# The Use of Electrophysiologic Techniques in Vision Research

# Course organizers

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# Presentations

Presenters and presentations may change.

| **Time**  | **Topic** | **Speaker**  |
| --- | --- | --- |
| 8:30-8:45 | Welcome and Introduction | Mitchell Brigell, PhD FARVOOccuphire Pharma (Consultant) |
| I will give a brief introduction to basic electrophysiological techniques. Potocols for isolating rod and cone pathways will be presented, as well as extended protocols for probing the visual pathways. Components of the full-field electroretinogram, pattern electroretinogram, multifocal electroretinogram, and pattern-reversal visual evoked potential will be presented. The conservation of these responses across species enables their use in translational research. The use of these techniques to understand the pathophysiology of disease and the functional impact of therapeutic intervention will be introduced. |
| 8:45-9:15 | Cellular Origins of the electroretinogram | Laura Frishman, PhD, FARVOUniversity of Houston |
| The electroretinogram (ERG) is a useful tool for noninvasively assessing retinal function in the clinic and the laboratory, using electrodes that make corneal or skin contact. The ERG arises from extracellular currents flowing in retinal tissue that result from neuronal signaling and in some cases, potassium buffering by glial cells. With appropriately applied stimuli and analyses, the ERG can provide useful, objective information about the functional integrity of the cells and retinal circuits that contribute to its generation. This lecture will examine the current state of knowledge with regard to the origins and mechanisms of generation of ERGs evaluated in standard clinic testing, and in more specific applications. A variety of approaches have been used to study the origins of the ERG, largely in animal models: intraretinal recordings under different stimulus conditions, pharmacologic manipulations of neurotransmission to dissect responses generated by the different retinal cells and circuits, selective lesions of particular cell types, and studies of genetically altered retinas. Current knowledge about origins of ERG waves using these approaches will be summarized in this lecture. |
| 9:15-9:45 | Applications of Isolated Photoreceptor Electrophysiologic techniques | Vladimir Kefalov, PhDUniversity of California Irvine |
| The quantitative analysis of rod and cone photoreceptor function is critical for understanding the mechanisms that regulate the physiology of these cells in health and disease. Although classical in vivo electroretinography is very useful in determining the general health of the outer retina and the diagnosis of associated visual dysfunctions, it is not easily amenable to more detailed analysis. One alternative approach that has gained popularity in animal research is the recording of electric signals from the isolated retina. When combined with genetic tools, in the case of mice, this method allows for isolation of the responses from rod and from cone photoreceptors in a wide range of light conditions. Pharmacologically blocking synaptic transmission and the propagation of the signal to bipolar cells that produce the b-wave, unmasks the full waveform of the photoreceptor response that is otherwise largely blocked by the b-wave. Such a pharmacological approach also enables the derivation of downstream bipolar cell and Müller cells responses from isolated retinas. Selectively blocking phototransduction in either rods or cones by genetic tools allows the isolation of the corresponding cone or rod signal in scotopic, mesopic, and photopic conditions. Finally, light adaptation and dark adaptation following exposure to bright bleaching light can also be studied selectively in rod or cone photoreceptors after isolating their responses. Commercial and 3D-printed recording chambers make it possible to adapt existing in vivo ERG systems for ex vivo electroteninography of retinal tissue of any kind, from mouse to fish, primate, and human. This approach also allows for the rapid screening of drugs applied to the isolated retina through the perfusion solution. |
| 9:45-10:15 | Application of electrophysiologic techniques to understanding of chromatic processing | Jay Neitz PhDUniversity of Washngton |
| Objective measures of cone function in humans and other mammals have the challenge that our retinas have a rod-to-cone ratio of 20:1 or greater. Thus, to study cones using the electroretinogram (ERG), we must overcome the fact that their signals are relatively small against a background of signals from rods. To overcome this challenge, we have adapted a standard psychophysical technique, flicker photometry, to the ERG, whereby the intensity of a flickering test stimulus is adjusted until it produces an ERG signal equal to that produced by a flickering reference light. Rod responses are eliminated by using frequencies (i.e., 31.25 Hz) they cannot follow. Averaging over many stimulus presentations produces a high signal-to-noise ratio. Moreover, any changes that occur in the ERG over a test session will be equally reflected in the responses to both test and reference lights and have minimal influence on the measured relative spectral sensitivity function. In dichromatic eyes, one can constrain the ERG spectral measurements to reflect the signals of only a single class of cone. Such measurements, made noninvasively, have illuminated the variety and distribution of cone photopigments in mammals, including common laboratory rodents and domestic animals such as cats, cows, and horses. Moreover, by using the ERG to measure the spectral peaks of variant middle (M) to long (L) wavelength-sensitive photopigments in primates and correlating them to their gene sequences, it has been possible to discover the amino acid changes responsible for tuning the spectra of the cone photopigments in the middle-to-long wavelengths, solving a central problem in vision science. In human trichromats, ERG flicker photometric spectral sensitivity functions can be used to estimate the ratio of L:M cones. A survey of normal individuals revealed a large range of individual differences in L:M cone ratio. These differences have surprisingly little effect on some measures of color vision but significantly impact others. The L:M cone ratio also differs considerably between different human groups, and it affects a person’s susceptibility to myopia. Finally, ERGs can be recorded using “cone isolating” stimuli in both flicker and longer flash paradigms. This has been used to assess S-cone function and evaluate the efficacy of gene therapies that aim to add functional photopigments to the eye. |
| 10:15-10:30 | Morning Break |  |
| 10:30-11:00 | Probing the Visual System with ISCEV Extended Protocols | Anthony Robson, PhDMoorfields Eye Hospital |
| The International Society for clinical electrophysiology of vision (ISCEV) specifies minimum standards for routine clinical testing of the visual system in order to optimise basic methods and to allow meaningful inter-laboratory comparisons. The ISCEV also publishes extended protocols for non-standard testing, with the aim of promoting convergence of techniques that are used in the clinical setting, often as an addition to the minimum ISCEV procedures. This presentation will outline the rationale underpinning the main ISCEV extended electroretinogram (ERG) protocols including the dark-adapted red flash ERG, photopic On-Off ERG, S-cone ERG and other techniques. Typical clinical applications will be illustrated in cases of inherited and acquired retinal disease, highlighting how the extended protocols may be used to complement standard methods, to aid diagnosis and patient management and to probe and detail retinal function phenotypes. |
| 11:00-11:30 | Application of visual electrophysiology in animal models of outer retinal disease | Neal Peachey, PhDCole Eye Institute / Cleveland VMAC |
| Non-invasive electrophysiology continues to provide an important bridge between animal models of human retinal disease and their human counterparts. In particular, electroretinogram (ERG) signals that reflect the activity of the major classes of retinal cells may be evoked using strategies already in use for clinical diagnosis and research. This presentation will review these approaches and present examples from the animal model literature highlighting the different functional phenotypes that can be captured using the ERG including several that are more readily obtained from mice than human. |
| 11:30-12:00 | Electrophysiological outcome measures in therapeutic trials for Inherited Retinal Degenerations (IRDs) | David Birch, PhD FARVORetina Foundation of the Southwest |
| This talk will describe the ways that the full-field electroretinogram (ERG) has been used during the past 20 years in treatment trials for IRDs. From trials with nutritional supplementation to current Phase 1/2 gene therapy trials, the ERG has been a useful outcome measure for both safety and efficacy. Techniques have been developed to provide objective and sensitive measures of both photoreceptor and inner retinal function. We will discuss limitations of electroretinography in IRDs including the lack of measurable signals in advanced patients, the inter-visit variability, and the insensitivity to local variations in retinal function. Trial designs and results will be presented where the ERG was used in x-linked retinitis pigmentosa, x-linked retinoschisis and autosomal dominant retinitis pigmentosa. |
| 10:30-10:45 | Questions |  |
| 12:15-1:15 | Lunch  |  |
| 1:15-1:45 | Application of visual electrophysiology in pediatric ophthalmology: clinical and research | Arlene Drack, MDUniversity of Iowa |
| Special techniques for acquiring electrophysiology data in children will be presented. Presentation of cases in which electrophysiology was critical to diagnosis will be accompanied by how these results can help guide molecular genetic testing. Use of electrophysiology as a research tool, for example in subretinal gene therapy, will also be explored. |
| 1:45-2:15 | The use of electroretinography in diabetic retinopathy | Machelle Pardue, PhDFARVOGeorgia Tech |
| Diabetic retinopathy is currently diagnosed by the appearance of retinal vascular pathology which can take many years to develop after diabetes develops. There is an urgent need to develop new biomarkers for diabetic retinopathy that would provide early detection and monitoring of this disease. Electroretinography (ERG) studies have revealed retinal dysfunction in early-stage diabetic retinopathy, prior to vascular pathology. Additionally, ERG responses show progressive changes with the duration of disease. This talk will review data from diabetic animal models and humans that show changes in ERG parameters, including delays in oscillatory potentials in response to dim flash stimuli. The utility of ERG for early detection of diabetic retinopathy in order to apply neuroprotective approaches such as dopamine treatment will be discussed. |
| 2:15-2:45 | The use of visual electrophysiology in Diabetic Retinopathy -Clinical | Jason McAnany, PhDUniversity of Illinois, Chicago |
| The electroretinogram (ERG) is a noninvasive, objective technique that has become increasingly important for understanding retinal dysfunction in diabetic eye disease. In this seminar, I will summarize findings from recent clinical studies that have used the full-field ERG, multifocal ERG, and pattern ERG to evaluate neural dysfunction in patients with diabetes. The strengths and limitations of the ERG as applied to diabetic eye disease will be reviewed. Recent work from my laboratory, and others, has provided evidence for possible photoreceptor dysfunction in early-stage disease, whereas pattern ERG data and photopic negative response analyses indicate inner retina dysfunction. Multifocal ERG studies that have shown spatially localized neural abnormalities that can predict the location of future microaneurysms will also be reviewed. |
| 2:45-3:00 | Afternoon Break |  |
| 3:00-3:30 | Application of visual electroretinography in rodent models of glaucoma | Bang Bui, PhD University of Melborne |
| Pre-clinical studies of optic nerve injury models have led to significant insight into the mechanism underlying ganglion cell neurodegeneration. During the process of ganglion cell injury, morphological changes can occur prior to cell death. Similarly, following injury changes in ganglion cell responses can occur in the absence of substantive structural changes. Subtle changes to ganglion cells and the retina can often be detected using functional tools such as the electroretinogram. Moreover, the electroretinogram is a sensitive and complementary means to quantify treatment efficacy. This presentation will describe *in vivo* electroretinography for assessing ganglion cell injury in rodent models. |
| 3:30-4:-- | Electrophysiologic measures of retinal ganglion cell dysfunction in glaucoma | Suresh Viswanathan, PhDFARVOSUNY College of Optometry |
| This presentation will include the following:1. An outline different types of visual electrophysialogical measures that are employed to assess retinal ganglion cell/optic nerve function
2. Recording paradim for each electrophysiological measure that will give the best results
3. How to choose the appropriate elecctrophysiological measure for the targeted population
4. Highlight results from published and unpublished studies of glaucoma involving either animal models or human patient to illustrate the use of electrophysiological measures for detection, monitoring progression and and assessing treatment effects on retinal ganglion cell function
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| 4:00-4:30 | Questions for Afternoon Speakers |  |
| Open discussion |
| 4:30 PM | Adjourn |  |