Documenta Ophthalmologica **108:** 107–114, 2004.

Standard for clinical electroretinography (2004 update)*

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Key words: clinical electrophysiology, electroretinogram, standards

Abbreviations: SF – standard flash

1. Introduction

Full-field electroretinography (ERG) is a widely used ocular electrophysiologic test. In 1989 a basic protocol was standardized so that ERGs could be recorded comparably throughout the world [1]. This standard was updated most recently in 1999 [2]. Standards for five commonly obtained ERGs were presented:

(1) ERG to a weak flash (arising from the rods) in the dark-adapted eye
(2) ERG to a strong flash in the dark-adapted eye
(3) Oscillatory potentials
(4) ERG to a strong flash (arising from the cones) in the light-adapted eye
(5) ERGs to a rapidly repeated stimulus (flicker)

This document is an updated version of the standard. There are no major changes in the basic ERGs, but readers should note the intensity range of the 'standard flash (SF)' which had been printed differently in the 1999 version. An additional dark-adapted ERG to a higher-intensity stimulus is also suggested to users, as it is now being used widely and has diagnostic value. However, it has not yet been characterized sufficiently to be considered a required part of the standard. Because the stimulus for this additional ERG is brighter than the SF, we no longer use the term 'maximal' for the dark-adapted ERG to a SF.

This standard is intended as a guide to practice and to assist in interpretation of ERGs. The five basic ERGs 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 ERGs. It is 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 standard ERGs be used widely, but not to the exclusion of other ERGs or additional tests that individuals laboratories may choose or continue to use. There are also specialized types of ERG, which may provide

*This document was approved by the International Society for Clinical Electrophysiology of Vision in Nagoya, Japan on April 4, 2003.

**The authors represent the ERG Standardization Committee (Dr. Marmor, Chair) of the International Society for Clinical Electrophysiology of Vision (ISCEV).

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

Macular or focal EFG
Multifocal ERG (see published guidelines [4])
Pattern ERG (see published standard [5])
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 ERG)
Dark and light adaptation of the ERG
Stimulus intensity–response amplitude analysis (Naka-Rushton)
Saturated *a*-wave slope analysis

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], guidelines for recording the multifocal ERG [4], and standards for the pattern ERG [5], electro-oculogram [6] and visual evoked potentials [7] have also been published.

Because of the rapid rate of change of ERG techniques, this standard will be reviewed every four years. We have made recommendations that commercial recording equipment should 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
  - Light diffusion
  - Electrodes
  - Light sources
  - Light adjustment and calibration
  - Electronic recording equipment
- Clinical protocol
  - Preparation of the patient
- Specific ERGs
  - Rod ERG
  - Standard combined ERG
  - Oscillatory potential
  - Single-flash cone ERG
  - 30 Hz Flicker ERG
- ERG measurement and reporting
- Pediatric ERG recording

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 signal amplitude is reduced as the point of ocular contact moves away from the corneal apex. Topical anesthesia is necessary for contact lens electrodes but may not be required for other types of corneal and conjunctival electrodes. It is necessary that all electrophysiologists master the technical requirements of their chosen electrode, to ensure good ocular contact, to ensure

proper electrode impedance, to ensure that waveforms are comparable to standard ERGs, and to define both normal values and variability (which may be different with different electrodes) for their own laboratory. Skin electrodes are not generally 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 5 kΩ or less impedance measured between 10 and 100 Hz[3]. If more than one skin electrode is used (e.g., for reference and ground) they should all have similar impedance.

Electrode stability
The baseline voltage in the absence of light stimulation should be stable, whatever corneal and reference electrode system is used. 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 (if not disposable) be suitably cleaned and sterilized after each use to prevent transmission of infectious agents. The cleaning protocol should follow manufacturers' recommendations and current national 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.

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 can be used to enhance the separation of rod and cone ERGs, but this is not part of the standard [Note 1].

Stimulus strength–standard flash
A *standard flash* (*SF*) is defined as one of stimulus strength (in luminous energy per square meter) at the surface of the Ganzfeld bowl of 1.5–3.0 photopic $cd \cdot s \cdot m^{-2}$ (candela-seconds per meter squared) [Note 2]. This is equivalent to luminance·time, measured as $cd \cdot m^{-2} \cdot s$. Note that these are photometric units and that $3.43\ cd \cdot m^{-2} = 1$ fL (foot-Lambert).

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. A white background is used for this standard, 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 recording 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 other ERG tests will be more extensive and more stringent, and we recommend that equipment manufacturers exceed the minimum standard [Note 3].

Stimulus and background calibration
The stimulus strength (in luminance·time) produced by each flash within 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 should 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 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., capacitively coupled) and capable of handling offset potentials that may be produced by the electrodes [Note 4].

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 ERGs at a rate of 1 kHz or higher per channel. With computer-aided systems, it is important that ERG waveforms 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.

Pre-adaptation to light or dark
The recording conditions outlined below specify 20 min of dark adaptation before recording rod ERGs, and 10 min of light adaptation before recording cone ERGs. 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 min of dark adaptation may be needed for recovery after lens insertion.

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 ERGs to weak flashes before the mixed and cone ERGs to more intense flashes, to minimize light adaptation, and to reduce the time that the patient wears an 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.

Specific ERGs

Rod ERG

We recommend that the patient be dark-adapted for at least 20 min before recording the rod system ERG (and longer if the patient had been exposed to unusually bright light). The rod ERG should be the first signal measured after dark adaptation, since it is the most sensitive to light adaptation. The recommended stimulus is a dim white flash of strength 2.5 log units below the white SF (see above); we advise a minimum interval of 2 s between flashes. A blue stimulus is equally appropriate if equated to the white standard [Note 1].

Standard combined ERG

The standard ERG from combined rod and cone systems is produced by the white SF in the dark-adapted eye. We recommend an interval of at least 10 s between stimuli. This ERG is normally produced by a combination of cone and rod systems.

Higher-intensity ERG (*suggested only*)

Since publication of the last version of the ERG standard, the origins of ERG components have become better understood. It has become apparent that only the first 10–12 ms of the *a*-wave reflect photoreceptor activity. A number of laboratories have found that the dark-adapted *a*-wave is shown more clearly with the use of a brighter stimulus than the SF (approximately 10 $cd \cdot s/m^2$). Measurement of both *a*-wave amplitude and implicit time is simpler as there is generally a very well-defined single peak. There is not yet sufficient experience or universality of usage to mandate this ERG as a required part of the ERG standard, but users should be aware of its increasing acceptance and value, and consider adding it to their protocols after the standard combined ERG. An interval of 20 s is recommended between flashes of this intensity.

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 potentials vary with stimulus repetition rate and change after the first stimulus. To standardize the oscillatory potentials, we recommend that white SF stimuli be given 15 s apart to the dark-adapted eyes (1.5 s apart to light-adapted eyes), and that only the second or subsequent waveforms be retained or averaged. The conditions of adaptation should be reported.

Single-flash cone ERG

We propose the white SF as the stimulus, and advise that to achieve stable and reproducible cone system ERGs the rods be suppressed by a background with a

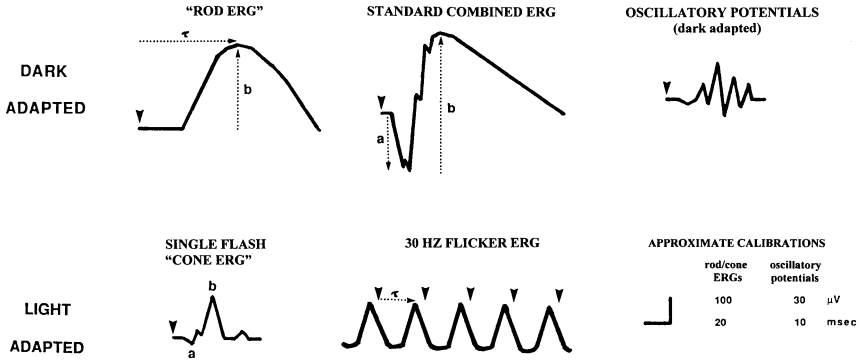

Figure 1. Diagram of the five basic ERGs defined by the Standard. These waveforms are exemplary only, and are not intended to indicate minimum, maximum or even average values. *Large arrowheads* indicate the stimulus flash. *Dotted arrows* exemplify how to measure time-to-peak (*t*, implicit time), *a*-wave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

luminance of 17 to 34 cd·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 min before recording the cone ERG, since the cone ERGs may increase during this period. Stimuli should not be repeated at intervals less than 0.5 s. Note that the term 'single-flash cone ERG' is used to distinguish this signal from flicker ERGs; it does not preclude averaging (if necessary) to improve the signal-to-noise ratio.

30-Hz flicker ERGs

Flicker ERGs also represent the cone system, and should be obtained with SF stimuli, under the same conditions of light-adaptation as the single-flash cone ERG. Recording the flicker ERG in the light-adapted state reduces discomfort and allows the photopic adaptation to be standardized. 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 ERG to the flickering stimulus will be a single-flash waveform; thus, the first few waveforms 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.

ERG measurements and recording

Measurement of the ERG

In general, *b*-wave amplitude and time-to-peak (implicit time) should be measured for all ERGs (except oscillatory potentials), and the *a*-wave should also be measured when recognizable as a distinct component. 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 5]. 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 ERGs with the recommended types of electrodes. Averaging a limited number of ERGs may decrease variability and help to reduce background noise if present. Averaging may also be used to identify and measure very weak pathologic ERGs. Artifact

rejection must be a part of any averaging system. Signal repetition rates should not exceed the recommendations in the standard for each type of ERG.

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 ERGs) 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 user's 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 ERGs (in other words, the 95% confidence limits determined by direct tabulation of ERGs). Although circadian variations of the ERG appear to be small under ordinary recording conditions, we recommend that the time of ERG recording be noted on all records since it could become relevant for certain diseases or repeat measurements.

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 ERGs 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 ERG 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 normal values and their variances (that must be provided on all reports). Finally, reports should note the time of testing, pupillary diameter, and 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. 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 the ERG.

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 that create difficulty with 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

The ERG matures during infancy, and newborn and infant signals must be interpreted with great caution. Later infantile and young childhood ERGs approach adult waveform and size. Pediatric ERGs 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 repetitions of each ERG be recorded in order to recognize reproducible waveforms and choose the best examples. Standard protocols may occasionally need to be abbreviated in order to obtain the ERGs most critical to the diagnostic question under investigation. More intense stimuli may sometimes help to reveal poorly developed ERGs. Reports should note the degree of cooperation and any medications used.

Notes

1. Chromatic stimuli offer certain advantages in the separation of cone and rod ERGs, but the calibration of colored stimuli and the relation of the ERGs produced by them to the standard ERG requires special procedures. We recommend that white flashes be used for the standard ERGs, whether or not other stimuli are used in addition.
2. 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 ensure 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 ERGs will be 2.5 log units below a scotopically-calibrated standard flash.
3. We recommend that the flash source of commercial instruments be capable of generating strengths at least 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.
4. DC (direct-current) amplification can produce signals identical to those from AC amplification, but it is extremely difficult to use because of drift in baseline and offset potentials; we strongly advise AC recording except for laboratories with special requirements and expertise.
5. 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 document is available on the ISCEV website <www.iscev.org>

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