RET*eval* Complete Option

The **RET***eval* Complete option makes the **RET***eval* device a full-featured, ISCEV standard compliant (Marmor, et al. 2009) ERG device. The DR Assessment protocol and the protocols in the Flicker ERG option provide quick results for a number of diseases that can be assessed via cone responses. Nevertheless, there are many other diseases for which a rod assessment and single-flash assessments provide valuable insight into the state of the visual system. These protocols will take significantly longer to perform due to the dark adaption periods required to assess rod function.

Additionally, a protocol is provided for ISCEV-compliant flash VEP testing (Odom, et al. 2010).

The ISCEV standard full field ERG measurements have been useful for a number of diseases. Textbooks have been written (Heckenlively and Arden 2006), (Fishman, et al. 2001) as well as a journal (*Documenta Ophthalmologica*) dedicated to clinical electrophysiology of vision.

Through a protocol chooser, the test protocol can be selected from single-flash options in addition to the flicker options and the protocol specifically designed for vision-threatening diabetic retinopathy.

By purchasing an adapter cable for DIN electrodes (part number 91-194), you can use any 1.5 mm safety DIN electrode with the **RET***eval* device. Chapter 17 in (Heckenlively and Arden 2006) enumerates many electrodes that are acceptable for ERG recordings. Refer to the documentation provided by the electrode manufacturer and in the ISCEV standards for proper placement, skin preparation, cleaning, and disposal of these DIN electrodes. When performing a test, the **RET***eval* device will prompt the operator to specify the electrode type. This information will be stored in the results and appropriate normative data (when available) will be displayed. The red lead is the positive connection, the black lead in the negative connection, and the green lead is the ground / right leg drive connection.

The ERG signal amplitude is lower with skin-contact electrodes such as Sensor strips than with corneal-contact electrodes. For ERGs recorded with the active electrode on the skin, signal averaging is used. Skin electrodes may not be suitable to evaluate attenuated pathologic electroretinograms. It is recommended that users recording electroretinograms should master the technical requirements of their chosen electrode to obtain good contact, consistent electrode positioning and acceptable electrode impedance.

RET*eval* *Complete protocols*

The **RET***eval* device supports single-flash and flicker ERG testing. Brief flashes of light are provided at the beginning of each stimulus period. A background light is also generated by providing brief flashes of light at 283 Hz, which is well above the human critical fusion frequency and therefore is perceived as steady illumination. These protocols provide dark adaptation timers as well as an approximate ambient light level during the dark adaption. The ambient light level is approximated by taking the geometric mean of the light level measured inside the integrating sphere (ganzfeld) by a photodiode with an ambient light optical filter bonded onto it.

Many of the protocols have constant retinal illuminance, which are described by the Troland unit (Td). These protocols are identified with "Td" in the user interface and PDF reports. In these protocols, the **RETeval** device measures the pupil size in real time and continuously adjusts the flash luminance to deliver the desired amount of light *into* the eye regardless of the size of the pupil according to the following formula: Troland = (pupil area in mm^2) (luminance in cd/m^2). Thus, pupils do not need to be dilated to achieve consistent results. Even when using mydriatics, people dilate to different diameters and results can be made more consistent by using the Troland-based stimuli. While Troland-based tests make results less dependent on pupil size, secondary factors such as the Stiles-Crawford effect and/or changes in the distribution of light on the retina prevent Troland-based tests from being completely independent of pupil size. The built-in ISCEV Troland protocols attempt to match the ISCEV candela protocols by assuming a 6 mm pupil diameter (28.3 mm^2 pupil area). For example, the Troland equivalent to the dark adapted 3.0 ERG, which has a flash luminance of 3 $cd{\cdot}s/m^2$, has a stimulus of (3 $cd{\cdot}s/m^2$)(28.3 mm^2) = 85 Td·s. If the pupil diameter is 6 mm, the 85 Td·s stimulus will be the same as a 3 $cd{\cdot}s/m^2$ stimulus and the resulting ERGs will therefore be the same.

There are cases where the stimulus compensating for pupil size may be inconvenient. These protocols are identified with "cd" in the user interface and PDF reports. For example, the patient cannot keep their eyelids sufficiently open for the device to measure the pupil, there is a desire to stimulate the eye through a closed eyelid, or there is a desire to match the stimulus of a previous publication. When looking for the presence of any retinal function, a bright constant luminance stimulus may be sufficient.

Subtests in these protocols display the waveform results after each measurement period and enable the operator to repeat the step as many times as desired. Automated cursor placements are computed to the average cursor placement across all repetitions. Any subtest can be skipped without affecting the rest of the protocol.

For the dark adapted 0.1 Hz 85 Td·s and 3 $cd{\cdot}s/m^2$ tests, oscillatory potentials and cursors are reported. The oscillatory potential waveform is obtained by applying an 85 Hz – 190 Hz bandpass filter. Up to 5 cursors are automatically placed on the oscillatory potential peaks and troughs and are indicated on the report as black dots on the waveform. Implicit times (time to peak) and amplitudes (peak to following trough) are reported for each individual cursor. The sums of implicit times and amplitudes for all cursors are also reported. When interpreting the summed cursor times and amplitudes, you should examine the cursor dots on the waveform to ensure that no waves are missed.

For dark adapted tests, the display is automatically dimmed and reddened. The green power status LED is also turned off to assist in dark adaptation. The display and LED are automatically brightened at the end of the dark adaptation tests.

To create the visual stimulus, the **RETeval** device generates variable-duration flashes of white light, made from red, green, and blue LEDs all being on for the same duration. The maximum energy flash of white light is 30 $cd{\cdot}s/m^2$, which has a flash duration of 5 ms. For the constant Troland tests, the flash duration may be longer than 5 ms for pupil sizes smaller than 1.9 mm. Modeling of the 3 stage activation phase of phototransduction, as described by (Cideciyan and Jacobson 1996) in equation A5, shows very small differences in rod or cone photocurrent between having an instantaneous flash and flash energies uniformly spread into flash durations as long as

10 ms as long as all measurements are considered relative to the center of the flash, as done by the **RET*eval*** device. If the pupil size is sufficiently small that the required flash energy for a Troland protocol is not obtainable, the **RET*eval*** device will produce its maximum flash energy.

The signal processing for the non-flicker tests uses the followings steps. A zero-phase 0.3 Hz high-pass filter reduces electrode drift and offset while preserving waveform timing. Measurements from multiple flashes are combined to improve the signal to noise ratio using a trimmed mean to reduce the effect of outliers after removing outlier replicates whose amplitudes exceed 1 mV. The resulting waveform is then processed using wavelet-based denoising (Ahmadi and Rodrigo 2013) where wavelets are attenuated based on the signal to noise power between the post-stimulus (signal) and pre-stimulus (noise) portions of the waveform. Oscillatory potential analysis does not use the wavelet denoising.

The number of flashes combined is specified in the tables below. If a different number of flashes is desired, a custom protocol can be created by modifying a protocol in the EMR/built-in-protocols folder and placing it in the Protocols/ folder on the device. Any text editor can be used to edit the protocol (e.g., Emacs or Notepad). Because of the relatively few flashes combined for the non-flicker tests, reducing the noise is more important in these tests; consequently, skin preparation is suggested for all patients in order to reduce the electrode contact impedance.

ERG protocols

The following tables describe the ERG protocols in detail.

This protocol (**ISCEV 6 step, light adapted first, cd**) performs the light adapted tests first, and assumes light adaptation occurs before the tests start. Some clinicians use room lights to do the light adaptation. ISCEV recommends 20 minutes of dark adaptation and 10 minutes of light adaptation.

ISCEV 6 step, light adapted first, cd				
Description	**Eye**	**Flash luminance energy (0.33, 0.33 white)**	**Background luminance (0.33, 0.33 white)**	**# flashes**
Light adapted 3.0 ERG	Right	3 cd·s/m² @ 2 Hz	30 cd/m²	30
Light adapted 3.0 flicker ERG	Right	3 cd·s/m² @ 28.3 Hz	30 cd/m²	141 – 424
Light adapted 3.0 ERG	Left	3 cd·s/m² @ 2 Hz	30 cd/m²	30
Light adapted 3.0 flicker ERG	Left	3 cd·s/m² @ 28.3 Hz	30 cd/m²	141 – 424
Dark adaptation timer	Both	Off	Off	
Dark adapted 0.01 ERG	Right	0.01 cd·s/m² @ 0.5 Hz	Off	9
Dark adapted 3.0 ERG	Right	3 cd·s/m² @ 0.1 Hz	Off	5
Dark adapted 10.0 ERG	Right	10 cd·s/m² @ 0.05 Hz	Off	5
Dark adapted 0.01 ERG	Left	0.01 cd·s/m² @ 0.5 Hz	Off	9
Dark adapted 3.0 ERG	Left	3 cd·s/m² @ 0.1 Hz	Off	5
Dark adapted 10.0 ERG	Left	10 cd·s/m² @ 0.05 Hz	Off	5

This protocol (**ISCEV 6 step, dark adapted first, cd**) switches the testing order to do the dark adapted tests first. The **RET*eval*** device performs a calibration at the beginning of every protocol. So that the calibration light flashes do not affect the dark adaptation state of the subject, place the device on the patients forehead when requested by the device. Skin color has a small, but measureable, effect on the light output (due to the skin's reflectance); thus, the test subject's forehead should be used. In this protocol, there is a light-adaptation timer for each eye to be adapted to 30 cd/m^2. ISCEV recommends 20 minutes of dark adaptation and 10 minutes of light adaptation.

ISCEV 6 step, dark adapted first, cd				
Description	**Eye**	**Flash luminance energy (0.33, 0.33 white)**	**Background luminance (0.33, 0.33 white)**	**# flashes**
Dark adaptation timer	Both	Off	Off	
Dark adapted 0.01 ERG	Right	0.01 cd·s/m² @ 0.5 Hz	Off	9
Dark adapted 3.0 ERG	Right	3 cd·s/m² @ 0.1 Hz	Off	5
Dark adapted 10.0 ERG	Right	10 cd·s/m² @ 0.05 Hz	Off	5
Dark adapted 0.01 ERG	Left	0.01 cd·s/m² @ 0.5 Hz	Off	9
Dark adapted 3.0 ERG	Left	3 cd·s/m² @ 0.1 Hz	Off	5
Dark adapted 10.0 ERG	Left	10 cd·s/m² @ 0.05 Hz	Off	5
Light adaptation timer	Right	Off	30 cd/m²	
Light adapted 3.0 ERG	Right	3 cd·s/m² @ 2 Hz	30 cd/m²	30
Light adapted 3.0 flicker ERG	Right	3 cd·s/m² @ 28.3 Hz	30 cd/m²	141 – 424
Light adaptation timer	Left	Off	30 cd/m²	
Light adapted 3.0 ERG	Left	3 cd·s/m² @ 2 Hz	30 cd/m²	30
Light adapted 3.0 flicker ERG	Left	3 cd·s/m² @ 28.3 Hz	30 cd/m²	141 – 424

The next two protocols are the same as the previous two with the exception that the 10 $cd{\cdot}s/m^2$ white flash is not performed.

ISCEV 5 step, light adapted first, cd				
Description	**Eye**	**Flash luminance energy (0.33, 0.33 white)**	**Background luminance (0.33, 0.33 white)**	**# flashes**
Light adapted 3.0 ERG	Right	3 cd·s/m² @ 2 Hz	30 cd/m²	30
Light adapted 3.0 flicker ERG	Right	3 cd·s/m² @ 28.3 Hz	30 cd/m²	141 – 424
Light adapted 3.0 ERG	Left	3 cd·s/m² @ 2 Hz	30 cd/m²	30
Light adapted 3.0 flicker ERG	Left	3 cd·s/m² @ 28.3 Hz	30 cd/m²	141 – 424
Dark adaptation timer	Both	Off	Off	
Dark adapted 0.01 ERG	Right	0.01 cd·s/m² @ 0.5 Hz	Off	9
Dark adapted 3.0 ERG	Right	3 cd·s/m² @ 0.1 Hz	Off	5
Dark adapted 0.01 ERG	Left	0.01 cd·s/m² @ 0.5 Hz	Off	9
Dark adapted 3.0 ERG	Left	3 cd·s/m² @ 0.1 Hz	Off	5

ISCEV 5 step, dark adapted first, cd				
Description	**Eye**	**Flash luminance energy (0.33, 0.33 white)**	**Background luminance (0.33, 0.33 white)**	**# flashes**
Dark adaptation timer	Both	Off	Off	
Dark adapted 0.01 ERG	Right	0.01 cd·s/m² @ 0.5 Hz	Off	9
Dark adapted 3.0 ERG	Right	3 cd·s/m² @ 0.1 Hz	Off	5
Dark adapted 0.01 ERG	Left	0.01 cd·s/m² @ 0.5 Hz	Off	9
Dark adapted 3.0 ERG	Left	3 cd·s/m² @ 0.1 Hz	Off	5
Light adaptation timer	Right	Off	30 cd/m²	
Light adapted 3.0 ERG	Right	3 cd·s/m² @ 2 Hz	30 cd/m²	30
Light adapted 3.0 flicker ERG	Right	3 cd·s/m² @ 28.3 Hz	30 cd/m²	141 – 424
Light adaptation timer	Left	Off	30 cd/m²	
Light adapted 3.0 ERG	Left	3 cd·s/m² @ 2 Hz	30 cd/m²	30
Light adapted 3.0 flicker ERG	Left	3 cd·s/m² @ 28.3 Hz	30 cd/m²	141 – 424

The next two protocols are similar to the ISCEV 5 step protocols above, except pupil-tracking is used to provide constant retinal illuminance, making pupil dilation optional. A 6 mm pupil was assumed to convert the ISCEV standard dilated luminances to Trolands.

ISCEV 5 step, light adapted first, Td				
Description	**Eye**	**Flash luminance energy (0.33, 0.33 white)**	**Background luminance (0.33, 0.33 white)**	**# flashes**
Light adapted 85 Td·s ERG	Right	85 Td·s @ 2 Hz	848 Td	30
Light adapted 85 Td·s flicker ERG	Right	85 Td·s @ 28.3 Hz	848 Td	141 – 424
Light adapted 85 Td·s ERG	Left	85 Td·s @ 2 Hz	848 Td	30
Light adapted 85 Td·s flicker ERG	Left	85 Td·s @ 28.3 Hz	848 Td	141 – 424
Dark adaptation timer	Both	Off	Off	
Dark adapted 0.28 Td·s ERG	Right	0.28 Td·s @ 0.5 Hz	Off	9
Dark adapted 85 Td·s ERG	Right	85 Td·s @ 0.1 Hz	Off	5
Dark adapted 0.28 Td·s ERG	Left	0.28 Td·s @ 0.5 Hz	Off	9
Dark adapted 85 Td·s ERG	Left	85 Td·s @ 0.1 Hz	Off	5

ISCEV 5 step, dark adapted first, Td				
Description	**Eye**	**Flash luminance energy (0.33, 0.33 white)**	**Background luminance (0.33, 0.33 white)**	**# flashes**
Dark adaptation timer	Both	Off	Off	
Dark adapted 0.28 Td·s ERG	Right	0.28 Td·s @ 0.5 Hz	Off	9
Dark adapted 85 Td·s ERG	Right	85 Td·s @ 0.1 Hz	Off	5
Dark adapted 0.28 Td·s ERG	Left	0.28 Td·s @ 0.5 Hz	Off	9
Dark adapted 85 Td·s ERG	Left	85 Td·s @ 0.1 Hz	Off	5
Light adaptation timer	Right	Off	848 Td	
Light adapted 85 Td·s ERG	Right	85 Td·s @ 2 Hz	848 Td	30
Light adapted 85 Td·s flicker ERG	Right	85 Td·s @ 28.3 Hz	848 Td	141 – 424
Light adaptation timer	Left	Off	848 Td	
Light adapted 85 Td·s ERG	Left	85 Td·s @ 2 Hz	848 Td	30
Light adapted 85 Td·s flicker ERG	Left	85 Td·s @ 28.3 Hz	848 Td	141 – 424

VEP protocols

Flash VEP protocols flash light in the eye and measure the visual system's response on the back of the head. There are two flash VEP protocols: a 3 cd·s/m^2 @ 1 Hz protocol and a 24 Td·s @ 1 Hz. The two protocols are equivalent when the pupil diameter is 3.2 mm (8 mm^2 area). Both use 64 flashes to average the response. These protocols require gold cup or other surface electrodes placed according to the ISCEV standard (Odom, et al. 2010). In order to perform these VEP tests, the adapting cable 91-194 must be purchased in order to connect your DIN electrodes to the **RET*eval*** device. Flash VEP measurements depend on the response from the retina being transmitted through the optic nerve to the occipital cortex and therefore can be used as an indicator of visual function. Flash VEP measurements are highly variable among individuals, but are fairly repeatable for one individual. Running replicates, which is an option in these tests, can help distinguish the evoked response from other biological signals.

Custom protocols

If there is a protocol that you would like to run that is not built-in, the **RET*eval*** device has support for extending the number of options through custom protocols. An exemplary custom protocol would be to perform replicates of a stimulus in order to assess test precision. Contact LKC (email: info@lkc.com) if you would like a custom protocol.

Custom protocols can be placed in the Protocols folder on the device. The built-in protocols can be viewed on the device in the folder EMR/built-in protocols, which can be a starting point for creating your own custom protocols. Protocols are written in the full-featured Lua programming language.

RET*eval* *Complete test results*

Incremental results are shown on the **RET*eval*** device after each test, with the option to repeat the test or continue to the next test. Successful cursor placement is indicated by dashed lines on the waveform indicating their location. If you do not see the successful cursor placement indication, repeat the measurement.

Historical results can be seen from the main menu **Results** option. Scroll up and down through the list and select the desired test result. The results are stored in chronological order; with the most recent result first. The results include the stimulus, electrical amplitudes, timings, and waveforms recorded by the Sensor strips for each eye for each step in the protocol. The graphs display the average cursor placements. A flash occurs at time = 0 for all tests. When referring to the literature for clinical interpretation, it is important that your testing be done at the same flash intensity and background light level. The ISCEV standard states that each laboratory should establish or confirm typical reference values for its own equipment, recording protocols, and patient populations.

An example PDF report for the **ISCEV 5 step, light adapted first, Td** protocol is shown below.

Test Protocol: ISCEV 5 Step Light First Td

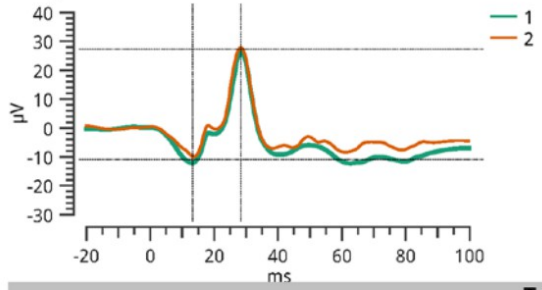
Electrodes: Sensor strips

Test #1

Flash: 85 Td·s, Chromaticity (0.33, 0.33) at 2 Hz

Right Eye

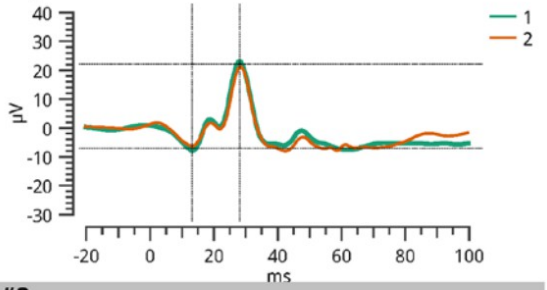
Trial	a-wave ms	a-wave µV	b-wave ms	b-wave µV
1	13.0	-12.1	28.4	38.9
2	13.5	-9.9	28.4	37.7
Avg	13.3	-11.0	28.4	38.3



Background: 848 Td, Chromaticity (0.33, 0.33)

Left Eye

Trial	a-wave ms	a-wave µV	b-wave ms	b-wave µV
1	13.2	-7.9	28.0	31.0
2	13.1	-6.2	28.3	27.5
Avg	13.2	-7.0	28.2	29.2

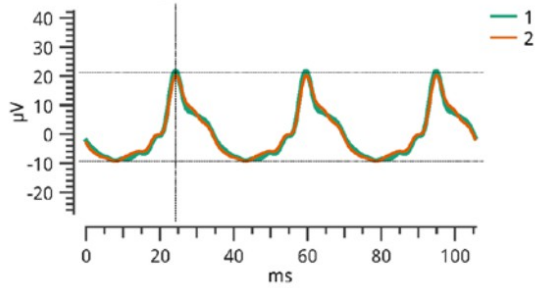


Test #2

Flash: 85 Td·s, Chromaticity (0.33, 0.33) at 28.3 Hz

Right Eye

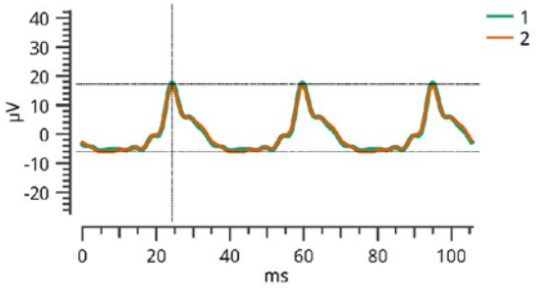
Trial	ms	µV
1	24.2	31.0
2	24.4	29.7
Avg	24.3	30.3



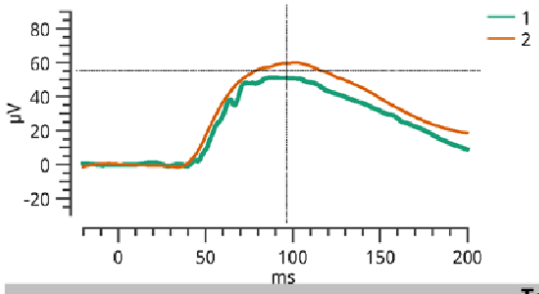
Background: 848 Td, Chromaticity (0.33, 0.33)

Left Eye

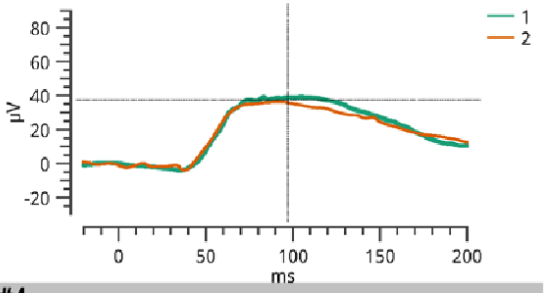
Trial	ms	µV
1	24.3	23.5
2	24.2	22.6
Avg	24.3	23.1



2	99.4	59.9	
Avg	96.3	55.5	



2	90.6	36.3	
Avg	97.1	37.7	



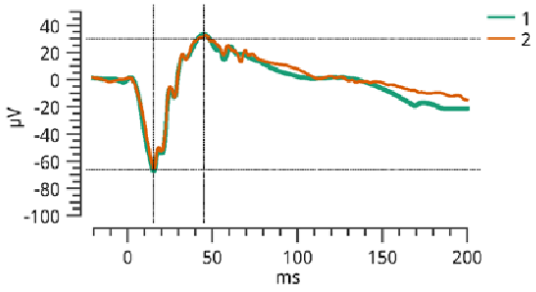
Test #4

Flash: 85 Td·s, Chromaticity (0.33, 0.33) at 0.1 Hz

Background: 0 Td

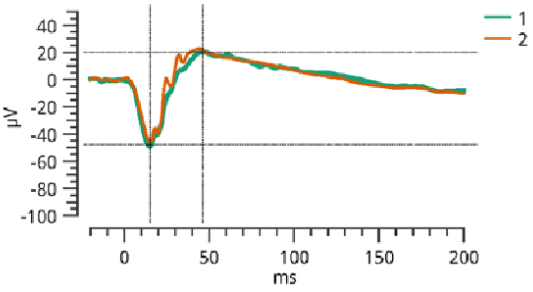
Right Eye

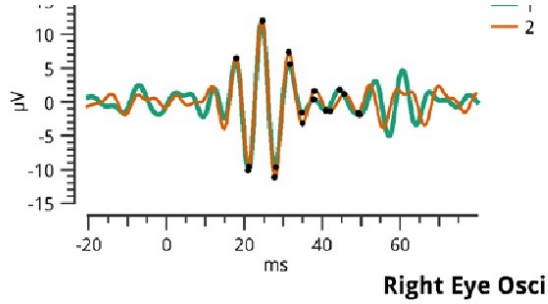
Trial	a-wave ms	µV	b-wave ms	µV
1	15.3	-67.4	44.6	97.2
2	15.4	-65.3	45.1	95.5
Avg	15.4	-66.4	44.8	96.3

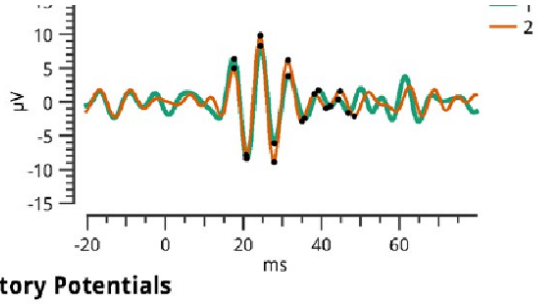


Left Eye

Trial	a-wave ms	µV	b-wave ms	µV
1	14.8	-49.3	48.4	68.5
2	15.2	-46.1	44.0	67.4
Avg	15.0	-47.7	46.2	68.0







Right Eye Oscillatory Potentials

	OP1		OP2		OP3		OP4		OP5	
Trial	ms	µV	ms	µV	ms	µV	ms	µV	ms	µV
1	17.9	16.6	24.5	21.7	31.7	7.2	37.8	1.6	44.5	3.8
2	18.0	16.1	24.7	23.2	31.4	10.6	38.0	2.9	45.7	2.8

Left Eye Oscillatory Potentials

	OP1		OP2		OP3		OP4		OP5	
Trial	ms	µV	ms	µV	ms	µV	ms	µV	ms	µV
1	17.7	14.7	24.4	14.4	31.5	6.3	39.2	2.4	44.3	2.0
2	17.6	12.9	24.3	18.8	31.5	9.1	38.1	2.1	44.8	3.7

An example flash VEP report is shown below. To make this recording, an adapting cable (LKC part number 91-194, **RET***eval* adapter cable for DIN electrodes) was used in lieu of the Sensor strip lead. A gold cup recording electrode (LKC part number 95-018) was placed 2 cm above the inion. The negative (reference) electrode was placed on the center of the forehead, just below the hairline (Ag/AgCl electrode, LKC part number 95-005). The ground / right leg drive electrode was a gold cup ear clip (LKC part number 95-004). For all the electrodes, the skin was prepared using alcohol pad containing a mild abrasive (LKC part number 95-017). The recording electrode was filled with electrode cream (LKC part number 95-006) which both holds the electrode in place and makes the electrical connection. Electrode gel (LKC part number 95-007) was used on the ear clip to ensure a low impedance connection.

Test Protocol: ISCEV Flash VEP: 24 Td·s

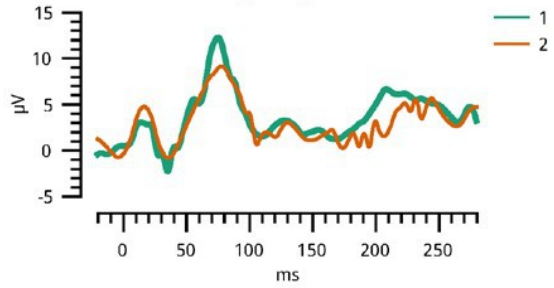
Electrodes: Gold cup

Test Results

Flash: 24 Td·s, Chromaticity (0.33, 0.33) at 1 Hz

Background: 0 Td

Right Eye



Left Eye