**TITLE:**

Recording Horizontal Saccade Performances Accurately in Neurological Patients Using Electro-oculogram

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**SHORT ABSTRACT:**

The article describes a practical method for recording horizontal eye movements with high accuracy by electro-oculogram in neurological patients, using a cup Ag-AgCl electrode with a wide plastic fringe. Stable measurement requires proper selection and fixation of electrodes, taking sufficient time for light adaptation to occur, and re-calibration as needed.

**LONG ABSTRACT:**

Electro-oculogram (EOG) has been widely used for clinical eye movement recording, especially horizontal saccades, although the video-oculography (VOG) has largely taken the place of it nowadays due to its higher spatial accuracy. However, there are situations in which EOG has clear advantages over VOG, *e.g.,* subjects with narrow eye clefts or having cataract lenses, and patients with movement disorders. The present article shows that if properly implemented, EOG can achieve an accuracy almost as good as VOG with substantial stability for recording, while circumventing problems associated with VOG recording. The present paper describes a practical method for recording horizontal saccades using oculomotor paradigms with high accuracy and stability by EOG in neurological patients. The necessary measures are to use an Ag-AgCl electrode with a wide plastic fringe capable of reducing noise, and to wait for sufficient light adaptation to occur. This waiting period also helps to lower the impedance between the electrodes and the skin, thereby ensuring stable signal recorded as time goes by. Furthermore, re-calibration is performed as needed during the task performance. Using this method, the experimenter can avoid drifts of signals, as well as contamination of artifacts or noise from the electromyogram and electroencephalogram, and can collect sufficient data for clinical evaluation of saccades. Thus when implemented, EOG can still be a method of high practicability that can be widely applied to neurological patients, but may be effective also for studies in normal subjects.

**INTRODUCTION:**

There are three major ways to record eye movements, the conventional EOG, the VOG recorded by the video-based eye tracking system, and the scleral search coil (SSC) method. Among them, EOG has been frequently used for recording eye movements in patients since the 1970s because of its simplicity. Widely applicable to the clinical population, this method has been extensively utilized for the diagnosis of neurological patients and has provided useful information about the pathophysiology underlying the disorders1-5. In addition, it is still the only technique that can be feasibly used for recording eye movements during sleep (rapid eye movement during REM sleep and other forms of eye movements).

Since the eyeball is positively charged in its anterior aspect including the cornea relative to its posterior aspect, there is a voltage difference between the anterior and posterior aspects of the eyes termed the corneo-retinal potential. Due to the presence of this potential, the right electrode will become more positive than the left when the subjects turns their gaze toward the right, and become negative when they turn their gaze to the left. Since the voltage difference between the left and right electrodes correlates significantly with the rotation angle of the eyeballs for horizontal saccades, it can be used to measure horizontal eye movements. However, this correlation does not hold for the vertical direction, although vertical EOG still can be used to measure eye movements6. On the other hand, some studies mainly use vertical EOG for monitoring blinks.

Recently, however, VOG has largely taken the place of EOG due to its higher spatial accuracy reaching up to 0.25–0.5 degrees, and has now become the standard method for saccade recording in the clinical setting. Meanwhile, EOG has come to be considered rather outdated, since its spatial accuracy, at most 0.5 degrees, is inferior to that of VOG.

However, VOG also has its own drawbacks if used in the clinical setting. There are cases in which VOG is not feasible; for example, eye tracking becomes inaccurate in subjects with a narrow eye cleft such as when the greater area of the cornea is occluded by the eyelids. In patients with cataract lenses, aberrant reflection of the infrared light hampers reliable recording of the gaze direction. Furthermore, EOG can offer advantages for some people for whom their movement disorder makes VOG recording difficult. In addition, the VOG system is more expensive compared to the setup of EOG, which often makes the former unavailable in ordinary medical facilities.

On the other hand, the SSC method is regarded as the gold standard for measuring eye movements. Compared with VOG and EOG, this method provides the highest spatial accuracy, down to 0.1 degrees, and is especially useful when the recording involves high-frequency head motion6. However, this method is potentially invasive, *i.e.*, painful and very irritating to the eyes, and allows recording for only a brief period, approximately under 30 min or shorter7-10. This short duration makes it a method unsuitable for extensive clinical application, although it has been used successfully in some specialized facilities11.

Based on previous studies recording more than 250 neurological patients and 480 normal subjects by the same group12-19, the present study shows that EOG can be accurate enough to serve as a standard technique of eye movement recording, and widely applicable to the clinical population, while circumventing various drawbacks of VOG and SSC. The present article describes a stable EOG recording method, using an electrode with a wide fringe to allow wide and stable contact with the skin, similar to that of an EEG electrode attached securely on the scalp by collodion for recording a long time period. The impedance of the electrode goes down and the recording becomes stable with time, thereby effectively reducing the artifacts from facial muscles and electroencephalography. This method is compared with simultaneously recorded VOG. When properly prepared and implemented, EOG is as good as VOG in terms of accuracy for recording saccades in neurological patients, and EOG may even be more amenable to saccade recording in normal subjects.

**PROTOCOL:**

All experimental procedures in this study were approved and conducted according to the guidelines of the institution’s human research ethics committee after obtaining informed consent.

1. **Prepare the Subject and the Room for Recoding**
   1. Perform recording in a room with low ambient illumination, to allow sufficient light adaptation.
   2. Have subjects sit in front of a black, concave dome-shaped screen measuring 90-cm in diameter that contains light-emitting diodes (LEDs) embedded in pinholes, which serve as the fixation points and saccade targets used for the oculomotor paradigms.

Note: The LEDs are arranged in horizontal, vertical and oblique arrays in a black, concave dome-shaped screen, *i.e.*, in 8 directions separated by 45 degrees from the center and at an interval of 5 degrees from the center, as was originally devised by Kato *et al*.20 for behavioral and physiological studies, and modified for human use by Hikosaka et al.21

* 1. For controlling the oculomotor tasks, make each subject hold a microswitch button connected to a microcomputer, which allows the subject to initiate and terminate a task trial by pressing and releasing the button.

Note: Tasks and data acquisition are controlled by a custom-made program operating on a typical Windows PC.

* 1. Stabilize the subject’s head position by chin and forehead rests, as well as by a head band.

1. **Place the Electrodes for EOG**
   1. Use an Ag-AgCl cup electrode for recording EOG (**Figure 1**), which has a diameter of 1.8 cm and a thickness of 3.5 mm. The bottom of the cup comprises the Ag-AgCl electrode and the lateral wall is surrounded by a plastic fringe of 5 mm thickness, enabling wide contact with the skin.
   2. Wipe the skin with an alcohol swab.
   3. Fill the cup with electrode paste.
   4. Stably fixate the electrode on the skin by placing double-stick adhesive tape beneath the plastic and attach the fringe to the skin.
   5. For recording horizontal saccades by EOG, place the electrodes at the bilateral canthi of the eyes, whereas for recording vertical saccades, place the electrodes above and below one eye.
2. **Set Up the Amplifiers for Recording**
   1. Use a direct current (DC)-amplifier for recording the EOG, with the signal digitized at 500 Hz.
   2. Record VOG simultaneously, using the video-based eye tracking system, which records ocular fixation position data at a sampling rate of 500–1,000 Hz.
   3. Feed the analog output of the horizontal and vertical eye positions and set the filter of the data acquisition system, with the signal low-pass filter at 20 Hz.
   4. Also, set the filter to attenuate the high-intermediate frequency noise, such as electromyography and electroencephalography.

Note: For analysis, a further smoothing process is necessary for calculating saccade velocity profiles from the eye position data (here, 3-point smoothing was performed three times).

* 1. If possible, measure the impedance between the electrode and the skin and keep it below 20 kΩ.

1. **Waiting Period after Placing the Electrodes for Light Adaptation**
   1. Wait 10–20 min after placing the EOG electrode on the skin, until sufficient light adaptation takes place.
   2. Allow the recording to stabilize and the impedance between the electrode gel to decrease.

1. **Calibrate the EOG and VOG Signals**
   1. Perform eye movement calibration before each test session by having the subjects look at 5 pre-specified locations.
   2. More specifically, make subjects view visual targets in the center and those that appear 20 degrees to the left, right, upwards, and downwards of the fixation point, both for EOG and VOG.
   3. Adjust the gain of EOG as the subjects fixate on these spots, so that using the custom-built data acquisition system for monitoring the current eye position displayed on the computer screen matches the target position displayed on the screen.
2. **Record Saccades Using the Oculomotor Paradigms and Recalibrate the Eye Positions as Needed During the Session**
   1. Instruct the subjects about the oculomotor paradigms.

Note: Two oculomotor tasks are typically used for clinical studies, the visually guided saccade (VGS) and antisaccade (AS) tasks. Briefly, in VGS, when subjects press the button, a central spot is lit in the center of the dome, and the subjects are first required to fixate on this spot. 1.5–2 s later, a target is presented, randomly at a location 5, 10, 20, or 30 degrees horizontally to the left or right of it, at the same time as the central fixation spot is extinguished. The subjects are instructed to make a saccade toward that target. In the AS task, make the subjects first press the button and then require them to fixate on the central fixation point as it appears. 1.5–2 s later, the target jumps to the left or right of it, similar to above. The subjects are required to make a saccade towards a mirror-symmetrical position across the central fixation spot.

* 1. Have the subjects press the button and begin the trials.
  2. During the session, adjust the gain of EOG during the task performance, so that the current eye position displayed on the monitor is always aligned with the target position simultaneously displayed on the same screen. Both for EOG and VOG, perform re-calibration for adjustment when necessary throughout the experiments.
  3. To compare the performances of the two methodologies, analyze the filtered and digitized EOG signals from the DC-amplifier and VOG by a custom-built computer program and show the EOG and VOG signals together in the same trace.

**REPRESENTATIVE RESULTS:**

**Figure 2** shows representative simultaneous records of EOG and VOG in a normal subject. 8 trials of VGS are superimposed for EOG (gray curves) and VOG (red curves; **Figure 2A**). Calibrated by the present method, EOG and VOG data are known to be linear over a range of 5–30 degrees, and the spatial accuracy of the data is 0.5 degrees.

The records obtained by the two methods largely overlap with each other. Also, the saccade parameters, such as latency and amplitude, are almost comparable for the two recording methods, although the velocity is slightly smaller for EOG (VGS: **Figure 2B**, AS: **Figure 2C**).

For EOG, electromyography and electroencephalography can confound eye movement records, which usually necessitates the use of low-pass filtering for proper eye recording. The use of low-pass filtering at 20 Hz has been reported to decrease the peak velocity, and to increase the onset of saccades slightly; the velocity of saccades is smaller by up to 10% for VGS and MGS, and the latency measured by EOG is longer than that measured by VOG by 2–3% (or on the order of 8–10 ms), whereas the amplitude of saccades is largely comparable22. On the other hand, previous studies by other groups have reported larger saccade velocity for EOG as compared to both VOG and SSC7-10, and the discrepancy between studies was considered due to the use of a smoothing procedure to calculate the saccade velocity profile and the low-pass filtering as mentioned.

**FIGURE LEGENDS:**

**Figure 1: The electrode and adhesive tape used for recording EOG.** The electrode has a diameter of 1.8 cm and a thickness of 3.5 mm, where the bottom comprises the Ag-AgCl electrode and the lateral wall is surrounded by a plastic fringe of 5 mm thickness. This enables wide contact with the skin, allowing close and stable fixation, and serves to reduce the impedance between the skin and the electrode. Due to this, the recording becomes stable with time, thereby effectively reducing the artifacts from facial muscles and electroencephalography.

**Figure 2: Representative traces of saccades recorded by EOG and VOG.** (**A**) Representative traces recorded by EOG and VOG. 8 VGS traces are superimposed, time-locked to the signal instructing the start of saccades. Saccades towards targets of four different eccentricities (5, 10, 20, and 30 degrees) to the left and right from the central fixation spot are recorded. The horizontal axis gives the time and the vertical axis gives the eye position (upper trace) or velocity (lower traces). The red curves are for VOG and the gray curves are for EOG. Ticks below are marked at a 100 ms interval. Gray curves are for EOG measured by the DC amplifier, and the red curves are for VOG measured by the video-based eye tracking system. When the eyes move to the right, the traces deflect upwards and when the eyes move to the left, they deflect downwards. Note the substantial overlap between the traces, except that the gray traces (EOG) are slightly displaced towards the right compared to the red traces (VOG), implying a slightly longer latency for EOG relative to VOG. (**B**) Comparison of EOG and VOG traces in the VGS task. Red curves are for EOG and black and blue curves are for VOG of the left and right eyes, respectively. The upper trace is for eye position, and the lower figure is for eye velocity. Again note the substantial overlap between the traces for EOG and VOG, but the latency is slightly longer for EOG, and the velocity curve of EOG shows a slightly lower peak velocity than that of VOG. (**C**) Comparison of EOG and VOG traces in the AS task. Similar EOG and VOG traces when subjects perform the AS task. Note again the substantial overlap and that the latency is slightly longer for EOG, and the velocity curve for EOG shows a slightly lower peak velocity than that for VOG.

**DISCUSSION:**

Although nowadays, the prevailing method for recording saccades has become the VOG, the present study showed that EOG can achieve an accuracy almost comparable to that of VOG if properly implemented (**Figure 2**). The present EOG method has been shown to achieve a good correlation with VOG when recording horizontal saccades and has been successfully used in many previous studies by the same group12-19.

Admittedly, VOG has a higher spatial accuracy than EOG and has largely replaced EOG in the clinical setting, but the higher accuracy of VOG and SSC should not always be taken at face value. EOG has been recorded in combination with VOG or SSC, and has shown performance comparable to the latter two despite small differences7-9. Comparison of saccade peak velocities simultaneously measured by EOG, VOG, and SSC consistently showed that peak velocity measured by EOG is slightly but consistently faster than those measured by the other two methods7-9. This faster velocity measured by EOG is generally ascribed to the greater noise level for EOG recording, such as contamination from the alpha and beta bands of EEG9. On the other hand, peak velocity measured by VOG is also higher than that recorded by SSC simultaneously7. This difference is attributed to the load of the search coil, influencing the saccade dynamics; possible slippage of the coil over the cornea, especially during blinks, may reduce the accuracy of eye movement measurement, leading to a slightly larger peak velocity measured by SSC than by VOG. In the present study, the peak velocity was lower when measured by EOG as compared to VOG. Presumably, this is because the low-pass filtering used here tends to decrease the peak velocity. Therefore, the difference in “accuracy” of each methodology may be due not only to the confounding noise, but also to how signals are processed (*e.g.,* low-pass filtering) as well as to inherent limitations of each recording method (*e.g*., slippage of the search coil).

Meanwhile, EOG has a clear advantage over the other eye movement recording methods in certain recording situations, *i.e*., subjects with narrow eye clefts and with cataract lenses. To adjust the method for the narrow eye cleft, experimenters can tape up the eyelids of subjects while recording, but this can irritate the eyes and result in excessive blinking and tears, which hampers reliable recording. In contrast, EOG can be used in patients with cataract lenses. For VOG, the signal is lost due to aberrant reflection associated with cataract lenses. Similarly, blinks may virtually “truncate” the VOG records, because the signal is lost during blinks. In contrast, horizontal EOG is less affected by blink artifacts, although small “spikes” corresponding to blinks in the records may be seen.

EOG requires only a short time for preparation, and may even be applicable to many patients with movement disorders that are less severe. Some neurological patients may have difficulty in stabilizing their trunk. Such movements may be detrimental for recording VOG as well. Considering these aspects, EOG shows a sufficient level of accuracy for clinical assessment; it is not that EOG is inherently “inaccurate” as a method for recording eye movements.

A practical guideline for recording the EOG in clinical applications has been published in 201723. The protocol here extends this proposal by including some additional procedures to further stabilize the EOG recording. The corneo-retinal potential can fluctuate with time, due to factors such as the alertness of subjects or environmental influences such as ambient light. The magnitude of the corneo-retinal potential difference is affected by various conditions and increases during light adaptation, while dark adaptation causes a decrease24,25. With sufficient dark adaptation, therefore, the corneo-retinal potential is expected to stabilize, leading to reduced drift. To reduce fluctuation further, the gain of EOG was continuously monitored throughout the experiment, and re-calibration was also performed for adjustment when necessary throughout the experiments. This re-calibration procedure took only 10–20 s to perform, so this did not intervene much with the recording procedures, and reduced the fluctuation of EOG signal. If the experimenter waits for 10–20 min after placing the electrodes, sufficient light adaptation will take place and the impedance between the electrode and the skin will also decrease and gradually asymptote to a low level (down to 20kΩ). The waiting period enables the recorded potential to stabilize dramatically from the beginning of the recording and to become increasingly stable with time.

Instead of the specialized dome embedding LEDs as described in this article, any board with LEDs embedded in a similar arrangement may be used. An alternate current (AC) amplifier can be used instead of a DC amplifier, but in this case, the amplitude of recorded saccades will not be reliable enough for qualitative assessment because of the signal decay. Electrodes having a wide fringe, which also serves to maintain close and wide contact with the skin, may be substituted for the electrode described in this article.

Some drawbacks of EOG should also be acknowledged. EOG is generally only adequate for recording horizontal eye movements, as raised in the Introduction. Furthermore, it is difficult to reliably assess microsaccades by the EOG method, whereas VOG has the capability to do so. This issue is especially important because of the saccadic spike potential and its fingerprint in the high frequency range26. Although these aspects could be problematic in the clinical context, they cannot be solved even with the present protocol and remains to be addressed in future studies. On the other hand, the eye position signal recorded by EOG can be contaminated by artifacts and noise, such as electromyography from facial muscles and electroencephalography. Also, when a DC amplifier is used, the recorded EOG signal can drift with time. These issues can be largely resolved by using an electrode with a plastic fringe that allows close and stable fixation as well as reduction of impedance between the skin and the electrode, effectively reducing the surrounding noise. Secondly, increasing the contact area between the gel and the skin by using a cup-electrode as described above, helps to lower the impedance at skin contact. Another way to avoid the drift is to wait for a period of 10–15 min after electrode placement, until sufficient light adaptation takes place. This waiting period also helps to further lower the impedance between the electrode (gel) and the skin, and the recorded EOG signal usually stabilizes as the time elapses. Repeating calibration and setting the gain of the gaze signal appropriately during the performance of oculomotor tasks can further help to improve the recording quality. The drift of eye position signal can pose a problem when recording smooth pursuit for which recording is usually made for an extended period. However, for recording saccades, whose duration lasts only for several tens of milliseconds, this is usually not an issue.

In summary, for achieving “accurate” EOG recording, it is not the methodology itself that matters, but how the experimenter implements it. The critical step is how to cope with the instability of the recording. The necessary measures are to use an Ag-AgCl electrode with a wide plastic fringe capable of effectively reducing noise, and to wait for sufficient light adaptation. This waiting period also helps to lower the impedance between the electrodes and the skin, thereby ensuring a stable signal recorded. Furthermore, re-calibration is performed as needed during the task performance. Thus implemented, EOG can still be a method of high clinical practicability that can be widely applied to neurological patients, especially for recording saccades in the horizontal direction. Indeed, EOG can be a preferable method when only this is available for economic reasons or in practical clinical situations where a readily implemented method is required and where omission of data is not permissible.

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The authors have nothing to disclose with regard to this study.

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