Adaptive Optics Imaging of Outer Retinal Diseases

Principal Investigator (PI):
Daniel X. Hammer, PhD
Deputy Director

FDA/CDRH/OSEL/DBP
10903 New Hampshire Ave
WO62-1106
Silver Spring, MD 20993
301-796-9320
daniel.hammer@fda.hhs.gov

Co-PI:
Zhuolin Liu, PhD
Staff Fellow

FDA/CDRH/OSEL/DBP
10903 New Hampshire Ave
WO62-1103
Silver Spring, MD 20993
301-796-7914
zhuolin.liu@fda.hhs.gov

Medical Advisor/Co-PI:
Emily Chew, MD
Director of the Div. of Epidemiology and Clinical Applications
National Eye Institute
National Institutes of Health
10 Campus Drive
Bethesda, MD 20810
301-496-6583
echew@nei.nih.gov

Associate Investigators (AI):

Name	*Organization*	*Role*	*Email*
Anant Agrawal, PhD	FDA/CDRH/DBP	Co-Investigator	anant.agrawal@fda.hhs.gov
Achyut Raghavendra, PhD	FDA/CDRH/DBP	Team Member	achyut.raghavendra@fda.hhs.gov
Catherine Cukras, MD, PhD	NIH NEI	Co-Investigator	cukrasc@nei.nih.gov
Kapil Bharti, PhD	NIH NEI	Co-Investigator	kapil.bharti@nih.gov
Tiarnán Keenan, BM BCh, PhD	NIH NEI	Co-Investigator	tiarnan.keenan@nih.gov
Tharindu de Silva, PhD	NIH NEI	Co-Investigator	tharindu.desilva@nih.gov
Johnny Tam, PhD	NIH NEI	Co-Investigator	johnny.tam@nih.gov
Laryssa A. Huryn, MD	NIH NEI	Co-Investigator	laryssa.huryn@nih.gov
Wadih M. Zein, MD	NIH NEI	Co-Investigator	zeinw@nei.nih.gov
Alisa Thavikulwat, MD	NIH NEI	Co-Investigator	alisa.thavikulwat@nih.gov

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Abstract

Objective: The objective of the study is to collect adaptive optics (AO) retinal images from human subjects with outer retinal diseases (diseases of the outer retina including photoreceptor, retinal pigment epithelium (RPE), basement membrane or choroidal pathologies) to develop new diagnostic methods, biomarkers, and clinical endpoints.

Study Population: Up to fifty (50) healthy volunteers without eye disease (Cohort 1) and up to fifty (50) affected participants with any type of outer retinal disease (Cohort 2) will be enrolled.

Design: This is a longitudinal study protocol where participants will be imaged with investigational multimodal AO (mAO) retinal imaging systems that include optical coherence tomography (OCT) and scanning laser ophthalmoscopy (SLO) channels over three years. High resolution OCT and SLO videos will be collected while the instruments automatically detect and correct for image distortion caused by ocular aberrations. In general, videos of different retinal cellular structures will be acquired from several retinal locations using various imaging modes.

Outcome Measures: The primary outcomes for this protocol are development of new diagnostic methods and disease biomarkers, investigation of cellular morphological and functional changes due to various outer retinal diseases, and development of new AO clinical endpoints for novel therapies.

1.0 Introduction

Adaptive optics (AO) retinal imaging is a diagnostic technology that has the capacity to vastly aid detection and treatment of ocular diseases by providing direct visualization, quantification, and functional assessment of retinal cells [1-2]. AO works by sensing and correcting an individual's ocular aberrations, providing cellular level access to the living eye. AO has for many years had the ability to routinely resolve cone photoreceptors. Recent progress includes imaging rod photoreceptors [3], retinal pigment epithelium (RPE) [4-7], choriocapillaris [8], and ganglion cells [9-11]. These advances may transform the diagnosis of ocular diseases, including highly prevalent ones such as age-related macular degeneration (AMD), glaucoma, and diabetic retinopathy.

However, AO has yet to achieve full clinical translation, partially because of system complexity and cost but also because the clinical benefit to patients remains unrealized. The FDA and NIH are uniquely positioned to collaborate in support of technology translation through their public health missions. This protocol supports a collaborative project between FDA/CDRH/OSEL and NIH/NEI to develop models and methods for AO image quality assessment and standardization, discover new AO imaging-based biomarkers of retinal diseases, and develop novel modalities suited to specific cellular targets, function, and disease. For example, we aim to extend the multimodal AO (mAO) scanning modes to capture RPE motility from subjects with normal and diseased eyes [12]. To support these projects and others, we have built two investigational mAO retinal imaging platforms that will be used in a set of human subject experiments [13,14].

The FDA mAO systems are particularly suited to study diseases of the outer retina and retinal degenerations (e.g., AMD, cone-rod dystrophies, hydroxychloroquine toxicity, retinitis pigmentosa, etc.). Common to outer retinal degenerations is the progressive degeneration of photoreceptors either prior to, simultaneously with, or subsequent to RPE dysfunction, depending on the specific retinal disease. The resolution that can be obtained using AO would enable further parsing of the magnitude and location of abnormalities both by retinal layer and spatial distribution. These observations could further inform the sequence of retinal degeneration as well as provide a finer quantitation of the extent of disease allowing for the development of AO-based outcome measures in clinical trials. Also common to retinal degenerations is the slow progression of disease. While changes over many years are observable using current technologies, an instrument that would enable the observation of changes on a smaller scale would have the potential to demonstrate meaningful differences over a shorter time compared to blunter tools which would require larger changes before detection. Having such outcomes would greatly improve the design and feasibility of interventional clinical trials for outer retinal degenerative disease.

In retinal toxicity due to hydroxychloroquine, the retina demonstrates a paracentral loss of photoreceptors which is seen on exam as a "bulls-eye" appearance, on optical coherence tomography (OCT) as a loss of the inner/outer segment (IS/OS) reflectivity, and functionally as paracentral scotomas. The mechanism of retinal toxicity from chloroquine and hydroxychloroquine is not completely understood. Chloroquine has been shown to disrupt lysosomal function in the RPE and lead to accumulation of membranous cytoplasmic bodies in the cone photoreceptors in animal models. Being able to image human retinas exposed to hydroxychloroquine with AO-OCT might further demonstrate the mechanism of action of this widely used medication in the development of toxicity. Further, observing the changes in cases of clinical evidence of toxicity may enable an earlier detection of damage that could be meaningful to this public health problem.

Other than the fact that AO imagers produce a tighter spot on the retina, which can be accounted for in the laser safety analysis, AO works like any other minimally-invasive, commercial, diagnostic retinal imager, of which there are dozens of manufacturers with hundreds of devices imaging millions of subjects daily. AO was invented in the late 1990s and instruments have been used in a research setting for decades, including at the University of Rochester since 1997, the University of Houston since 1998, the University of California, Berkeley since 2005, the University of California, San Francisco since 2010, and the Medical College of Wisconsin since 2011. No light-exposure-related incidents in humans have been reported at these institutions. Moreover, the Medical College of Wisconsin has established an International Adaptive Optics Consortium (IAOC) with the goal of conducting multi-center, large-scale AO clinical trials. IAOC members with similar

systems include the Medical College of Wisconsin, the University of Pennsylvania, the New York Ear and Eye Infirmary, Moorfields Eye Hospital (UK), the University of California, San Diego, and NIH, National Eye Institute (NEI). The IAOC AO instruments have been used to screen over 1,000 patients at four sites with no light-exposure-related incidents. Commercially, Imagine Eyes has several AO instruments that have received the CE mark in Europe and have imaged thousands of patients. In the U.S., Physical Sciences Inc. has also taken steps toward commercialization with various AO imagers. In collaboration with researchers at University of Pittsburgh Eye Center, Children's Hospital Boston, and others, their instruments have imaged hundreds of patients.

The FDA AO devices used in this study are investigational devices that qualify for investigational device exemption (IDE) abbreviated filing requirements according to 21 CFR 812. Appendix 3 contains a summary of the relevant sections of 21 CFR 812 related to abbreviated filing requirements and the justification for the automatic IDE status for the FDA AO devices.

2.0 Study Objectives

The objective of the study is to collect AO retinal images from affected participants with outer retinal diseases and healthy volunteers to develop new diagnostic methods, biomarkers, and clinical endpoints.

3.0 Participants

Fifty (50) healthy volunteers (Cohort 1) and fifty (50) affected participants with outer retinal disease (Cohort 2) will be accrued for this study. Outer retinal diseases include but are not limited to AMD, inherited retinal degenerations such as retinitis pigmentosa and cone-rod dystrophy, macular dystrophies (Stargardt's disease), and choroidal dystrophies (choroideremia). It also includes conditions, systemic diseases, or treatments that may affect the outer retina, for example hydroxychloroquine toxicity.

3.1 Inclusion Criteria

Participants will be eligible if they:

1. Are 21 years of age or older,
2. Have the ability to cooperate with instructions during adaptive optics imaging (similar to instructions given during a clinical eye exam),
3. Have the ability to understand and sign an informed consent. (Non-English speaking participants will not be enrolled into the study), and
4. Have been diagnosed with outer retinal disease or condition (Cohort 2).

3.2 Exclusion Criteria

Participants will not be eligible if they:

1. Have a condition which prevents adequate images from being obtained (e.g. unstable fixation or media opacity),
2. Have visual correction outside of the range +4 diopters (D) to -8 D,
3. Have a history of adverse reaction to mydriatic drops,
4. Have a predisposition to (i.e., narrow iridocorneal angle) or any history of acute angle closure glaucoma (AACG), or
5. Are working under the direct supervision of Drs. Hammer, Chew and Liu, or any of the NIH/NEI AIs.

All participants will be initially screened for the eligibility criteria using existing medical records (for existing participants) or during an initial eye exam (for new participants) conducted at the NEI Eye Clinic, NIH Clinical Center (CC) after initial consent is given.

3.3 Recruitment

All study participants (Cohorts 1 and 2) will be recruited from the NIH patient population through direct individual recruitment (by Dr. Chew or the other NIH/NEI AIs) by email, phone call, or in person. A copy of a sample recruitment email is included in Appendix 9, which also includes the information that will be conveyed in any phone or in-person recruitment. The recruitment period may continue while the study is active.

Participants will not be involved in any aspect of the experimental study (design, conduction, or analysis). The existing ophthalmic record for potential participants from the NIH study population will be screened by Dr. Chew and/or the other NIH/NEI AIs with permission to review medical records, particularly when targeting participants with a specific outer retinal disease.

3.4 Anticipated Benefit

Participants derive no direct benefit from participating in this study. However, research using data from this protocol may yield generalizable knowledge regarding methods for assessing ophthalmic variables in various eye diseases.

3.5 Consent Process

All study participants will be consented by Dr. Chew or the NIH/NEI AIs prior to any activity using a standard script (Appendix 4). Dr. Chew and/or the NIH/NEI AIs will determine from the participant's medical record or during the exam if they meet the inclusion/exclusion criteria listed above. In particular, she will judge their eligibility based upon the clarity of their anterior optics (e.g., no dense cataracts), their fixation ability, and their ability to keep their head motion minimal while sitting in a chin rest.

Potential participants who have been screened initially (see recruitment section above) will receive a verbal explanation of the purposes, procedures, and potential risks of the study. Potential participants will have the opportunity to carefully review the informed consent form (ICF) and ask questions regarding this study prior to signing, and they will be informed that they may withdraw from the study at any time without prejudice to themselves. Potential participants will also be allowed to take the ICF home overnight to carefully consider the risks prior to consent. All participants must sign an IRB-approved ICF. A signed copy of the ICF will be provided to the participant to take home.

The signed ICF will be scanned and stored in the NIH CC Electronic Medical Record (EMR) system. Upon arrival at the FDA, participants will receive a verbal explanation of the study purpose, procedures, and risks from the FDA PI and/or Co-PI. The FDA PI and/or Co-PI will reference the consent the participant signed at the NIH and ask the participant to affirm he/she wants to proceed with the study. These steps will be completed at the beginning of each visit to the FDA. Participants will not be asked to complete a consent form again at the FDA.

3.6 Alternatives to Participation

Participants do not receive any treatment in this study or forego any treatment in order to participate in this study. The alternative, therefore, is not to participate.

3.7 Compensation

Participants will receive a $25 gift card at the end of each FDA visit to compensate for small travel expenses. Participants will be given stamps to park without cost on the NIH campus. FDA parking is free of charge in the visitor's lot. Participants who withdraw from or end the study prior to completion will no longer continue to receive compensation.

3.8 Withdrawal

Participants may withdraw from the study at any time. Participants will not be penalized or lose any benefits to which they otherwise qualify upon withdrawal. The PI and co-PIs may end participation for any participant if he/she feels that continued participation is not fruitful (quality data cannot be obtained) or overly burdensome (excessively arduous, fatigue-generating, or time-consuming) to the participant, based upon the collective clinical and research experience of the study investigators. Participants will be informed if any significant new findings are discovered that may affect their participation in the study. Data collected prior to withdrawal will remain part of the study findings.

3.9 Consent Monitoring
The Emmes Company, LLC (Emmes) has been assigned to conduct monitoring of study participants' informed consent forms. Monitoring visits will be conducted at least annually at the NIH. Emmes staff will not monitor data or safety events and does not have direct access to or interaction with study participants.

4.0 Study Design and Methods
This is a longitudinal study of healthy volunteers and affected participants with outer retinal diseases to develop new diagnostic methods, biomarkers, and clinical endpoints.

4.1 Adaptive Optics Imagers
The AO imagers used in the study are investigational multimodal systems that includes OCT and SLO channels [13,14]. Two similar instruments will be used in the study. The first instrument, called the FDA mAO imager, scans two near-infrared wavelength beams, one centered at ~760 nm for SLO and one centered at ~830 nm for OCT, across the retina to produce images. The total power delivered to the retina for the mAO imager is below ANSI maximum permissible exposure limits, as described in Appendix 2a. The OCT channel in the second instrument uses a Fourier domain mode-locked laser (FDML). Therefore, the second is called the FDA AO-FDML imager, and uses three infrared beams at ~785 nm (SLO), ~850 nm (AO beacon), and ~1060 nm (OCT). The AO-FDML imager also includes a visible light stimulus channel for functional imaging experiments. The total power delivered to the retina for the AO-FDML imager is below ANSI maximum permissible exposure limits, as described Appendix 2b. The primary upgrade in the second instrument is the OCT imaging speed and pixel density, which has increased from 2.3 volumes (300×300 pixels) per second to 13.4 volumes (500×500 pixels) per second. In both instruments, ocular aberration sensing and correction is achieved with a Hartmann-Shack wavefront sensor (WS) and deformable mirror (ALPAO, DM97-08, France), respectively. Both instruments use a pupil camera and a fixation target to position the participant's head and eye for scanning a particular retinal region within 15 degrees from the fovea. Data collected generally include OCT and/or SLO videos; and less frequently WS videos (temporal sequences) and data on ocular aberrations. All patient procedures (alignment, fixation, imaging scans) and data formats used for the two FDA AO imagers are identical.

4.2 Initial Eye Exam for Screening and Risk Mitigation
Prior to AO imaging at FDA, each participant will undergo a comprehensive eye exam at the NIH CC to determine if they are eligible for continued participation in the study. FDA and NIH clinical study participants never incur any costs for care related to their participation in a study. NIH and FDA will enter into a reliance agreement for NIH to rely on the FDA's IRB for review and approval of this study. Research activities at the NIH will not commence until a Reliance Agreement, also known as a signed IRB Authorization Agreement (IAA) between the NIH and FDA, is in place and all related institutional requirements have been met.

The participants will be examined by the project co-PI/medical advisor, Dr. Chew, or another NIH/NEI AI who is a board-certified ophthalmologist. The participants will undergo a complete eye examination. This will include the measurement of baseline visual acuity (VA) in both eyes with a standard Snellen chart as well as slit lamp examination, intraocular pressure check, and anterior exam to check for any predisposition to a dilation-induced AACG event (narrow iridocorneal angle). Instillation of an ocular anaesthetic (Proparacaine 0.5%) and topical fluorescein will be required to check intraocular pressure and perform gonioscopy as per routine office protocol for the eye examination. If there is no risk for angle closure, the participant's eyes will be dilated with a mydriatic drug (Tropicamide 1%), and any possible adverse event monitored by Dr. Chew or another NIH/NEI AI for 30 minutes. A dilated examination will also be completed at this time. The following testing will be performed on each participant using legally-marketed devices:

1. Color fundus photography,
2. Autofluorescence imaging,
3. OCT retinal imaging,
4. Biometry (eye length),
5. Visual field perimetry (as needed depending on disease process and analysis).

All five tests are standard-of-care procedures for participants with retinal disease or those suspected of having retinal disease. Because the tests are standard-of-care procedures and FDA-approved or cleared devices are used, they pose no greater risk to the participant than a standard clinical eye exam. Such tests are performed on a routine basis in eye clinics across the world on both healthy and diseased eyes. These tests are used for both screening for pathology as well as to follow participants with existing, known pathology. As they are used for screening, particularly in optometric offices, the tests involve risks that the participant may normally encounter in their medical care.

The tests are necessary to collect additional information on each participant's eyes that will be used to guide further AO imaging. Color fundus photography, autofluorescence, and OCT imaging will help to determine where to perform small-field AO imaging. Biometry is necessary to properly correct the AO imaging field-of-view (important for quantitative measurement of retinal cell and structure dimensions). Perimetry provides a functional visual field test and allows location of spatial regions and severity of disease with which to compare the imaging results. All tests will be performed under the supervision of Dr. Chew and/or the NIH/NEI AIs at the NIH CC. The information they provide will be extremely valuable to increase the probability of success for the research study.

For Cohort 1, should any ocular abnormality or disease be detected on eye examination, the participant will be advised to follow up at an appropriate interval (as determined by Dr. Chew or and the NIH/NEI AIs) with an eye care professional for monitoring or treatment. If there are no adverse events or other exclusions, as determined by Dr. Chew, the participant will be cleared to enter the study. Participant information from the eye exam will be kept at the NIH eye clinic per routine office protocol and with strict confidentiality abiding by NIH policies and all relevant regulations regarding medical records. Participant records for the initial eye exam will be kept together with records from other NIH-sponsored clinical studies. Participant information from the eye exam will be transferred to the FDA investigators only with the participant's consent. Written consent will be obtained with a description of the exact information transferred to the FDA investigators when it is necessary to use information obtained from the clinical exam.

The initial eye exam will mitigate the risk of an adverse event from mydriasis to every extent possible. For emergency situations at the FDA site, the FDA investigators will call 911 and if necessary (i.e., an ambulance is not deployed), take the participant to the nearest ER (Adventist HealthCare White Oak Medical Center, 11890 Healing Way, Silver Spring MD 20904). Dr. Chew and the NIH/NEI AIs will also be available for non-urgent questions from study participants and follow-up. For emergency situations at the NIH CC, the NIH investigators will call the appropriate emergency code response team. It is highly unlikely that a study participant will have an adverse event on subsequent imaging sessions if they have not had one during the initial eye exam. However, should a participant experience eye pain after administration of dilating drops (other than the minor stinging that typically accompanies the eye drops), the participant will be immediately brought by the FDA PI to NIH for immediate monitoring by the NIH Co-PI or AIs. Also, should a participant complain of any change in vision, they will be brought by the FDA PI to NIH and have their eye examined and their VA re-tested. Either circumstance will constitute an adverse event.

Drs. Hammer and Liu will administer dilating eye drops during the FDA imaging sessions. Both Drs. Hammer and Liu have previously conducted human subject studies on adaptive optics and have administered dilating eye drops to participants. It is estimated that Dr. Hammer has imaged over 100 participants and Dr. Liu has imaged over 75 participants with adaptive optics retinal imaging devices. It is common in eye clinics for technicians (i.e., non-MDs) to deliver dilating eye drops to participants.

4.3 AO Imaging Procedures

The exposure time limits and restrictions outlined in Appendix 2a and 2b will be followed in order to ensure that there is minimal risk of retinal damage. In particular, the output power level at each wavelength will be recorded with a calibrated power meter (Newport Power Meter Model 1918-C with 918D-ST-IR head) for every imaging session day prior to participant imaging.

All participants will be asked to attend an imaging session at the FDA. Participants may be asked to be re-imaged multiple times to characterize test-to-test variability or other measures related to system performance or physiological changes over time. Additionally, participants may be asked to complete more than one imaging session due to safety reasons, inadequate/incomplete data acquisition during the previous imaging session, and/or if low-quality images were captured during the previous session. The maximum number of imaging sessions for any participant will be five per year and the time between imaging sessions will be no less than one week. For longitudinal experiments, the participants will be imaged over three years. Either or both eyes may be imaged in a given session. Each imaging session will take ~two hours and no participants will undergo more than one imaging session a day. The exact duration will depend upon the participant themselves (ability to hold fixation, number of rest stops, etc.) and on attributes of the experimental design (the scanning mode, number of retinal locations imaged, number of eyes imaged, etc.), and will be known (approximately) and communicated to the participant prior to any given session. Participants will consent to participation as described above. They will be informed of the approximate session duration and that they may ask for a rest period at any time. They will be offered rests frequently during testing and between test blocks, and a rest will be required any time a participant shows sign of fatigue.

Each participant's pupils will be dilated with Tropicamide 1%. One drop will be delivered to the eye(s) at least 20 minutes prior to imaging by the Dr. Hammer or Dr. Liu. Two additional drops of Tropicamide may be administered for insufficient dilation. Adverse reaction to the mydriatic agent will be monitored for 30 minutes following administration by the PI or co-PI. Participants will be instructed to blink whenever the need arises; we do not require participants to hold their eyes open during imaging. Participants will also be provided single-use, preservative-free artificial tears as needed, either self-administered or administered by the FDA investigators.

After pupillary dilation, the following procedures will be performed:

1. Alignment: The participant will be instructed to sit at the instrument and place their chin in the chin-rest while their head is positioned to align their eye to the imaging beams.
2. Closed-loop AO operation (aberration measurement and correction): The participant will be instructed to look at a specific fixation target. The instrument will measure the wave-front aberrations of the participant's eye and the AO control system will correct ocular aberrations to allow acquisition of high-resolution images.
3. Retinal image acquisition: The participant will be instructed to look at a specific fixation target. Image sequences, with various durations depending on scanning mode, will be acquired at different retinal locations. The participant will be allowed to take periodic breaks during imaging as needed.

A subset of subjects will undergo photoreceptor stimulation experiments concurrent with AO-OCT imaging. This involves delivery of a sequence (1-10) of short duration (5-100 ms) pulses of visible light stimulation to the eye. The visible light pulse duration is precisely controlled with a shutter and synchronized to the image acquisition. Sets of AO-OCT volumes will be collected as usual. The visible light stimulus is included in the light safety analyses (Appendix 2a and 2b) and introduces no additional risk of retinal injury.

4.4 Location

The AO imagers are housed in the FDA/CDRH/OSEL laboratory space on the White Oak Campus, Building 62, Room G238.

4.5 Study Duration

The duration of the study, including enrollment, data collection, analysis, and dissemination, is expected to last five years. Follow-up visits will be allowed depending on the quality of the data collected and the availability of the participant. Each AO imaging session will occur within six months of a visit to the NEI. No more than three NEI clinical visits and five FDA AO imaging sessions will be performed in a year.

5.0 Data Management Plan

5.1 Storage of Data and Samples

No blood, tissue or other samples will be collected or stored in this study. Participant records from the NIH eye exams will be kept at the NIH eye clinic as described in Section 5.3. NIH clinical retinal images and data will be transferred to the FDA investigators only with participant consent. In general, the only NIH clinical information and data necessary to transfer to the FDA investigators will be the age and sex of the participant (to balance study demographic characteristics), lens status, ocular disease state and status, ocular axial length, fundus images, OCT images, and visual field maps. Section 4.2 describes the reasons this data needs to be shared with the FDA investigators. To protect participant's personal identifiable information (PII) and protected health information (PHI) and prevent its release to non-investigators, NIH clinical data (especially images) transferred to the FDA will be de-identified by the NIH by removing all PII, including name, address, social security number, and date-of-birth age. All NIH clinical data will be transferred by the FDA PI or Co-PI and stored only on a password-protected Ironkey external hard drive (i.e., not transferred to an FDA computer) and kept in a locked file cabinet in the FDA PI's office (WO62.1106). The external hard drive uses Data Locker software (FIPS 140-2 compliant) which meets all FDA and NIH IT requirements.

The only site of AO data and image collection will be the high-resolution imaging laboratory of FDA/CDRH/OSEL/DBP on the White Oak Campus. The acquired data will be initially stored on the AO system computers, which are password protected accessible only by study investigators. Because of the potential size of volumetric image datasets, which can be tens of gigabytes for a single participant for a single imaging session or more, data will be transferred to a dedicated server, which is password protected and accessible only by study investigators. All data collected at the FDA will be coded and will not contain any PII. AO images and data will be shared with the NIH/NEI co-PI and AIs for analysis and publication preparation. In general, the only data necessary to transfer from FDA to NIH will be processed images. The FDA AO data will be transferred to the NIH investigators via a password-protected Ironkey external hard drive and will be stored and accessed on the NIH CC EMR system or the NIH investigator's password protected (2-step authentication via PIV card) local computers. Since the images are already free of identifiable markers, there will be no need for de-identification.

5.2 Privacy

All research activities will be conducted in a private setting, with only the participant and investigators in the imaging laboratory.

5.3 Confidentiality

All records will be kept confidential and will only be reviewed by Drs. Hammer, Liu, Chew, and the NIH and FDA AIs. Electronic copies of the signed ICFs will be stored on the NIH CC EMR system. The Certificate of Confidentiality issued by the FDA in accordance with new requirements under the 21st Century Cures Act is included in Appendix 7.

For the NIH eye exam, records will be kept at the NIH eye clinic in the same manner as records from participants in other NIH-sponsored clinical studies. Information will be stored in the NIH's EMR system, which is accessible only by trained staff on password protected computers. Some information is collected in encrypted electronic records, which are stored on password protected computers. Except coded clinical data and retinal images discussed above in Section 5.1, all participant information remains on-site at the NIH eye clinic and is accessible only by trained staff and maintained with confidentiality abiding NIH policies and all relevant regulations regarding medical records.

For the FDA imaging session, a participant's name, age, and sex will be available to the FDA investigators to ensure a diverse population is represented, when age-matched participants are required, and to compare retinal images across different age groups. A participant number will be assigned to each participant and all information collected during the imaging session (image videos and ocular aberration data) will be coded using their participant number. Unlike clinical retinal images, AO images are not considered PII because the retinal

patch imaged is too small (≤2 deg.) to reconstruct a complete map of retinal vasculature for participant identification and no medical diagnosis will be made using the AO images. The only link between participant number and PII (name, age, and sex) will be a single cross-referenced list, which will be kept in a password-protected excel spreadsheet. Any data publicly disclosed cannot be linked back to the participant. Only Drs. Hammer, Liu, and Chew and the NIH and FDA AIs will have access to this electronic document.

5.4 Analysis Plan

De-identified AO data, particularly OCT and SLO videos will be analysed with custom software written by the investigators in LabVIEW and Matlab or commonly available image processing (ImageJ, Photoshop) and statistical programs. Examples of image processing and analysis code for AO applications include image registration, montaging or mosaic generation, capillary tortuosity and flow, and photoreceptor, RPE, and ganglion cell identification and quantification (cone packing, tessellation, Voronoi analysis). We may also work with collaborators external to this study on new analysis methods where de-identified participant data is shared. Because only de-identified information will be processed, a direct breach of confidentiality is not possible.

5.5 Data Sharing

De-identified AO images and other data from the study will be shared with internal and external professional colleagues, students, and other trainees during scientific and educational interactions. De-identified data at the NIH/NEI site may also be shared with collaborating laboratories at NIH or outside of NIH and/or submitted to NIH-designated repositories and databases. Repositories receiving data and/or samples from this protocol may be open-access or restricted access. Required approvals from the collaborating institution will be obtained and materials will be shipped in accordance with NIH and federal regulations.

Any new knowledge gained on disease mechanism, progression, diagnostic methods, clinical endpoints, and new disease biomarkers will be shared with the public in the form of peer-reviewed journal articles and public presentations at scientific conferences. Data sharing and participant engagement will be accomplished on an individual basis upon request. Upon IRB approval, the investigation will be registered on 'Clinicaltrials.gov'. Because this is an exploratory study on multiple disease types, the outcome measures reported to clinicaltrials.gov are not specified at this point. In general, for any AO structural or functional measure tested, we will compare results from a disease group and an age-matched control group. We will use all available resources (registries, websites, etc.) to share the results of this study with the public and patient advocacy groups. Confidentiality will be maintained through-out the reporting and data sharing process.

5.6 Quality Assurance

The FDA PI and co-PI will oversee the integrity and security of clinical data and images acquired within the purview of this protocol that are stored on the FDA campus. The NIH co-PI will oversee the integrity and security of clinical data and informed consents stored on the NIH EMR system. Quality assurance monitoring will be performed by the FDA PI at least annually. Quality assurance monitoring will include: a. Assuring that the patient spreadsheet is up-to-date and accurate; b. Assuring that all stored clinical data is de-identified, removed from any temporary locations, including the system computers, and archived properly on the dedicated server; and c. Assuring that all AO imaging data (which already contains no identifiable information) is archived properly on the dedicated server. The PIs and AIs will be responsible for the collection, processing, storage, retrieval, quality control and annotation of images and data collected during this study. All monitoring of the informed consent documents and process will be performed as specified in Section 3.9.

5.7 Study Conclusion

At the conclusion of the study, all FDA AO data will be removed from all computers and Ironkey external hard drives and archived on the dedicated server. Only the study investigators will have access to the dedicated server, which is housed in a locked FDA laboratory.

6.0 Patient Risks

6.1 Summary of Potential Risks

In general, there are three potential risks to the participants in this imaging study:

1. The participant has an allergic reaction or other complication to the mydriatic agent (increase in intraocular pressure),
2. The participant is exposed to a laser light level that exceeds the maximum permissible exposure (MPE) limit that damages the retina, or
3. The participant becomes injured during imaging from their presence in a research laboratory setting near scientific instrumentation.

Mydriatic drugs work by inhibition of the parasympathetic (i.e., contractive) response of the eye by blockage of muscarinic acetylcholine receptors or by stimulus of the sympathetic response of the eye by blockage of the re-uptake of noradrenaline. Side effects and complications associated with mydriatic drugs include photophobia, and in extremely rare cases, an increase in intraocular pressure leading to acute glaucoma. Mydriatic drugs are necessary for this investigation because the benefit from adaptive optics is optimal for dilated pupils larger than six (6) mm. Only those participants known (from clinical administration conducted by Dr. Chew) not to have severe adverse reactions to mydriatic agents will be enrolled in the study. All participants will experience some level of photophobia similar to that associated with a routine visit to their ophthalmologist. The effect of the drugs usually lasts four to six hours.

The second risk, light exposure to focused (ANSI) Class II or higher laser power levels, has to do with the interaction between imaging beams that are directed to the participant's eyes and their retina (the only reason the devices are minimally invasive rather than non-invasive). The laser safety analyses in Appendix 2a and 2b describe the technical details of the risk. Given the mitigation strategy, this risk has a very low likelihood of occurring but moderate severity if it does occur.

The third risk involves injuries that can occur when a participant unfamiliar with research enters a laboratory. The optics laboratory is not a wet lab, and so does not have harmful chemicals. The participant has one ingress/egress path to the AO imaging systems. Therefore, this risk has a very low likelihood of occurring and low severity if it does occur. This protocol and the imaging procedures described, as well as the eye exam, have been examined by the CDRH safety officer and found to be acceptable (Appendix 5).

6.2 Risk Mitigation Strategy

We will recommend that each participant avoids sunlight and wears sunglasses outside to mitigate photophobia. More severe complications (allergic reaction to mydriatic drugs) will be monitored by Dr. Chew or the NIH/NEI AIs for 30 minutes after administration during the initial NIH eye exam and by the PI and co-PI during the FDA imaging sessions. The eye exam is designed to provide significant reduction in risk of reaction by screening participants with a predisposition or history of AACG.

The primary mitigation for the second risk Is to maintain power levels that are below ANSI MPE limits. The power at the cornea will be measured each day an imaging session takes place and recorded in a log-book. The SLO and OCT illumination sources are superluminescent diodes (SLD), the output power of which are controlled from the computer. The SLD electronic drive boards have current limits that prevent excessive output power being delivered to the participant's eye. When SLDs fail, their power output will drop significantly or altogether (i.e., the power never increases for device failure). Scanner failure will direct a focused beam to the retina but also stop imaging, which will be nearly immediately noticed by the operator (within seconds), who can tell the participant to sit back from the instrument. The visible light stimulus source delivery is controlled with a shutter that precisely sets the illumination timing (pulse duration, etc.).

The third risk is mitigated by common laboratory safety practices that are in place at DBP and described in our BSC safety manual, which contains the emergency response plan as well as a description of procedures for dealing with chemical safety, biosafety, laser safety, and issues related to hazardous waste. OSEL complies

with all Occupational Safety and Health Administration (OSHA) regulations and conducts routine safety training for its staff.

6.3 Classification of Risk
The FDA IRB has classified this protocol as a greater-than-minimal risk study.

7.0 Data and Participant Safety Monitoring and Adverse Event Reporting

7.1 Data Monitoring
Besides ICF and consent process monitoring, which will be performed by Emmes at NIH (see Section 3.9), all data monitoring will be continuously self-administered by the pIs, Co-pIs, and aIs. Data integrity procedures will be reviewed at least annually. NIH clinical data will be stored on the secure EMR system (see Section 5.3), and data monitoring will mainly involve routine audits (at least once a year) to ensure that the data hasn't been corrupted or compromised. FDA AO images and data will be stored on the AO system computer in the short-term and moved to a dedicated, secure server for long-term storage and archiving. Any clinical data transferred from NIH to FDA will be kept on a password-protected Ironkey external drive (see Section 5.1). Data monitoring at the FDA will include routine audits of the AO system computer, external drive, and secure server to ensure the data is secure and uncorrupted.

7.2 Participant Safety Monitoring
The pIs, co-pIs, and aIs will continuously monitor participant safety related to this protocol. Participant safety procedures will be reviewed at least annually. Participants will be monitored for adverse events by the FDA PI, FDA and NIH co-pIs, and NIH/NEI aIs during all NIH ophthalmic exams and FDA imaging sessions. FDA and NIH policies, standard operating procedures, and oversight (managerial and various safety officers) are also in place to govern general safety concerns related to these exams and imaging sessions.

7.3 Adverse Event and Unanticipated Adverse Device Effects Reporting
The PI is responsible for detecting, documenting, and reporting Unanticipated Problems Involving Risks to Subjects or Others (UPIRTSOs), suspensions, terminations, adverse events (aEs), including serious adverse events (SAEs), and non-compliance with FDA policy, IRB requirements, and federal regulations. Adverse events can include conditions that affect either or both eyes (e.g., pain) or the whole body (e.g., dizziness). Unanticipated problems, as defined by the FDA SOP on the topic [15], will be reported, including death, regardless of whether the death is expected and/or unrelated to the research.

Unanticipated adverse device effects will also be reported to the FDA within 10 working days after the effect per 21 CFR 812.150(b)(1). The PI will report all such events to the FDA IRB and provide any additional information which the committee may require to evaluate the severity of the event. Unanticipated adverse device effects that present an unreasonable risk to subjects (as determined by the FDA PI, who is also the sponsor) will result in study termination within 5 working days per 21 CFR 812.46(b)(2). Terminated studies may not be resumed without FDA and IRB approval.

The risk probability of adverse events is expected to be extremely low. Participants will be asked at the end of imaging if they notice any changes in vision, apart from those normally experienced during pupil dilation. Participants will be provided with the contact information of the PI (on the ICF) to report any delayed effects of imaging.

7.4 Policy Regarding Research-Related Injuries
The NIH Clinical Center will provide short-term medical care for any injury resulting from participation in the research. In general, no long-term medical care or financial compensation for research-related injuries will be provided by the FDA, NIH, the NIH CC, or the Federal Government. However, participants have the right to pursue legal remedy if they believe that their injury justifies such action.

8.0 Investigator Roles and Qualifications

Daniel X. Hammer, PhD, is the PI and is responsible for the conduct and oversight of all aspects of the study. He has 25 years of experience in biomedical research and has been the PI on many human subject protocols,

including for AO imaging. He will be a primary operator of the AO systems during human subject imaging sessions. With Dr. Liu, he designed and built the FDA multimodal AO systems.

Zhuolin Liu, PhD, is a Co-PI and will be a primary operator of the AO systems during human subject imaging sessions. She is first author on several seminal AO-OCT papers and has participated in several human subject investigations. With Dr. Hammer, she designed and built the FDA multimodal AO systems.

Emily Chew, MD, is the NIH NEI PI and medical advisor for the protocol. She is Division Director for the Div. of Epidemiology and Clinical Applications at NIH/NEI. She will perform the initial ophthalmic exam, train personnel in the administration of eye drops, obtain informed consent, and provide oversight. Dr. Chew is an active clinician and ophthalmic surgeon. She is the principal investigator on several clinical trials in her current position at the National Eye Institute.

Anant Agrawal, PhD, is a research scientist in FDA/CDRH/OSEL/DBP and participates in many of the AO imaging studies, both those that involve human subjects and others. He will operate the AO system occasionally. He is leading the AO retinal phantom development effort and has over 12 years of experience with OCT instrumentation, phantom development, and data analysis.

Achyut Raghavendra, PhD, is a post-doctoral fellow in the FDA AO laboratory and participates on the AO glaucoma imaging study. He will operate the AO systems occasionally and help with AO data processing and analysis.

Tiarnán Keenan BM BCh, PhD, Laryssa A. Huryn, MD, and Wadih M. Zein, MD, Alisa Thavikulwat, MD are retinal physicians at the NEI. They will perform the initial ophthalmic exam, train personnel in the administration of eye drops, obtain informed consent, and provide oversight. They are all principal investigators and/or co-investigators on various clinical trials in their current positions at the NEI.

Tharindu De Silva, PhD, is an imaging researcher in the Unit on Clinical Investigation of Retinal Disease at the NEI at the NIH/NEI. He has background in bioengineering and image analysis.

Kapil Bharti, PhD, has expertise in experimental models of eye disease and has been published widely on their biophysical, molecular, and developmental characteristics.

Catherine Cukras, MD, PhD is an NIH special volunteer and formerly the NIH NEI PI for this protocol. She will provide consultive support to the protocol on clinical matters.

Johnny Tam, PhD, is an investigator at NIH/NEI and is an active collaborator with the FDA team. He has an AO imaging system in the NIH clinical space, where he has a human subject protocol to image subjects with retinal degeneration and other diseases.

8.1 Conflicts of Interest

No investigator has a conflict of interest according to FDA and NIH guidelines on conflicts of interest.

9.0 References

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10.0 Appendices

1. Informed consent form
2. Laser safety analysis for:
    a. mAO imager
    b. AO-FDML imager
3. IDE abbreviated filing requirements justification
4. NIH consenting script
5. CDRH Safety Officer recommendation
6. CDRH Laser Safety Officer recommendation memo
    a. mAO imager light safety analysis review
    b. AO-FDML imager light safety analysis review
7. Certificate of Confidentiality
8. NIH (a) and FDA (b) maps and security checkpoint instructions for non-FDA employees
9. Sample recruitment email
10. Investigators CV and CITI training certification