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Standard for clinical electroretinography (1999 update)

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Introduction

Full-field electroretinography (ERG) is a widely used ocular electrophysiologic test. In 1989 a basic protocol was standardized so that certain responses could be recorded comparably throughout the world [1]. This document was updated in 1994 [2]. Standards for five commonly obtained responses were presented:

(1) A response developed by the rods (in the dark-adapted eye)
(2) A maximal response in the dark adapted eye
(3) Oscillatory potentials
(4) A response developed by the cones (in the light-adapted eye)
(5) Responses to a rapidly repeated stimulus (flicker)

This document, an updated version of the standard, is intended as a guide to practice and to assist in interpretation of ERGs. The five basic responses represent the minimum of what an ERG evaluation should include. The standard describes simple technical procedures that allow reproducible ERGs to be recorded under a few defined conditions, from patients of all ages (including infants). Different procedures can provide equivalent ERG responses. It is

Dr. Marmor was Chair of the ERG Standardization Committee of the International Society for Clinical Electrophysiology of Vision (ISCEV) and Dr. Zrenner was President of the ISCEV. This document was approved by the International Society for Clinical Electrophysiology of Vision in Eilat, Israel on April 15, 1999

Table 1. *Specialized types of ERG (not covered by ISCEV standard)

Macular or focal ERG
Multifocal ERG
Early receptor potential (ERP)
Scotopic threshold response (STR)
Direct-current ERG
Long-duration flash ERG (on-off responses)
Bright-flash ERG
Double-flash ERG
Chromatic stimulus ERG (including S-cone response)
Dark and light adaptation of the ERG
Stimulus intensity-response amplitude analysis (Naka-Rushton)
Saturated a-wave slope analysis

incumbent on users of alternative techniques to demonstrate that their procedures do in fact produce signals that are equivalent in basic *waveform, amplitude, and physiologic significance* to the standard.

Our intention is that the standard method and responses be used widely, but not to the exclusion of additional responses or tests that individuals laboratories may choose or continue to use. We recognize that the investigation of certain eye conditions may not require all five of the standard responses. There are also specialized types of ERG which may provide additional information about retinal function (see Table 1) that are not covered by this standard. We encourage electrophysiologists to learn about and try expanded test protocols and newer tests to maximize the diagnostic value of the ERG for their patients. ISCEV guidelines for the calibration of electrophysiologic equipment [3], recommendations for recording the pattern ERG [4], and standards for the electro-oculogram [5] and visual evoked potentials [5] have also been published.

Because of the rapid rate of change of ERG techniques, these standards will be reviewed every four years. We have made recommendations that commercial recording equipment have the capability to record ERGs under conditions that are outside the present standard but that are nevertheless either widely used or likely to be needed in the future. Note that this document is not a safety standard and does not mandate particular procedures for individual patients.

The organization of this report is as follows:

Basic technology

There are refinements to the original standard throughout this document, and sections with major changes or additions are marked with an asterisk (*)

Basic technology

Light diffusion

Full-field (Ganzfeld) stimulation should be used. With focal flashes, the area of retinal illumination is not uniform, and its extent is unknown (although focal flashes may be used for certain specialized ERG tests). Full-field dome stimulators are generally preferable to ocular diffusers (e.g., 100-diopter or opalescent contact lenses) since it is difficult with the latter to measure the extent and intensity of retinal illumination. It is incumbent on manufacturers and users of lens diffusers to verify true full-field stimulation of determinable strength.

Electrodes

*Recording Electrodes. Electrodes that contact the cornea or nearby bulbar conjunctiva are strongly recommended for basic full-field recording. These include contact lens electrodes, conductive fibers and foils, conjunctival loop electrodes and corneal wicks For most users, contact lens electrodes will provide the highest amplitude and most stable recordings; such electrodes should be centrally transparent with an optical opening as large as possible, and incorporate a device to hold the lids apart. The corneal surface should be

protected during use with a non-irritating and non-allergenic ionic conductive solution that is relatively non-viscous (e.g., no more viscous than 0.5% methyl cellulose). More viscous solutions can attenuate signal amplitude. Other types of corneal and conjunctival electrodes require more skill to use but may have certain advantages. Users should be aware that as the point of ocular contact moves away from the corneal apex, signal amplitude is reduced. Topical anesthesia is necessary for contact lens electrodes but may not be required for other types of corneal and conjunctival electrodes. It is incumbent for all electrophysiologists to master the technical requirements of their chosen electrode, to insure good ocular contact, to insure proper electrode impedance, to insure that waveforms are comparable to standard responses, and to define both normal values and variability (which may be different with different electrodes) for their own laboratory. Skin electrodes are in general not recommended as active recording electrodes.

Reference electrodes. Reference electrodes may be incorporated into the contact lens-speculum assembly to make contact with the conjunctiva ('bipolar electrodes'). This is the most stable configuration electrically. Alternatively, electrodes can be placed near each orbital rim temporally as a reference for the corresponding eye. The forehead has also been used as a reference electrode site, although there is a theoretical risk of signal contamination by ocular cross-over or from cortical evoked potentials. Users are advised to avoid other positions.

Ground electrodes. A separate skin electrode should be attached to an indifferent point and connected to ground. Typical locations are on the forehead or ear.

* *Skin reference electrode characteristics.* The skin should be prepared by cleaning and a suitable conductive paste or gel used to insure good electrical connection. Skin electrodes used for reference or ground should have 5kΩ or less impedance measured [3] between 10 and 100 Hz. If more than one skin electrode is used (e.g., for reference and ground) they should all have similar impedance.

Electrode stability. Whatever corneal and reference electrode system is used, the baseline voltage in the absence of light stimulation should be stable. Some reference electrode systems may need to be made of non-polarizable material to achieve this stability.

Electrode cleaning. Recording the ERG involves the exposure of corneal electrodes to tears and exposure of the skin electrodes to blood if there has

been any abrasion of the skin surface. We advise that electrodes be suitably cleaned and sterilized after each use to prevent transmission of infectious agents. The cleaning protocol should follow manufacturers' recommendations and current standards for devices that contact skin and tears.

Light sources

* *Stimulus duration.* The standard is based on stimuli of duration considerably shorter than the integration time of any photoreceptor. Thus, the light stimulus should consist of flashes having a maximum duration of about 5 ms [Note 1]. Short-flash durations may be obtained from gas discharge tubes, from stroboscopes, and potentially from other devices.

Stimulus wavelength. Most flash stimuli in use have a color temperature near 7000° K, and they should be used with domes or diffusers that are visibly white. Colored filters are used by some laboratories to enhance the separation of rod and cone responses, but this is not part of the standard [Note 2].

* *Stimulus strength. A standard system* is defined as one that produces a stimulus strength (in luminous energy per square meter) at the surface of the Ganzfeld bowl of 1.5-4.5 photopic $cd \cdot s \cdot m^{-2}$ (candela-seconds per meter squared) [Note 3]. This is equivalent to luminance·time, measured as $cd \cdot m^{-2} \cdot s$. Note that these are photometric units and that 3.43 $cd \cdot m^{-2}$=1fL (foot-Lambert) [Note 4]. A flash of this strength will be called the *standard flash* (SF).

Background illumination. In addition to producing flashes, the stimulator must be capable of producing a steady and even background luminance of 17– 34 $cd \cdot m^{-2}$ (5 to 10 fL) across the full field. For this standard, a white background is used, but we recognize that colored backgrounds may also be used for special purposes.

Light adjustment and calibration

Adjustment of stimulus and background intensity. Methods of modifying both the stimulus and background intensity must be provided. We recommend that a standard system be capable of attenuating flash strength from the SF over a range of at least 3 log units, either continuously or in steps of no more than 0.3 log unit. The method of attenuation should not change the wavelength composition of either the flash or background luminance. We recognize that the stimulus and background requirements for a full range of ERG tests will be more extensive and more stringent, and we recommend that equipment manufacturers exceed the minimum standard [Note 5].

Stimulus and background calibration. The stimulus strength (in luminance·time) produced by each flash on the surface of the full-field stimulus bowl must be documented by the user or manufacturer, ideally with an integrating photometer (luminance meter) placed at the location of the eye. The light output per flash of most stroboscopes varies with the flash repetition rate; therefore, separate calibrations will need to be made for single and repetitive stimuli. The photometer should also record the background luminance of the stimulus bowl's surface, in a non-integrating mode. The photometer must meet international standards for photometric measurements based on the photopic luminous efficiency function (photopic luminosity curve), and must be capable of recording the total output of very brief flashes. Users are urged to consult the ISCEV guidelines for calibration of electrophysiologic equipment [3] for a more detailed treatment of calibration procedures. We recommend that manufacturers of stimulators supply a suitable photometer with their equipment.

Recalibration. See the ISCEV guidelines for calibration [3]. Light output from the dome varies with time from changes in the flash tube, the tube power source, line voltage, the background light bulbs, the attenuation systems, or the paint in the dome. This may be especially critical for background illumination provided by incandescent sources. Responsibility for electronic stability and warnings about sources of instability should rest with the manufacturers of the equipment; however, at present this cannot be presumed. A stabilizing transformer will minimize line voltage variations, if they are a problem. The frequency with which recalibration of flashes and backgrounds is required will vary from system to system and could be as high as weekly for some units. Self-calibrating units are to be encouraged.

Electronic recording equipment

* *Amplification* We recommend that the bandpass of the amplifier and preamplifiers include at least the range of 0.3 to 300 Hz and be adjustable for oscillatory potential recordings and special requirements. We advise that the input impedance of the preamplifiers be at least 10 MΩ. Amplifiers should generally be AC (alternating current) coupled (i.e., capacitatively coupled) and capable of handling offset potentials that may be produced by the electrodes [Note 6].

Patient isolation. We recommend that the patient be electrically isolated according to current standards for safety of clinical biologic recording systems in the user's country.

Display of data and averaging. We strongly recommend that the equipment that provides the final record be able to represent, without attenuation, the full amplifier bandpass. Good resolution can be achieved with oscilloscopes or computer-aided (digitizing) systems but not with direct pen recorders. To avoid a loss of information, digitizers should sample responses at a rate of 1000Hz or higher per channel. With computer-aided systems, it is important that responses be displayed promptly so that the operator can continuously monitor stability and make adjustments during the test procedure. Recording units that digitize ERG signals can usually average them as well, which may sometimes be useful.

Clinical protocol

Preparation of the patient

Pupillary dilation. We recommend that pupils be maximally dilated for all ERG recordings in this standard and that pupil size be noted if dilatation is, for any reason, less than maximal.

**Pre-adaptation to light or dark.* The recording conditions outlined below specify 20 minutes of dark adaptation before recording rod responses, and 10 minutes of light adaptation before recording cone responses. The choice of whether to begin with scotopic or photopic conditions is up to the user, as long as these adaptation requirements are met. If contact lens electrodes are used, the wearing time can be minimized by dark adapting first, and inserting the electrodes under dim red light at the end of the adaptation period. However, care should be used to avoid too bright a red light, and an additional 5 minutes of dark adaptation may be needed for recovery after lens insertion. Laboratories using non-contact lens electrodes may elect to record photopic responses before scotopic ones, since there is less risk of corneal irritation from these electrodes during the time of dark adaptation. In this case, photopic adaptation is unnecessary as long as the patient has been at photopic light levels for at least 10 minutes before recording. However, the dark adaptation period which follows photopic stimulation may need to be extended beyond 20 minutes depending on the duration and intensity of flash stimuli.

Pre-exposure to light. We advise that fluorescein angiography or fundus photography be avoided before ERG testing, but if these examinations have been performed, a period of dark adaptation of at least one hour is needed. It is usually preferable to record scotopic responses to weak flashes before

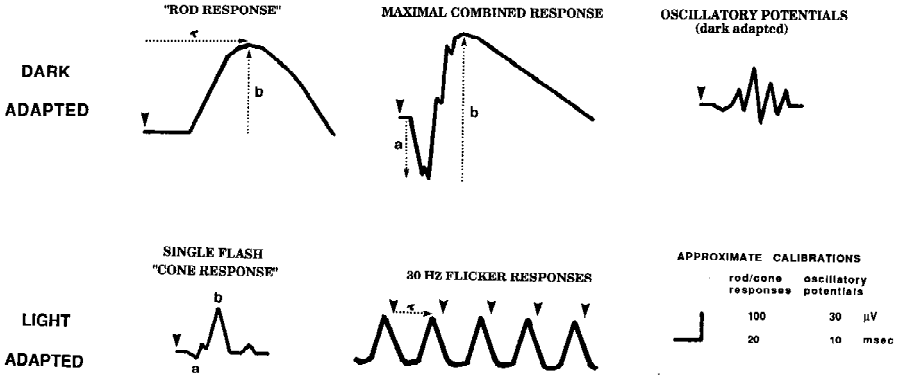

Figure 1. Diagram of the five basic ERG responses defined by the Standard. These waveforms are exemplary only, and are not intended to indicate minimum, maximum or even average values. *Large arrowheads* indicate the stimulus flash. *Dotted arrows* exemplify how to measure time-to-peak (τ, implicit time), a-wave amplitude and b-wave amplitude.

the mixed and cone responses to more intense flashes, to minimize light adaptation, and to reduce the time that the patient wears the contact lens electrode.

**Fixation.* A fixation point should be incorporated into stimulus domes. A stable eye is important so that eye movements do not alter the optimal corneal electrode position, produce electrical artifacts, or allow blockage of light by the electrode or eyelid. Patients who cannot see a fixation target may be instructed to look straight ahead and keep their eyes steady. Patients should be monitored to assess compliance, and account for difficulties in eye opening or fixation.

ERG measurements and recording

Measurement of the ERG. Both amplitude and implicit time should be measured for selected ERG signals. For practical purposes, the variables most often measured are the b-wave amplitudes of the 'rod response', maximal combined response and single flash 'cone response' and the b-wave time-to-peak of the single flash 'cone response' or 30 Hz flicker response. According to current convention, the a-wave amplitude is measured from baseline to a-wave trough, the b-wave amplitude is measured from a-wave trough to b-wave peak, and the b-wave time-to-peak is measured from the time of the flash to the peak of the wave (see Figure 1).

Oscillatory potentials. There is considerable debate in the literature about how to measure and describe oscillatory potentials [Note 7]. Their appearance

is highly dependent upon stimulus conditions, adaptation and amplifier filter characteristics, but most authors describe three major peaks often followed by a fourth smaller one. Simply observing the presence of these peaks, and their normality relative to the standards of the laboratory, may be adequate for many clinical purposes at our present state of knowledge.

Averaging. Averaging is not ordinarily required to record quantifiable ERG responses with the recommended types of electrodes. Averaging a limited number of responses may decrease variability and help to reduce background noise if present. Averaging may also be used to identify and measure very weak pathologic responses. Artifact rejection must be a part of any averaging system. Signal repetition rates should not exceed the recommendations in the standard for each response.

Normal values. We recommend that each laboratory establish or confirm normal values for its own equipment and patient population giving attention to an appropriate sample size. All ERG reporting (whether for local records, publication, or even for nonstandard responses) should include normal values and the *limits of normal*. Some manufacturers distribute norms for their standard protocols, and several large series have been published recently that give normative data. However, ERG norms for amplitude may have to be scaled up or down depending on where the users electrode rests on cornea or conjunctiva. Note that ERG parameters change rapidly during infancy and modestly with age thereafter. Because some ERG parameters (such as b-wave amplitude) are not necessarily normally distributed, calculations of standard deviation may be misleading. To describe the limits of normal, we recommend listing the median value (not the mean), and the actual values on either side of the median that bracket 95% of the normal responses (in other words, the 95% confidence limits determined by direct tabulation of responses).

Reporting the ERG. Standardization of ERG reporting is critical to the goal of having comparable data worldwide. We recommend that reports or communications of ERG data include representative waveforms of each of the standard responses displayed with amplitude and time calibrations and labeled with respect to stimulus variables and the state of light or dark adaptation. We suggest that when single flash stimuli are used without averaging, two waveforms of each response be displayed to demonstrate the degree of consistency or variability. The strength of stimulation ($cd \cdot s \cdot m^{-2}$) and the level of light adaptation ($cd \cdot m^{-2}$) should be given in absolute values. *The reporting forms should indicate whether the techniques of recording meet the international standard.* We recommend that patient measurements be listed along with the normal values and their variances (that must be provided on all reports). Fi-

nally, reports should note any conditions that are not specified by the standard, including type and position of electrode, sedation or anesthesia, and the level of compliance.

Pediatric ERG recording

The ERG can be recorded from infants and young children but some care must be taken to account for immature eyes and limited cooperation.

Sedation or Anesthesia. Most pediatric subjects can be studied without sedation or general anesthesia (topical anesthesia is necessary for contact lens electrodes). Small infants can be restrained if necessary. Unusually uncompliant children (especially ages 2–6 for whom containment can be difficult) may become compliant with oral sedation or anxiolysis. Medical guidelines should be followed with respect to indications, risks, medical monitoring requirements and the choice of a sedative/relaxant versus general anesthesia. Considering the variability of pediatric records, there will generally be little effect on ERG amplitude or waveform with sedation or brief very light anesthesia, although full anesthesia may modify responses.

**Electrodes.* Contact lens electrodes are applicable to infants and young children, but pediatric sizes will be required with speculum-containing models, and care must be used to minimize corneal and psychological trauma. Non-contact lens and skin electrodes vary in their applicability to children, and their greater comfort is often offset by greater movement or small signals which create electrical noise or artifacts. Special care is required with children to monitor electrode position and compliance in order to avoid artifactual recordings.

Normal values and measurement. ERG responses mature during infancy and newborn and infant signals must be interpreted with great caution. Later infantile and young childhood responses approach adult waveform and size. Pediatric ERG responses should ideally be compared to those from normal subjects of the same age, even though there may be little normative data available. Because movement and poor fixation can make pediatric records variable in amplitude and waveform, we recommend that several examples of each response be recorded in order to recognize reproducible waveforms and choose the best and largest of these reproducible responses. Standard protocols may occasionally need to be abbreviated in order to obtain the responses most critical to the diagnostic question under investigation. Reports should note the degree of cooperation and any medications used.

Specific responses

*Rod response

We recommend that the patient be dark-adapted for at least 20 minutes before recording the rod response (and longer if the patient had been exposed to unusually bright light). The rod response should be the first signal measured after dark adaptation, since it is the most sensitive to light adaptation. The standard stimulus is a dim white flash of strength 2.5 log units below the white SF (see above and Note 3); We advise a minimum interval of 2 seconds between flashes. A blue stimulus is equally appropriate if equated to the white standard [Note 2].

Maximal combined response

The maximal response is to be produced by the white SF, in the dark-adapted eye. We recommend an interval of at least 10 seconds between stimuli. This response is normally produced by a combination of cone and rod systems.

Oscillatory potentials

Oscillatory potentials are generally obtained from the dark-adapted eye, using the same white SF. They may also be recorded from the light-adapted eye. The high-pass filter must be reset to 75 to 100 Hz, so that an overall bandpass of 75 to 100 Hz on the low end and 300 Hz or above at the high end is achieved. Filters should attenuate sufficiently to achieve this result. Users should be aware that there are several types of electronic and digital filters, which may have different effects upon physiologic signals (e.g., phase shifts or ringing). More information about filter selection and use is presented in the ISCEV guidelines for calibration [3].

The oscillatory potential response varies with stimulus repetition rate and changes after the first stimulus. To standardize the response, we recommend that flashes be given 15 seconds apart to the dark-adapted eyes (1.5 seconds apart to light-adapted eyes), and that only the second or subsequent responses be retained or averaged. The conditions of adaptation should be reported.

*Single-flash cone response

The stimulus for the cone response is the white SF, to achieve stable and reproducible cone responses, the rods should be suppressed by a background with a luminance of 17 to 34 $cd \cdot m^{-2}$ (5 to 10 fL) measured at the surface of the full-field stimulus bowl. We recommend that the higher value of the background be chosen if the stimulus flash is at the upper end of the allowable

SF range and the lower background value chosen if the flash stimulus is at the lower end of the range. We recommend that patients light-adapt to the background luminance for at least 10 minutes before recording the cone ERG, since the cone responses may increase during this period. Stimuli should not be repeated at intervals less than 0.5 seconds. Note that the term ‘single-flash cone response’ is used to distinguish this signal from flicker responses; it does not preclude averaging (if necessary) to improve the signal-to-noise ratio.

30-Hz flicker responses

Flicker responses are to be obtained with SF stimuli, under the same rod-suppressing background illumination, after recording the single-flash cone response. Recording the flicker response in the light-adapted state reduces discomfort and facilitates the standardization of photopic adaptation. We advise strongly that flashes be presented at a rate of approximately 30 stimuli per second, and the rate that is chosen should be constant for the laboratory. The first response to the flickering stimulus will be a single-flash waveform; thus, the first few responses should be discarded so that stable conditions are reached. Some flash tubes do not produce full output while flickering, and separate calibration or a change in neutral density filtering may be needed to keep as closely as possible to the standard.

Notes

1. Prolonged-flash ERGs are currently used for studying slow potentials and for separating on- and off-responses that are outside the scope of the standard. We recognize that one can adjust the intensities of long flashes to produce response amplitudes equivalent to those produced by brief flashes, although the off components will be missing. This procedure requires careful comparison of ‘V/log I’ curves and particular care to account for off-responses and for signal attenuation by light adaptation (i.e., the interstimulus interval must be appropriately lengthened). The verification of equivalence to the standard ERG is recommended only for laboratories with special needs and expertise.

2. Chromatic stimuli offer certain advantages in the separation of cone and rod responses, but the calibration of colored stimuli and the relation of the responses produced to the standard ERG requires special procedures. We recommend that white flashes be used for the standard responses, whether or not other stimuli are used in addition.

*3 White stimuli produced by a combination of narrow band sources, such as red, green and blue light-emitting diodes (LEDs), may not be equivalent to broad-band white light as a stimulus for rods and cones. Manufacturers must

insure that appropriate photopic and scotopic filters are incorporated into their stimulation and calibration systems so that stimulus output is of equivalent intensity to the standard for all conditions. Separate scotopic calibration may be necessary for these LED systems, and if so the proper stimulus for eliciting rod responses will be 2.5 log units below a scotopically-calibrated standard flash.

4. This measurement can in practice be made with inexpensive lightmeters that integrate the flash output over time (see 'Light Adjustment and Calibration' section). Technically, the standard describes a source that delivers at the cornea the same number of quanta during the period of its flash as would be produced in 1 second by the Ganzfeld bowl when continuously illuminated by a source that produces a luminance of 1.5 to 4.5 $cd \cdot m^{-2}$.

5. We recommend that the flash source of commercial instruments be capable of generating strengths 2 log units above the SF and be attenuable through 6 log units below the SF. Regardless of whether attenuation is achieved by filters or electronic means, we also strongly recommend that commercial units incorporate a means of inserting additional colored and neutral density filters. These capabilities will allow electrophysiologists to perform a variety of useful protocols beyond the Standard, and meet possible future changes in the Standard. We also suggest that background luminance be adjustable to perform electro-oculography with the same equipment. Commercial units should also allow the insertion of colored and neutral filters into the background illumination system to meet a variety of needs.

6. DC (direct-current) amplification can produce identical responses but is extremely difficult to use because of drift in baseline and in offset potentials; we strongly advise AC recording except for laboratories with special requirements and expertise.

7. An overall index of oscillatory potential amplitude can be obtained by adding up measurements of the three major peaks, preferably from lines spanning the bases of the adjacent troughs, but alternatively from the adjacent troughs directly (to allow use of measuring cursors with digitized systems). Some authors advise measurement of individual peaks.

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Note: Printed reprints of this standard are not available, but the entire document is available on the ISCEV website <www.iscev.org>

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