to disk for off line analysis.

Figure 3. Schematic of the Adaptive Optics Scanning Laser Ophthalmoscope, modified with an additional relay telescope to accommodate the dual-Purkinje image eye tracker. The dPi tracker uses infrared light to track rotations of the eye on horizontal and vertical axes. The dichroic beam splitter passes visible light and reflects infrared, allowing both systems to operate without interference. PMT photomultiplier tube.

2.2. Dual-Purkinje image eye tracking

In order to track the eye that was being imaged with the AOSLO, a dual-Purkinje image eye tracker was positioned in front of the AOSLO system and a relay telescope was added in order to accommodate the physical constraints imposed by the eye tracker (Fig. 2). Horizontal and vertical eye position was sampled at 120 Hz with a resolution of one arc minute, the nominal resolution of this eye tracking system.

2.3. Video analysis

Fixational eye movements were extracted from the raw AOSLO videos using a cross-correlation procedure. All frames in the video were compared to a single reference frame in order to determine how the eye had moved during the course of the imaging. Because the eye moves constantly during retinal imaging, no frame ever matches the reference frame.
exactly, and the reference frame itself is distorted by eye motion. Video frames were therefore subdivided into strips, and each strip was independently matched to the corresponding region of the reference frame. After all frames in a video sequence had been analyzed, the distortion in the reference frame was removed by taking the average relative shift found for all other frames, on the assumption that the actual average shift is zero for a fixating eye. Because the fast scan direction is horizontal, a single scan line represents a snapshot of the retina and is virtually unaffected by most fixational eye motion. (A single horizontal scan captures its data in 25 microseconds, and even at the peak velocity of a microsaccade, the shift in this time is less than one pixel.) A strip of just a few lines is effectively a snapshot as well, unless a saccade occurs at that time. We typically use strips of 4 scan lines for our cross-correlation region, representing a compromise between enough image data to match reliably and having good temporal resolution. In the extractions reported here, strips from four equally spaced locations down each frame are analyzed, providing a sampling frequency of approximately 120 Hz (30 Hz video X 4 samples per frame). Temporal resolution up to 16 kHz is achievable if all scan lines in a frame are analyzed. The left and right halves of each frame are analyzed independently, so that torsion can be extracted from differences in the vertical shift of each half. With a pixel size of 16 arc seconds and a field size of 2 degrees, the center to center distance of our left and right half strips is 1 degree. A torsional change that produced a one pixel vertical shift difference in these two halves would have a magnitude of about 15 arc minutes. Subpixel matching techniques can improve the torsion axis resolution, but torsional rotation measures will always be considerably lower resolution than horizontal or vertical rotations. We focus here on the horizontal and vertical axis, for which we also have dual-Purkinje image tracker data. All the analysis was conducted using Matlab programs developed in house. Raw AOSLO videos in the form of AVI movies were first corrected for the scan nonlinearity, then each strip segment of each frame was cross-correlated to a larger, corresponding region in the reference frame. By default, the first frame of the movie was used as reference, but an alternate was chosen when the first frame had a large saccade or other obvious distortion.

3. Results

Eye movements extracted from one AOSLO video are plotted in Fig. 3, along with traces from the dPi tracker measured simultaneously. The video and eye traces were recorded by two separate computers with independent timing and calibrations, so the traces have been offset vertically and horizontally to provide the best match by eye. Flat portions of the records at about 2 seconds and 6.5 seconds on the time axis are missing data due to eye blinks. Overall, the agreement is as good as one can expect, given the nominal one arc minute sensitivity of the dPi tracker. Some discrepancies between the two traces are worth noting. For about one second after each eye blink, the discrepancy between the traces is larger than at other times. This may be due to some settling in the dPi tracker, which loses lock during eye closure. It may alternatively be due to torsion changes related to the eye blink. Estimates of torsion from the AOSLO data indicate a 0.5 degree change from before to after the first blink, and the difference in measurement axis between the AOSLO (visual axis) and dPi tracker (optic axis) would produce a discrepancy of about 4 arc minutes in the vertical traces for that much torsion.

Saccades are accompanied by overshoots in the dPi records, and smaller overshoots occur in the AOSLO derived traces at the same time. Note the vertical saccade at about 3 seconds in panel (d), and the corresponding spike in the horizontal record at the same time in panel (c). These artifacts are likely due to wobble of the intraocular lens, as has been reported previously for larger saccades. As predicted, the retinal image also shows wobble, but of smaller amplitude. For brief periods, on the order of one second, the dual-Purkinje image tracker predicts the motion of the retinal image to within about one arc minute, but over the course of a longer recording such as these the discrepancy can reach several minutes.

4. Discussion.

The level of agreement between these two methods serves to validate both as techniques for measuring drifts and microsaccades in fixational eye movements. Further development of the software to extract eye motion and correct for the distortions they produce will likely improve the sensitivity of the AOSLO-derived records even further. Conservatively, we can conclude from the comparison here that both systems are accurate to within one to four arc minutes, depending on the duration of recording. However, it seems likely that the discrepancies are due almost entirely to the dPi tracker, and that the actual resolution of the AOSLO records is on the order of one pixel; i.e. just a few arc seconds. Horizontal position estimates for the left and right halves of the AOSLO frames rarely exceed one pixel, demonstrating the precision with which the correlation peaks can be localized.
One goal of this research is to work toward precise compensation for eye motion, so that image distortions can be removed and multiple frames can be averaged together for improved signal to noise ratios and extended scan areas. Given that no practical eye tracker in existence can measure horizontal, vertical and torsional eye position to the precision required for complete correction of fixational movements, it seems likely that the approach taken here of offline image analysis will always be the final step in the correction of retinal images. However, an integrated eye tracker with sensitivity of a few minutes of arc could be extremely valuable in maintaining the scan on a region of interest and in steering the correlation algorithm for much more rapid analysis of the images. The combination of the two techniques could well form the basis for on-the-fly correction of retinal images in a clinical setting.

Figure 4. Horizontal (a) and Vertical (b) eye position traces extracted from 10 seconds of AOSLO video of a steadily fixating eye. Expanded plots of 1 second segments from the horizontal (c) and vertical (d) traces to show detail. The vertical axis shows eye position in arc minutes, plotted against time in seconds on the horizontal axis.

5. Acknowledgments
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6. References


