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Some Characteristics of the C-Wave of ERGs Recorded by a Pair of Electrodes on the Cornea and Sclera

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Summary. Light-evoked responses of the rabbit retina in situ were recorded between the electrode on the longitudinal line of the nasal side (positive) and the electrode on the opposite longitudinal line of the temporal side (negative). The c-wave was negative and the a-, b-wave was reversed when the negative (temporal) electrode was closer to the corneal center than the positive (nasal) electrode. The c-wave decreased its amplitude as the positive electrode came to the same latitudinal position as that of the negative one. Then, the c-wave was cancelled out in appearance on the recordings. When the negative electrode moved further into the posterior pole, the polarities of the c-wave as well as of the a-, b-wave were returned to those of routine ERGs.

Introduction

The electroretinogram (ERG) is routinely recorded between an active electrode on the cornea and a reference electrode on the tissue bio-electrically near the sclera. However, we have studied the action potential of the in situ retina as a mass, moving a pair of electrodes on the sclera, and reported some interesting findings for characteristics of the a-, b-wave and oscillatory potentials of the ERG (Honda, 1976a and b; Honda and Adachi-Usami, 1976). In this study, a slow potential (the c-wave of the ERG) was investigated, moving non-polarizable electrodes on the surface of the cornea and/or sclera, unlike routine electrode-positions of the ERG.

Methods

Albino rabbits weighing 3.5 kg each were used. Under a general anesthesia of urethane (1.0 g/kg body weight into the abdominal cavity), the upper and lower lids, the conjunctiva, and other surrounding tissue were removed. The skull was partially cut off in the orbital margin. A pair of non-polarizable zinc zinc-sulphate electrodes (Fig. 1) was

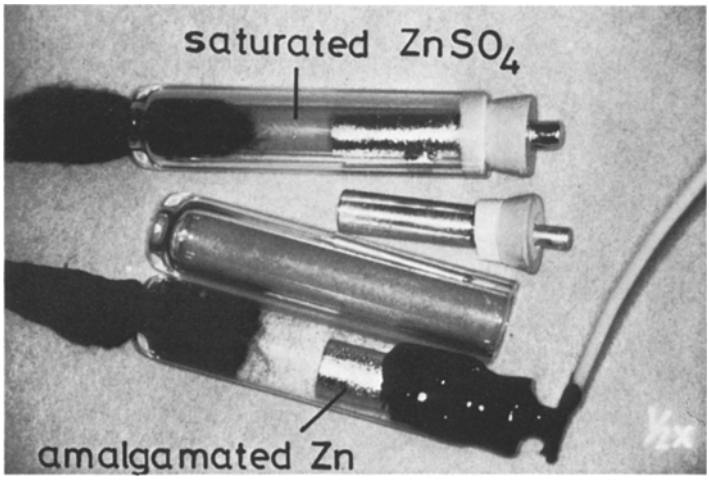


Fig. 1. Non-polarizable zinc zinc-sulphate electrodes were used in this study. The cotton-wick was solidified by gelatin with physiological solution, and the syringe was filled by saturated zinc-sulphate. The zinc pole was amalgamated by mercury and was inserted into the syringe. Then, the electrode was shielded from water and light (in this figure partially painted)

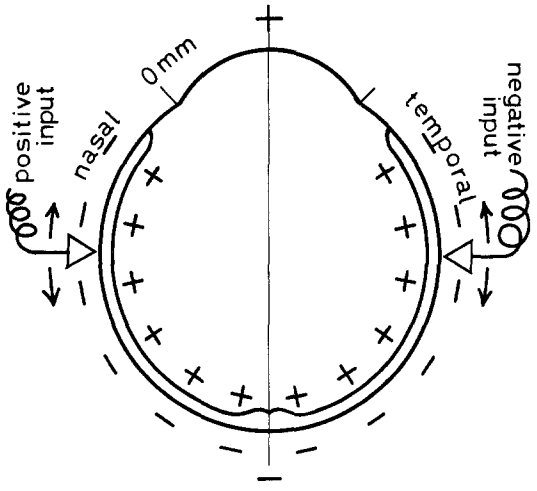


Fig. 2. The retina is a membrane attached on the inner surface of the eyeball. In this study, a pair of electrodes was attached on the surface of the cornea and/or sclera. The position of the electrodes is shown by the distance in longitude directly measured from the corneal limbus (Omm) (negative to the corneal center, positive distance to the optic nerve). The electrode on the nasal side was connected to the positive input of the amplifying system, and the electrode on the temporal side was connected to the negative input. The positivity of the positive input was recorded as an upward deflection. Responses were written on a pen-writing oscillogram continuously

held by microelectrode-manipulators, and cotton-wicks were attached on the surface of the cornea and/or sclera (Fig. 2). The position of the electrodes was shown by the distance in longitude directly measured from the corneal limbus (Omm) (negative latitudinal distance to the corneal center, positive distance to the optic nerve). The electrode on the longitudinal line of the nasal side was connected to an active input (positive) of

the amplifying system, and the electrode on the opposite (temporal) longitudinal line was connected to the negative input. The positivity of the active electrode was recorded as an upward deflection. Responses were written on a pen-writing oscillogram. The time constant of the amplifying system was 0.3 s.

A diffuse white light of incandescent tungsten filament from a photostimulator having a mechanical shutter (Nagata and Honda, 1970) was used as stimuli. The fre-

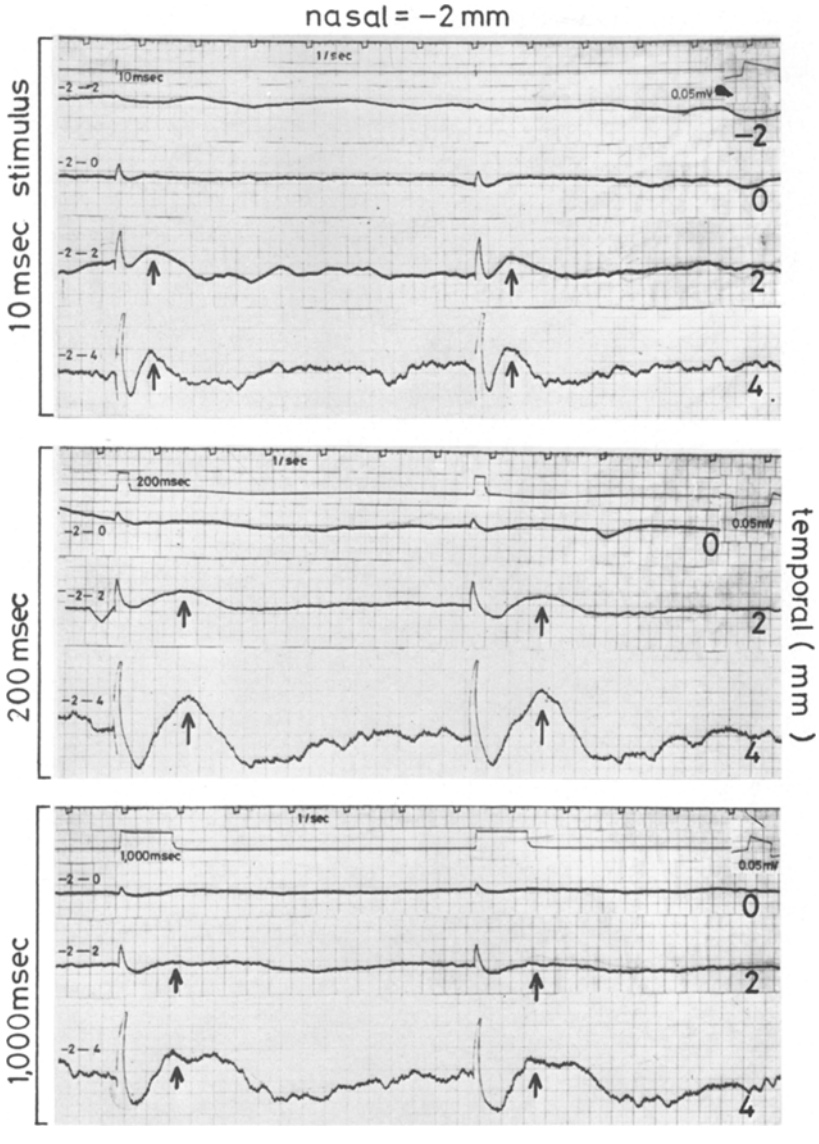


Fig. 3. In this figure, the nasal (positive) electrode is fixed in −2 mm (2 mm closer to the corneal center from the limbus). The position of the electrode on the temporal side is shown in mm on the right side of each recording. Three series of experiments by the stimuli of 10, 200 and 1000 ms are shown

quency of the stimulus was set at 1/s throughout the experiments. The duration of each stimulus was 10, 200, and 1000 ms. The stimulus intensity was approximately 2.1 x 10^3 ergs/cm^2· s at the entrance to the pupil. The pupil of the stimulated eye was dilated to the maximum by a mydriatica. The non-stimulated eye was shielded from the stimulus light by a black eye-patch, but was not enucleated. Before the beginning of each experiment, the eye was light-adapted under intermittent stimuli of 1/s.

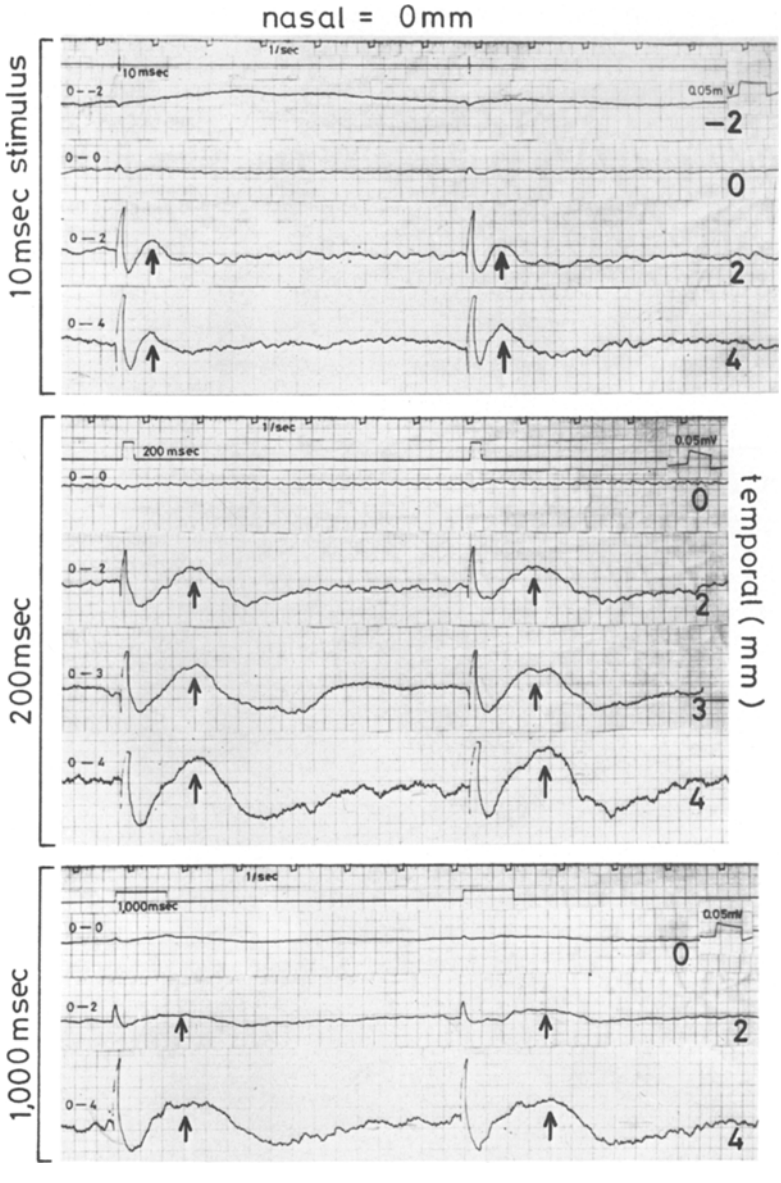


Fig. 4. The nasal electrode is fixed on Omm (corneal limbus). Three series of experiments by the stimuli of 10, 200, and 1000 ms are shown. The position of the temporal (negative) electrode is shown on the right side in mm

Results

All evoked responses in Figures 3 – 6 were recorded between the electrode on the nasal side (positive) and the electrode on the temporal side (negative). The electrodes were moved on the cornea and/or sclera in the manner mentioned above (Fig. 2). The c-waves in Figure 3 (nasal electrode fixed in –2 mm) and Figure 4 (nasal electrode fixed in Omm) were positive as those of usual ERGs. The amplitude increased as the temporal electrode moved to the posterior pole. Changes of the wave form were fundamentally equal among responses by the stimuli of 10, 200, and 1000 ms, although positive off-responses ap-

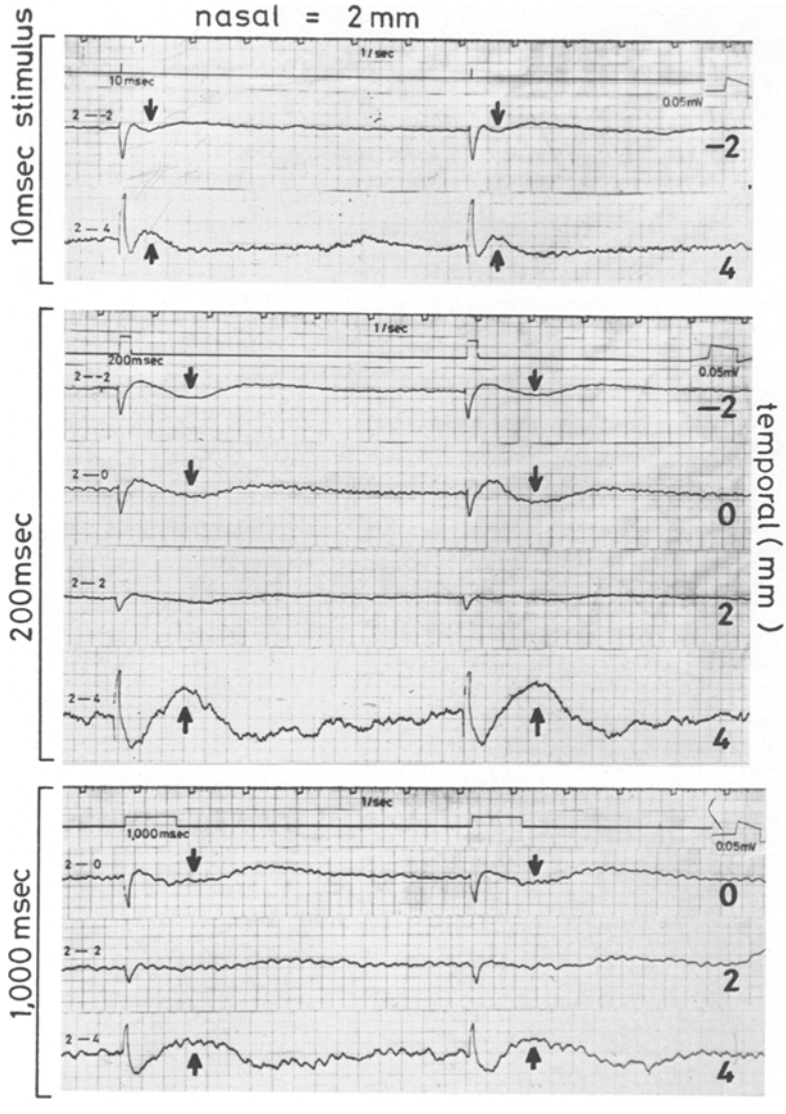
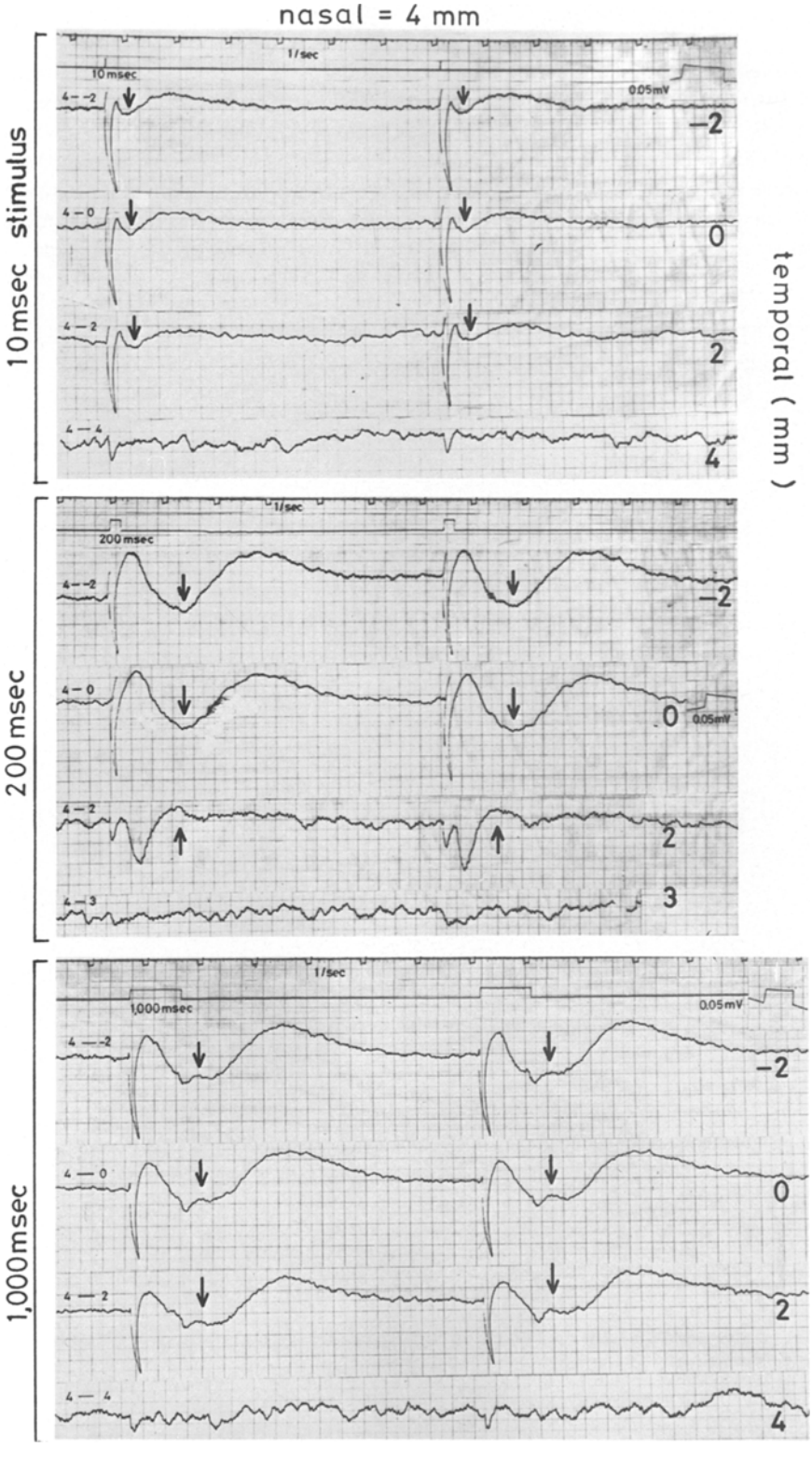


Fig. 5. The nasal electrode is fixed on 2 mm. The position of the temporal electrode is shown on the right side in mm


nasal = 4 mm
temporal (mm)
10msec stimulus
200 msec
1,000msec
1/sec
10 msec
200 msec
1,000msec
0.05mV
4 — 2
4 — 0
4 — 4
4 — 3
-2
0
2
4
3

peared by 1000 ms stimuli. The polarities of the a- and b-wave in Figures 3 and 4 were the same as those of usual ERGs, and the amplitude also changed in the same way the c-waves changed.

In Figure 6 (nasal electrode fixed in 4 mm), however, the c-waves were almost negative, and the positive a-wave and the negative b-wave were observed. They looked like reversed patterns of the responses in Figures 3 and 4 or routine ERG recordings. Amplitudes of the c-waves in Figure 6 decreased as the temporal electrode came to the same latitude as that of the nasal one. Changes of the wave form and the amplitude were fundamentally equal among responses evoked by the stimuli of 10, 200, and 1000 ms, although negative off-responses appeared on the descending phase of the negative c-waves by 1000 ms stimuli. In Figure 5, both forms of responses were mixed. The c-waves were positive only when the temporal electrode was deeper than that of the nasal one.

These observations on the wave form and the amplitude of the c-wave might be summarized as follows: The c-wave was negative and the a-, b-wave was reversed when the negative (temporal) electrode was nearer to the corneal center than the positive (nasal) electrode. The c-wave decreased its amplitude as the positive electrode came to the same latitudinal position as that of the negative one. When the negative electrode was moved further into the posterior pole, the polarities of the c-wave as well as of the a-, b-wave were returned to those of routine ERGs.

Discussion

In previous papers (Honda, 1976a and b) we have reported that polarities and amplitudes of the a-, b-wave of the ERG as a mass response are easily affected by the position of electrodes on the sclera. The wave form of the a-, b-wave depends on the balance of latitudinal position of electrodes on the sclera. The a-, b-wave seems to be cancelled out, isolating the oscillatory potentials of the ERG, when the latitudinal positions of the electrodes on the opposite sides of the eye are equal (Honda and Adachi-Usami, 1976). In this study employing an amplifying system of a longer time constant (that was not D.C. but was enough to follow the c-wave) and a pen-writing oscillogram, the c-wave was shown to change in a way similar to that of the a-, b-wave. The c-wave was cancelled out in appearance on the recordings when the latitudinal positions of the electrodes were equal. It might be said that the origin of the c-wave distributes in the retina of plane extent as that of the a-, b-wave. This study also showed indirectly that the origins of the c-wave and the oscillatory potentials are different, because, in the previous paper, the plane distribution of the oscillatory potentials has been shown to be different from that of the a-, b-wave (Honda and Adachi-Usami, 1976). These conclusions agree with many electrophysiological observations of the origin of the ERG components, dealing with the in vivo and in vitro retina as a membrane having thickness (Brown, 1968).

The retina is a membrane attached on the inner surface of the eyeball (Fig. 2) which is buried in the orbit. Early history of the clinical ERG was how to pick up effectively

Fig. 6. The nasal electrode is fixed on 4 mm. The position of the temporal electrode is shown on the right side in mm

the evoked responses from the retina in situ, and contact lens electrodes have been developed. However, employing an averaging computer clear ERGs were recordable, not only by the corneal electrode but also by skin-electrodes on the orbital margin (Schmidt and Straub, 1970). It should be remembered again that the "normal" pattern and the amplitude of routine ERGs by corneal electrodes are temporarily established in a situational relationship between the electrode and the eyeball, and that the evoked potentials are generating from wide retinal areas and the routine ERG is only a summed result monitored from the outside of the eyeball.

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