# Deconvolution of High Rate Flicker Electroretinograms

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***Abstract*— Flicker electroretinograms are steady-state electroretinograms (ERGs) generated by high rate flash stimuli that produce overlapping periodic responses. When a flash stimulus is delivered at low rates, a transient response named flash ERG (FERG) representing the activation of neural structures within the outer retina is obtained. Although FERGs and flicker ERGs are used in the diagnosis of many retinal diseases, their waveform relationships have not been investigated in detail. This study examines this relationship by extracting transient FERGs from specially generated quasi steady-state flicker and ERGs at stimulation rates above 10 Hz and similarly generated conventional flicker ERGs. The ability to extract the transient FERG responses by deconvolving flicker responses to temporally jittered stimuli at high rates is investigated at varying rates. FERGs were obtained from seven normal subjects stimulated with LED-based displays, delivering steady-state and low jittered quasi steady-state responses at five rates (10, 15, 32, 50, 68 Hz). The deconvolution method enabled a successful extraction of "per stimulus" unit transient ERG responses for all high stimulation rates. The deconvolved FERGs were used successfully to synthesize flicker ERGs obtained at the same high stimulation rates.**

## I. Introduction

The flash electroretinogram (FERG), one of the earliest electrophysiological responses of the retina, represents the impulse response of the eye [1]. This electrical response of the eye generated by a full-field flash stimulus has been a valuable clinical tool to evaluate the retinal function. It has also been used to diagnose many retinal diseases, such as cone and rod dystrophy, toxic retinopathy, retinitis pigmentosa and other disorders [2]. FERGs can be recorded non-invasively using different kinds of corneal or lower eyelid skin electrodes. The skin electrode montage adopted in this study produces slightly smaller signals but is more comfortable for the subjects, give that it produces less variability, and can be applied easily in a nonclinical laboratory [3].

In Fig. 1, a typical low rate, light adapted photopic FERG response obtained from a typical normal subject is shown on the top row. This response is dominated by a negative wave (a-wave) at around 20ms and a sharp positive peak (b-wave) at around 40ms, followed by a large broad negativity. As observed, oscillatory potentials (OPs) with frequencies at around 100-150Hz appear superimposed on the FERG. The a-wave is thought to result from the hyperpolarization of the cone photoreceptors and OFF bipolar cells, while the b-wave is elicited by Muller and ON bipolar cells [4, 5].

In relation to stimulation rate, two types of ERG responses are generated: the transient FERG responses and the steady-state (SS) responses. At low stimulation rates where the inter-stimulus interval (ISI) is longer than the evoked potential, a transient FERG response is acquired. At high stimulation rates (> 10Hz), the FERG evoked potentials overlap with each other, resulting in steady-state FERGs or flicker ERGs.

Due to their periodic nature, steady-state responses are typically analyzed in the frequency domain, losing most of the temporal peak information associated with specific retinal structures. The purpose of the present study is to investigate the temporal characteristics of the ERG waves at high stimulation rates in a population of normal subjects. This is achieved by extracting the "per-stimulus" unit response using the continuous loop averaging deconvolution (CLAD) algorithm successfully employed in other neurosensory evoked potentials [3,6].

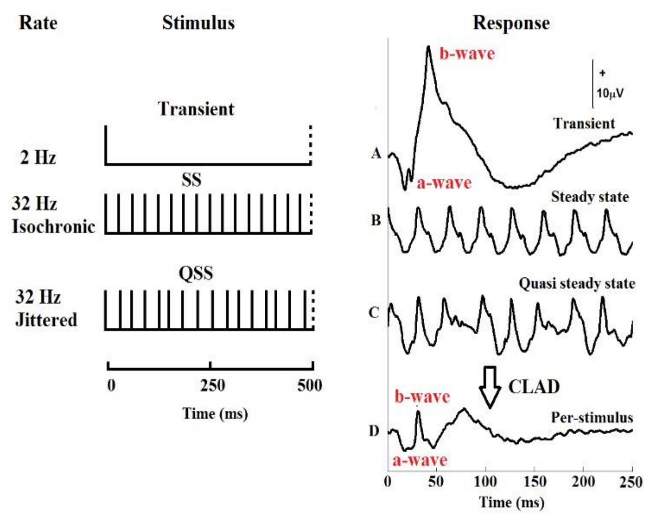


Figure 1. Transient FERG response (top row) at low rate of 2 Hz and steady-state flicker response at 32 Hz stimulation rate (two bottom rows). High rate ERGs are obtained using isochronic sequences generating steady-state (SS) responses (second row) or jittered sequences generating quasi steady- state (QSS) responses (third row). QSS flicker response can be deconvolved using the CLAD deconvolution algorithm to generate the "per stimulus" unit response generated by each flash at that rate (fourth right trace). Flash stimuli represented by vertical lines are shown on the left and the corresponding responses are displayed on the right columns.

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## II. METHODS

### A. Human Subjects

Seven young, healthy adults (5M, 2F) volunteered to participate, in accordance with the Institutional Review Board of University of Miami. The subjects' ages were from 20-27 years. They didn't have any history of ocular, retinal or neurological diseases and were not wearing eyeglasses or contact lenses.

### B. Stimulation

In this study, a specially designed white LED based visual display unit (VDU) covering an angle of 33$^o$ vertical by 38$^o$ horizontal with 300 cd/m$^2$ luminance was used [7]. This VDU was suitable for flicker response deconvolution since it was able to deliver 5 ms pulses of light with rise and fall times less than 200μs [3, 6].

Besides the conventional isochronic low rate stimulus of 1.98 Hz, five additional stimulation rates (10, 15, 32, 50 and 68 Hz) were investigated by delivering steady-state sequences (constant inter-stimulus intervals) and slightly jittered quasi steady-state (QSS) sequences. The QSS sequences were designed to have a low noise amplification factor (NAF) in the bandwidth (4Hz-150Hz) [6].

### C. Subject preparation and testing

Monocular FERGs were measured using gold cup skin electrodes placed on the forehead (ground) and under the eyelids. After skin preparation electrodes were connected as shown in Fig. 2. The electrode's impedance was kept below 3000 Ω. The active (positive) electrode was placed below the eyelid of the subject's test eye and the reference (negative) electrode was placed on the other lower eyelid. Non-test eye was occluded using a patch. All electrodes were filled with a medical grade conductive gel to enhance conductivity. After the electrode placement, the subject was seated in an electrically shielded dim recording chamber.

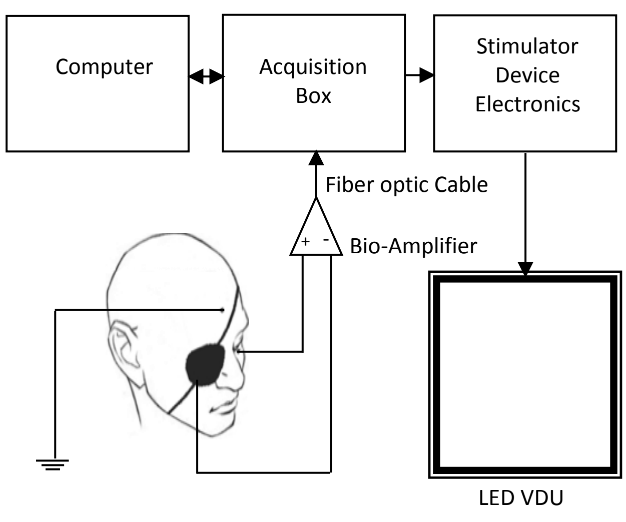


Figure 2 *Experimental acquisition set-up*

### D. Data collection

A commercial, two-channel Evoked Potential system with a continuous acquisition module (SEP-CAM, Intelligent Hearing Systems, Miami FL) was used to acquire the FERGs and deliver stimulation sequences. ERGs were amplified 100,000 times, band pass filtered (1-300Hz) and sampled at 2000 Hz.

Following the stimulation sequence, the acquisition system delivered trigger pulses, causing the visual display to turn on for 5ms. Meanwhile, the ERG responses were continuously acquired and stored for off-line analysis. For each stimulation condition, 128 sweeps with recording window duration of 500 ms were averaged (64 seconds). All evoked potentials (transient, SS and QSS) were processed offline using MATLAB. The averaging was done using an artifact rejection level of ± 30μv.

During acquisition, the subjects were instructed to remain relaxed, fixing their view on a marker at the center of the stimulator. After each acquisition, the subjects were allowed to rest briefly.

The conventional low rate (1.98Hz) FERG was obtained as reference. SS and QSS responses were obtained for each subject at stimulation rates of 10, 15, 32, 50 and 68 Hz. In total, eleven different responses (transient, SS and QSS) with three trials each were obtained from every subject. The typical recording time was around 40 minutes.

### E. Signal processing

The purpose of the signal processing was to deconvolve the QSS FERG to obtain the transient, per stimulus, FERG response when high-rate stimulation was used. The deconvolution process was then validated by assessing the capability of the deconvolved FERG to predict the acquired Steady State signal at the same rate.

As the ERG responses overlap with each other at high stimulation rates, introducing non-isochronic QSS sequences at high rates enabled the extraction of the "per stimulus" unit responses, a(t), using the CLAD algorithm [6]. The convolved signal v(t) can be presented in the time domain as the convolution of the per stimulus response a(t) with the stimulation sequence s(t) plus a residual noise coming from the process shown in (1).

$$v(t) = a(t) * s(t) + n(t) \quad (1)$$

Thus, by transforming the deconvolved response to the frequency domain, the convolution process becomes standard multiplication in (2). Therefore, extracting the unitary response now is achieved by dividing the measured response by the stimulus sequence and then transforming it back to the time domain as in (3).

$$V(f) = A(f) \cdot S(f) + N(f) \quad (2)$$

$$a(t) = FFT^{-1}\left[\frac{V(f)}{S(f)} - \frac{N(f)}{S(f)}\right], S(f) \neq 0 \quad (3)$$

Using the CLAD method all transient responses for each subject at each rate were averaged. The resulting "per-stimulus" responses were analyzed in time domain to obtain the amplitude and implicit times (latencies) of a-wave and b-wave in agreement with the ISCEV standard [8]. The amplitudes of the a-wave and the b-wave were measured from the baseline to the a-wave negative peak and from a-wave peak to b-wave peak, respectively. The implicit times of a-wave and b-wave were obtained from the stimulus onset to the wave peaks.

The ERG responses elicited by the five isochronic sequences (10, 15, 32, 50 and 68 Hz) were averaged to produce the real steady-state responses (384 sweeps / trace). These steady-state responses were de-noised using a comb filter that preserves the fundamental frequency and the nine harmonics. The resulting steady-state responses were analyzed in time and frequency domains. In time domain, the first peak implicit time and the trace peak to peak amplitude were measured as indicated by Birch [5]. In frequency domain, the spectrum of the signal was computed using the Fast Fourier Transform (FFT); the amplitude and phase of the fundamental frequency and its harmonics were also computed. Furthermore, depending on the assumption of steady-state superposition, the deconvolved transient responses were used to generate the synthetic steady-state in order to validate the obtained "per-stimulus" transient responses. The comparison between the real steady-state and synthetic steady-state was done both in time and frequency domains. In time domain, the correlation coefficient was used. In frequency domain, the phases and magnitude of the phasors was used.

## III. Results

The population averages of the convolved QSS responses, the deconvolved transient, real steady state, and the predicted synthetic steady state at the used stimulation rate are shown in Fig. 3. The population average of the convolved QSS and the conventional steady state are shown in Fig. 3A, and Fig. 3C. CLAD was successfully able to extract the per stimulus transient responses at the used high rate stimulation of 10, 15, 32, 50 and 68 Hz with clear and quantifiable a-wave and b-wave as shown in Fig. 3B. Averaged deconvolved waveform components are clearly visible and consistent in both implicit time and amplitude as shown in Fig. 3B. However, as the stimulation rate increased, component implicit time and amplitudes started to change. In Table I, means and standard deviations of amplitudes and latencies as a function of rate are listed.

The per-stimulus transient responses are mathematically convolved to generate synthetic steady waveforms as shown in Fig. 3D. Temporal analysis using the correlation coefficient between the synthetic and real steady-state of the main frequency and the first four harmonics except for 68Hz with the first two harmonics are consistent with 2, 10, 15, 32, 50 and 68 Hz as illustrated in Table II.

Additionally, frequency domain analysis of the steady state response and the synthetic steady response by studying the magnitude and phase differences were conducted. The spectral analysis showed that they both fit very well with minimal phase and magnitude differences as illustrated in Fig. 4.

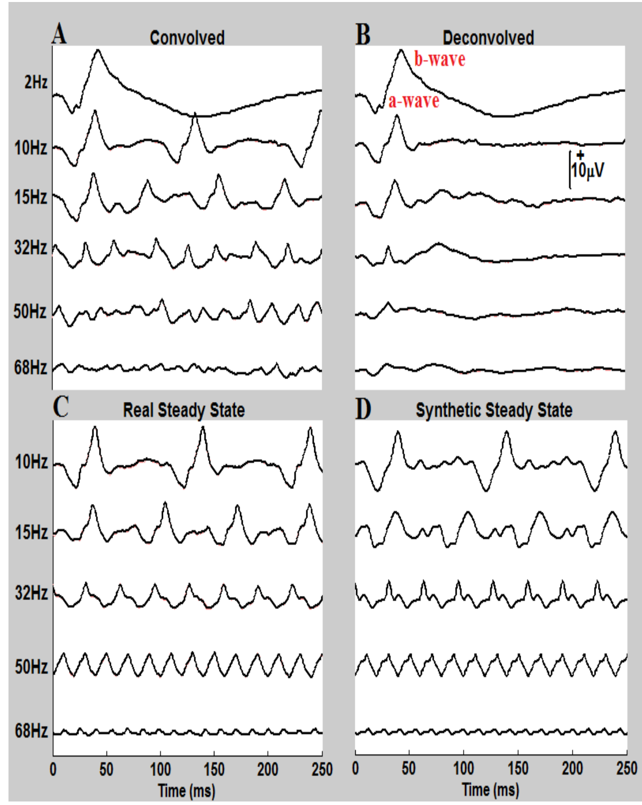


Figure 3. *Population average of ERG responses for the seven subjects. (A) The convolved responses to QSS sequences. (B) The extracted transient ERG responses. (C) The conventional steady state responses. (D) The predicted steady state responses.*

TABLE I. AMPLITUDES AND IMPLICIT TIMES OF THE MAJOR FERG COMPONENTS. AVERAGE VALUES AND STANDARD DEVIATION (IN PARENTHESES) ALONG THE POPULATION ARE SHOWN.

| Rate (Hz) | Amplitude (µV) | | Implicit time (ms) | |
|---|---|---|---|---|
| | *a wave* | *b wave* | *a wave* | *b wave* |
| 2 | -5.61 (2.13) | 18.12 (7.66) | 18.81 (0.76) | 41.39 (1.12) |
| 10 | -4.96 (1.04) | 12.98 (3.55) | 21.17 (2.19) | 38.53 (1.86) |
| 15 | -3.59 (1.17) | 10.53 (2.95) | 21.89 (1.82) | 36.31 (1.60) |
| 32 | -2.25 (1.04) | 5.14 (1.62) | 18.81 (2.63) | 30.53 (0.89) |
| 50 | -1.16 (0.48) | 4.38 (1.80) | 15.88 (1.32) | 30.1 (1.43) |
| 68 | -1.48 (0.76) | 3.20 (2.38) | 15.44 (2.19) | 30.1 (2.38) |

TABLE II. CORRELATION COEFFICIENT BETWEEN REAL STEADY STATE AND SYNTHETIC STEADY STATE AND SEQUENCE CHARACTERISTICS

| Rate (Hz) | Mean Rate (Hz) | Sequence Length | No. of stimulation | Correlation Coefficient |
|---|---|---|---|---|
| 2 | 1.95 | 1024 | 1 | N/A |
| 10 | 10 | 1000 | 5 | 0.97 |
| 15 | 15.03 | 1064 | 8 | 0.91 |
| 32 | 31.25 | 1024 | 16 | 0.95 |
| 50 | 50 | 1000 | 25 | 0.97 |
| 68 | 68.98 | 1015 | 35 | 0.82 |

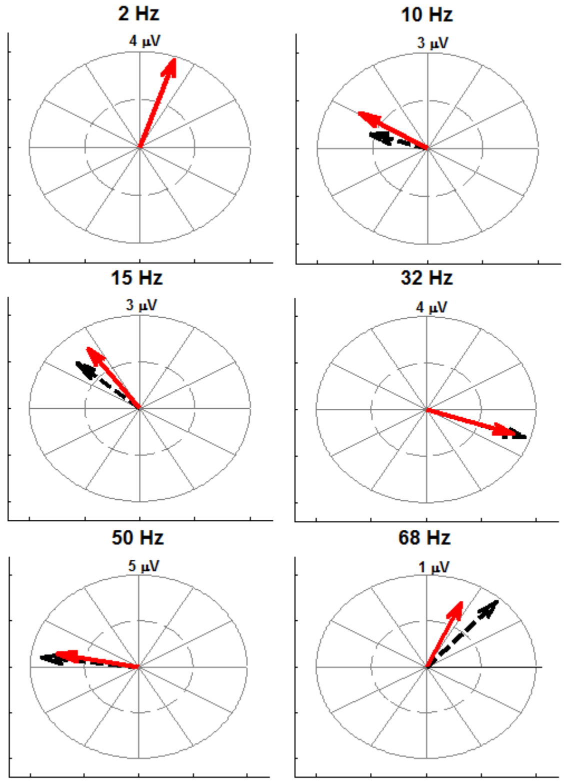


Figure 4. *Phasor of the fundamental frequency showing the comparison of magnitudes and phases between the real steady state (black-dotted arrows) and synthetic steady state (red arrows).*

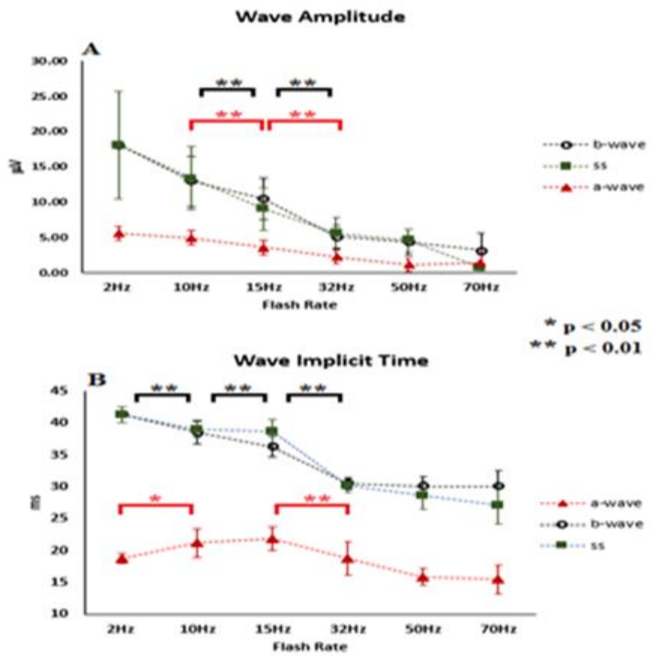


Figure 5. *Statistical analysis of a-wave, b-wave and real steady-state (SS) responses. (A) The amplitude of the waveforms components in microvolt. (B) The implicit time of the waveform components in millisecond.*

## IV. DISCUSSION

The transient and steady-state responses obtained in this study were consistent with the existing literature. The CLAD method enabled a successful extraction of the "per stimulus" transient ERG responses. For the first time, it was possible to analyze the ERG responses accurately in time domain at high frequency rates in a normal population. The a-wave and b-wave amplitudes and implicit times were clearly visible in the extracted transient responses at all stimulation rates. The experimental results showed that, as the stimulation rate increased, the a-wave and b-wave amplitudes decreased significantly between the 10 and 15 Hz and 15 and 32 Hz. Furthermore, the implicit time of a-wave increased between 2 and 10 Hz and decreased 15 and 32 Hz. Moreover, b-wave decreased significantly as the stimulation rate increased up to 32 Hz as shown in Fig. 5.

Interestingly, this study revealed new high-rate morphology of FERG waveform having a positive peak that follows b-wave and lasts for about 50 ms (Fig.1, bottom right). This peak was consistent across all stimulation rates but stronger at 32 Hz. The generators of this new peak need to be investigated in future studies.

The CLAD method assumes equal "per-stimulus" unit responses for each individual stimulus in the sequence. It is not possible to predict a-priori if this assumption will hold for the evoked responses and sequences used in this study. Excessive jitter may evoke different per-stimulus responses producing an unreliable deconvolved signal. Since the extracted per-stimulus responses used in this study successfully predicted the real recorded steady state responses, we can infer that the sequence jitter was adequate for this type of neural activity.

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