

## Comparison of Two Electroretinography Systems Used in Dogs: The HM<sub>s</sub>ERG and the RETIport

Man Bok JEONG<sup>1)</sup>, Won Gyun SON<sup>1)</sup>, Young Woo PARK<sup>1)</sup>, Se Eun KIM<sup>1)</sup>, Shin Ae PARK<sup>2)</sup> and Kang Moon SEO<sup>1)\*</sup>

<sup>1)</sup>Department of Veterinary Surgery and Ophthalmology, College of Veterinary Medicine and BK21 Program for Veterinary Science, Seoul National University, San 56-1, Sillim 9-dong, Gwanak-gu, Seoul 151-742, Republic of Korea and <sup>2)</sup>Department of Surgical and Radiological Sciences, School of Veterinary Medicine, University of California Davis, One Shields Ave, Davis, California 95616, U.S.A.

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**ABSTRACT.** The purpose of the study was to compare two different electroretinography (ERG) instruments used on the same animal in a laboratory setting. Retinal function in both eyes of 12 healthy miniature schnauzers was evaluated under general anesthesia. Scotopic and photopic ERGs were recorded by the HM<sub>s</sub>ERG and the RETIport using the Dog Standard Protocol recommended by the European College of Veterinary Ophthalmologists (ECVO). The waveforms of the ERGs obtained by the two ERG units were similar to those described in previous studies. The 90% reference ranges using the multiple of medians (MoM) showed that the changes in ERG parameters obtained by the HM<sub>s</sub>ERG unit were very similar to those of the RETIport for most ERG responses, except for a few. The results demonstrate that the two ERG systems are comparable for evaluating retinal function in dogs. Further, the results also show that it is necessary to establish ERG system-specific limits of normality in each laboratory or clinic in order to obtain a correct diagnosis, for example by using a graphical representation of the 90% reference range.

**KEY WORDS:** dog standard protocol, electroretinography, HM<sub>s</sub>ERG, retinal function, RETIport.

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Electroretinography is an important objective electrophysiological technique used to evaluate retinal function in humans and animals [27, 29]. As well as being used for the evaluation of retinal function prior to cataract surgery, ERG is also applied in the early diagnosis of inherited retinal degenerations, in the diagnosis of sudden acquired retinal degeneration (SARD) and optic neuritis, and more recently, in the monitoring of therapeutic responses and in the evaluation of new drugs in toxicity studies [5, 24, 30, 32].

The most important components of the ERG equipment are the stimulating and the recording systems. Many light stimulating systems or photostimulators have been developed in order to obtain stimulation of the retina, such as xenon strobos, tungsten bulbs and the light-emitting diodes (LEDs) [16–18, 21].

In order to obtain an overall response from retinal cells it has been shown to be important to provide full-field stimulation [4, 6]. For this purpose the so called Ganzfeld (or ‘full-field’) stimulator was developed and now recommended for use in both humans and animals [22, 25]. A contact lens electrode with a built-in LED together with a recording system, have been used in human and animal ERG studies during the past years [10, 12, 20]. These previous studies employed a similar type of light stimulator (a contact lens active electrode with built-in LEDs stimulator), which is the type used for the ERG equipment (RETIport, Roland Consult, Brandenburg, Germany) in the present

study. Further, a portable LED mini Ganzfeld ERG unit, also called the handheld multi-species ERG unit (HM<sub>s</sub>ERG, RetVet Corp. Inc., Columbia, MO, U.S.A.), was recently introduced into the field of veterinary ophthalmology [9, 28].

In the present study similar procedures for ERG recordings were performed using the two previously mentioned types of ERG instruments (HM<sub>s</sub>ERG and RETIport). The light stimulator and the provided light stimulation were different; however, one being a mini-Ganzfeld utilizing LED stimulation (HM<sub>s</sub>ERG), and the other a combined contact lens active electrode and LED-stimulator (RETIport). To our knowledge, no previous reports have been published comparing the newly developed mini-Ganzfeld ERG (HM<sub>s</sub>ERG) with a conventional table top ERG equipment (RETIport) in healthy normal dogs. The purpose of the present study was therefore to compare the two ERG instruments under similar laboratory conditions using the same animal.

### MATERIALS AND METHODS

*Experimental animals:* Both eyes of 12 healthy miniature schnauzers (6 males, 6 females) were evaluated in the present study. Animals ranged in age from 9–11 months (mean ± SD; 10.6 ± 0.7), weighing 3.4–6.8 kg (mean ± SD; 5.2 ± 1.1). Ophthalmic examination, including slit lamp biomicroscopy (SL-202P, Shih-nippon commerce, Tokyo, Japan) and indirect ophthalmoscopy (Vantage, Keeler instruments Inc., Broomall, PA, U.S.A.), showed no abnormalities in any of the eyes tested. The experiments adhered to the strict guidelines of the “Guide for the Care and Use of Laboratory Animals” of the Institute of Laboratory Animal

\* CORRESPONDENCE TO: SEO, K. M., Department of Veterinary Surgery and Ophthalmology, College of Veterinary Medicine and BK21 Program for Veterinary Science, Seoul National University, San 56-1, Sillim 9-dong, Gwanak-gu, Seoul 151-742, Republic of Korea.  
e-mail: kmseo@snu.ac.kr

Resources of Seoul National University.

**Animal preparation for ERG:** All animals were fasted for at least 12 hr before performing the ERG recordings. Maximal pupillary dilation was obtained by applying 1 drop of 1% tropicamide (Mydracil, Alcon Inc., Puurs, Belgium) to the test eye every 20 min from at least 1 hr (3 times within 1 hr) prior to beginning the ERG session. The size of the pupil was measured by a caliper under a focal light source (Heine 6-21-301, Heine, Berlin, Germany) at the beginning and at the end of ERG procedure. Each of 12 experimental animals was sedated using medetomidine HCl (Domitor, Pfizer animal health Korea Ltd., Seoul, Korea, 60  $\mu\text{g}/\text{kg}$ , IM) and was kept in a cage in a silent area under ambient light for approximately 5 min. After that, ketamine HCl (Yuhan Ketamine, Yuhan Corporation, Gunpo-si, Korea, 5 mg/kg, IM) was injected. The anesthesia was monitored by a veterinarian. The dogs were positioned in their sternal recumbency and the head was placed on a pack of towels. In order to avoid rotation of the globe, conjunctival stay sutures through the skin of upper eyelid were used to position the eyes.

**ERG procedures:** Prior to the ERG recordings both ERG equipments were calibrated using a custom made 260  $\mu\text{V}$  input and the amplitude of the resultant sine wave measured. This was performed in order to show that each ERG instrument could register the input provided by the calibrator. Flash intensity calibration of the photostimulators of both ERG units had been performed by each respective manufacturer within 6 months of the study.

ERGs were recorded from the left eye of each dog using the HM sERG system, and the fellow eye was tested using the same ERG instrument one week later. The same procedure was executed using the RETiport system, one week following the HM sERG recordings. Thus, each dog was anesthetized and tested four times. Platinum subdermal needle electrodes (Model F-E2, Astro-Med Inc., West Warwick, RI, U.S.A.) were used to record ERGs; a reference electrode and a ground electrode were placed approximately 3 cm caudal to the lateral canthus and over the external occipital protuberance, respectively. The cornea was anesthetized by topical 0.5% proparacaine hydrochloride

(Alcaine, Alcon, Puurs, Belgium). For the mini-Ganzfeld (HM sERG), an eyelid speculum was applied to the test eye. Then an active contact lens electrode (ERG-Jet, Universo Plastique, LKC Technologies Inc., Gaithersburg, MD, U.S.A.) was placed on the cornea, following the application of topical artificial tear gel (Optagel, Samil Pharm. Ltd., Seoul, Korea). For the RETiport, a contact lens electrode with a built-in white LED stimulator (Kooijman/Damhof ERG lens, Medical Workshop BV, Groningen, Netherland) was used. The electrodes were connected to a preamplifier, and signals were amplified with a band pass filter between 0.3 and 300 Hz for the HM sERG and between 1 and 300 Hz for the RETiport. Flash duration was 0.005 to 5 msec for the two ERG systems used.

Each ERG session consisted of scotopic and photopic ERGs for both ERG instruments. For scotopic ERGs 3 different responses were recorded: scotopic low intensity responses (S) for study of rod function, which was designated S1, S2, S3, S4, and S5; scotopic standard intensity responses (S-ST) and scotopic higher intensity responses (S-H), the latter two for stimulation of both rods and cones. Photopic ERGs consisted of two different responses including photopic single flash response (P) for evaluation of cones and photopic 30 Hz flicker responses (P-FL) for cones and the post-synaptic component responses [13, 14].

The HM sERG was positioned as close to the eye as possible without touching the animals (Fig. 1a). Scotopic low intensity responses were elicited at 0.01  $\text{cd}\cdot\text{s}/\text{m}^2$ , an average of 10 flashes with an interval of 2 sec every 4 min [i.e., at 4 (S1), 8 (S2), 12 (S3), 16 (S4) and 20 (S5) min] during 20 min of dark adaptation. After 20 min of recordings, the light stimulus intensity was increased to 3  $\text{cd}\cdot\text{s}/\text{m}^2$  and the responses to an average of 4 flashes with an interval of 10 sec were recorded. Scotopic higher intensity responses were recorded using 10  $\text{cd}\cdot\text{s}/\text{m}^2$  with an average of 4 flashes and an interval of 20 sec. After 10 min of light adaptation using the mini-Ganzfeld dome (background luminance: 30  $\text{cd}/\text{m}^2$ ), photopic single flash responses were recorded with a 3  $\text{cd}\cdot\text{s}/\text{m}^2$  flash stimulus, averaging 32 flashes at an

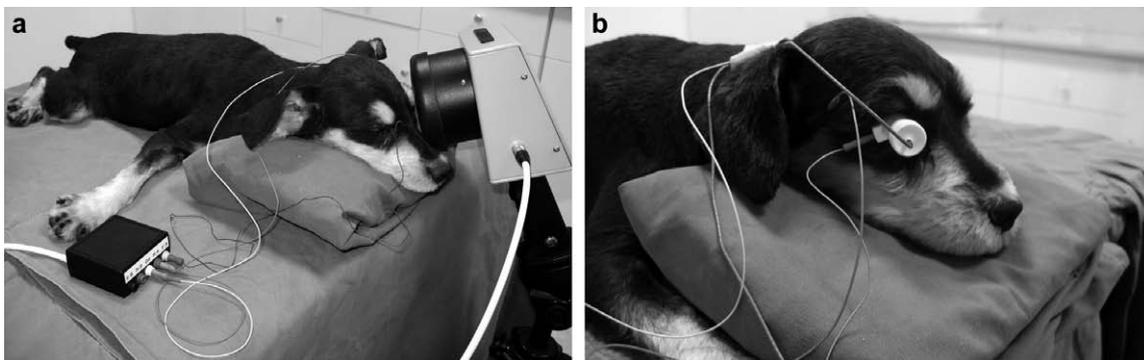


Fig. 1. ERG using the HM sERG (a) and the RETiport (b) in miniature schnauzer dogs. Note the difference between the portable LED mini-Ganzfeld stimulator used in the HM sERG and the contact active lens electrode with built-in LED stimulators (LED-electrode) used in the RETiport. The entire HM sERG unit is shown (a), while only the contact lens with the built-in stimulator is shown for the RETiport (b).

interval of 0.5 sec under light adaptation. Photopic 30 Hz flicker responses were thereafter recorded using the same light intensity.

The LED-electrode of the RETIport was placed on the cornea after topical anesthesia with 0.5% proparacaine hydrochloride (Alcaine, Alcon), following the application of topical artificial tear gel (Optegel, Samil Pharm. Ltd.) (Fig. 1b). A piece of tape was used to keep the contact lens in a correct and stable position on the eye by taping the cord of the LED-electrode to the head. This was done in order to prevent the LED-electrode from rotating downwards. A scotopic low intensity response was elicited at 0.025 cd·s/m<sup>2</sup> using single flashes (for rod responses) every 4 min during 20 min of dark adaptation. After the 20 min recording, the flash light intensity was increased to 2.5 cd·s/m<sup>2</sup>, and the response to a single flash (scotopic standard intensity response) was recorded. A scotopic higher intensity response was recorded using 7.9 cd·s/m<sup>2</sup> (the latter two for mixed rod and cone responses). After 10 min of light adaptation (background luminance: 25 cd/m<sup>2</sup>), the photopic single flash response to a single 2.5 cd·s/m<sup>2</sup> flash stimulation was recorded under light adaptation (for cone responses). Photopic 30 Hz flicker responses were recorded using the same light intensity (for cone and post-synaptic component responses).

The amplitude and implicit times of a- and b-waves were measured. The amplitude of the a-wave was calculated from the baseline to the first negative deflection, and the amplitude of the b-wave was measured from the trough of the a-wave to the positive peak of the b-wave. Implicit times of a- and b-waves were calculated from the onset of light stimulus to the peak of the a- and b-waves, respectively. For the scotopic low intensity responses and photopic 30 Hz flicker responses, only b-waves were recorded and thereby measured. Amplitudes of the photopic flicker responses were measured from the baseline to the positive peak and the implicit times from the light onset to the positive peak.

*Statistical analysis:* Repeated measures analysis of variance (Anova) was used to investigate the Least Square Means (LSM) differences between the two different types of ERG equipments, at a given level of light intensity. The repeated measures Anova was used since the same dog was measured under different conditions: two types of ERG equipments, different levels of light intensity, and both eyes (left and right). All possible two-way and three-way interactions were considered in the model. An F-test was used for multiple comparisons of each mean response (amplitudes and implicit times of a- and b-waves) measured using the two different ERG equipments for a given light intensity. In view of these multiple tests,  $P < 0.01$  was considered statistically significant to control type I error. All analyses were conducted using statistic computer software (SAS, version 9.1, SAS Institute Inc., Cary, NC, U.S.A.).

For data description purpose only, the multiple of the medians (MoM) and 90% reference range were used [8]. The MoM expresses the data points of the amplitude and

implicit time of the a- and b-wave as a proportion of the median value for the different types of ERG. The MoM was used to establish normative values for each type of ERG used in the MS breed of dogs, in the age group 9–11 months. The lower limit and the upper limit of the 90% reference range are defined as the 5th percentile and the 95th percentiles of the MoM distribution, respectively.

## RESULTS

Calibration of the two ERG instruments showed that with the 260  $\mu$ V input, the HMsERG showed a recording of 275  $\mu$ V and the RETIport 264  $\mu$ V.

*Waveforms of the ERG:* Figure 2 displays examples of typical normal ERG waveforms recorded from left eye of the same miniature schnauzer dogs by the HMsERG and the RETIport instruments, respectively. As shown by the Fig. 2, both instruments gave results that were similar, as to configuration of the waveform. The scotopic low intensity responses had a prominent b-wave (without an a-wave) that increased in amplitude and the implicit time increased, during the 20 min of dark adaptation. The scotopic standard intensity response and scotopic higher intensity response had prominent a- and b-waves. The photopic single flash response had smaller but discernible and faster a- and b-waves when compared to the scotopic standard intensity and the scotopic higher intensity responses. The photopic flicker responses consisted of b-waves only.

*A- and b- wave parameters of the ERG:* Figure 3 demonstrates the changes of LSM and 95% confidence intervals in amplitude and implicit time of the a- and b-waves obtained using the two ERG instruments in the same animal for each test session. For the scotopic low intensity responses, there were significant differences in the mean amplitudes of the b-waves obtained between two ERG equipments (Fig. 3c;  $P = 0.0016$  for S1 and  $P < 0.0001$  for S2 to S5). However, these was a significant difference in the mean implicit time of the b-waves between the two ERG equipments only for S4 (Fig. 3d;  $P = 0.0025$ ). For the scotopic standard intensity response, the mean implicit time of the a-wave recorded by the HMsERG was significantly different from that of the RETIport (Fig. 3b;  $P = 0.0059$ ). For the photopic single flash response, there were significant differences in the mean amplitude of the a-wave and in the mean implicit time of the b-wave (Fig. 3a and 3d;  $P = 0.0002$  and  $P < 0.0001$ , respectively). Figure 4 illustrates a graphical representation of reference ranges for each ERG parameter using the MoM and 90% reference range [8].

## DISCUSSION

The purpose of the present study was to compare the HMsERG and RETIport ERG instruments in the same animal. The results showed that the ERG waveforms obtained by the two ERG units were remarkably similar. The graphical representation of the 90% reference range showed that the changes in ERG parameters obtained by the HMsERG

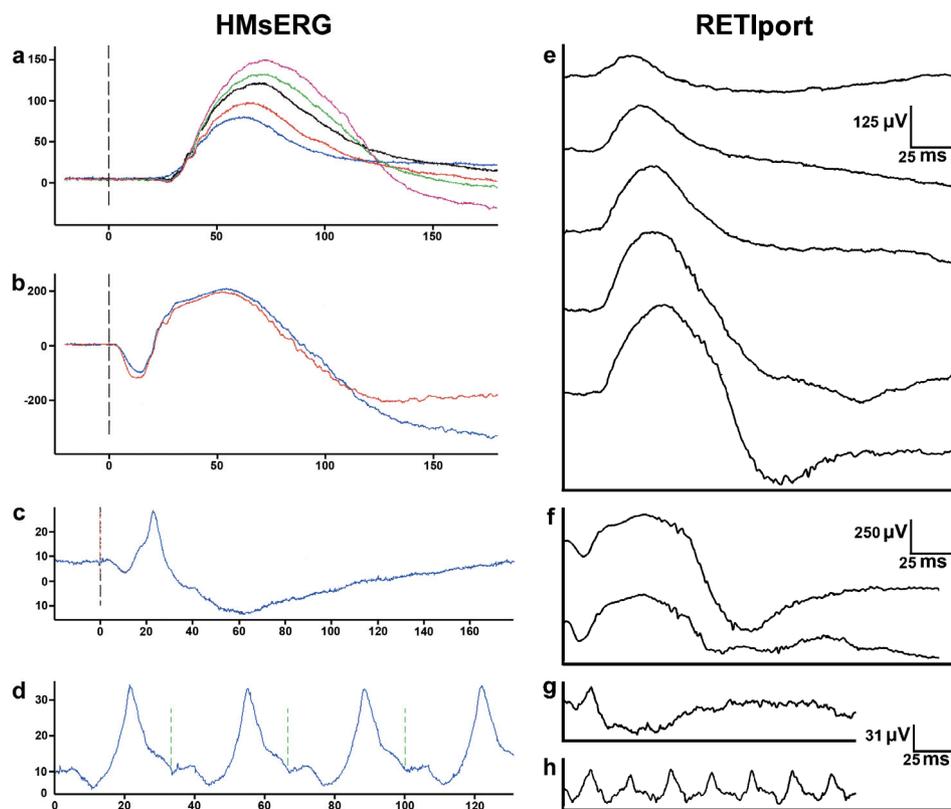


Fig. 2. Representative ERG waveforms recorded from left eye of the same miniature schnauzer dogs by the two ERG units. Scotopic ERGs (a and b, e and f) and photopic ERGs (c and d, g and h) are shown. Scotopic low intensity responses (A and E), which were elicited 4, 8, 12, 16, 20 min after dark adaptation (designated S1, S2, S3, S4, and S5 from bottom to top in A and from top to bottom in E, respectively), scotopic standard intensity responses (blue line in b and upper line in f), and scotopic higher intensity responses (red line in b and lower line in f). Photopic single flash responses (c and g) and photopic flicker responses (d and h) are shown. The vertical dashed line illustrates the onset of the light stimulus for the HMsERG, while the light stimulation occurs at the beginning of each recording for the RETIport.

unit were very similar to those of the RETIport for most ERG recordings, except for a few of the responses. When evaluating the specific ERG parameters obtained from each instrument some significant differences were found, however, most probably due to equipment-related factors.

Stimulus luminance for each ERG unit was measured by the respective companies. In order to investigate if both ERG instruments could provide reliable measurements, calibration of the two units was performed using an input of  $260 \mu\text{V}$  by a custom-made electronic device constructed by Linscan ultrasound Inc., Rolla, MO, U.S.A. Response amplitudes were obtained in the range of the expected amplitudes for both instruments, set at  $\pm 10\%$  by one of the manufacturers (Linscan ultrasound Inc.). According to guidelines of the International Society for Clinical Electrophysiology of Vision (ISCEV) for calibration, it is important to calibrate both the stimulation and recording systems on a regular basis in order to perform technically adequate ERG recordings [3].

ERG responses are influenced by equipment-related and physiological factors. The former may be intensity of the light stimulus used, filter and amplifier settings, conductor characteristics of the electrodes, and position of the electrodes [23, 35]. The latter may be the level of dark-adaptation [34], exposure to bright light prior to the ERG recordings [31], and level of anesthesia or sedation [11, 33]. Other variables that affect ERGs are breed of dog, age and the degree of pupil dilation [1]. All of these factors can cause problems comparing ERG results recorded in different clinics or laboratories, especially when using different equipments. For the present study, most procedures in relation to physiological and environmental factors were standardized as far as possible leaving the equipment-related factors as the main variables.

In regards to equipment-related factors there were some major differences between the two ERG units used in the present study. These were: the type of active electrodes used (ERG Jet lens versus Kooijman/Damhof ERG lens);

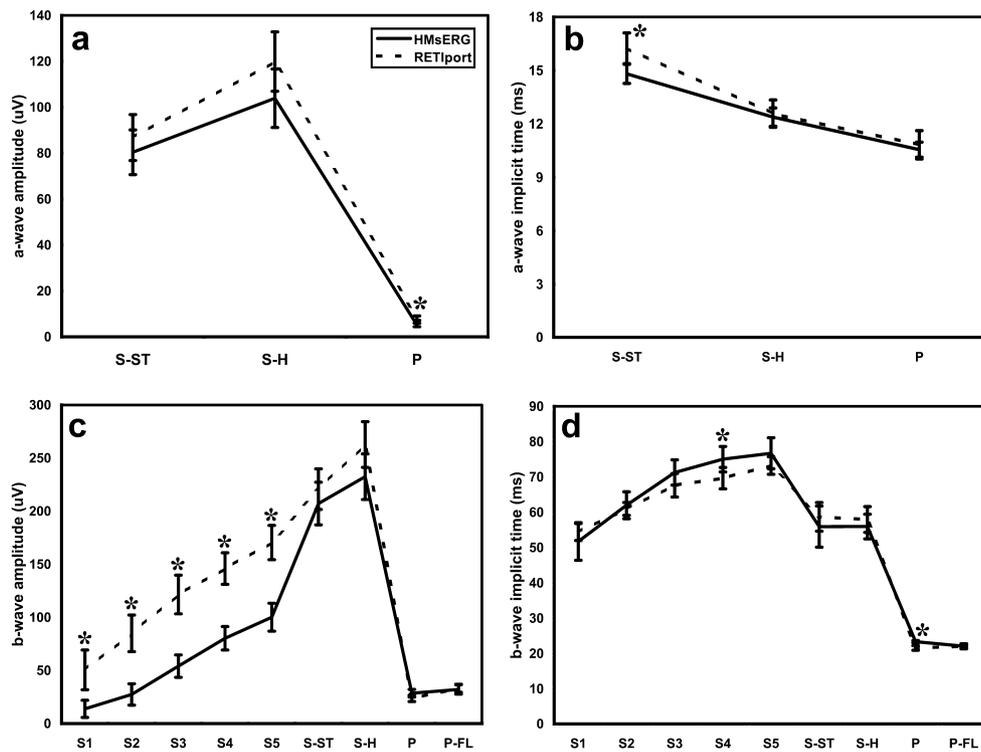


Fig. 3. Least Square Means and 95% confidence interval ( $\text{LSM} \pm 1.96 \text{ SE}$ ) of amplitude (a and c) and implicit time (b and d) of a- and b-waves recorded by the HMSeERG (a solid line) and the RETIport (a dotted line). \* indicates a significant differences between the HMSeERG and the RETIport. S1 to S5 depicts scotopic low intensity responses obtained using  $0.001 \text{ cd}\cdot\text{s}/\text{m}^2$  for HMSeERG and  $0.025 \text{ cd}\cdot\text{s}/\text{m}^2$  for RETIport of light stimulation 4, 8, 12, 16, and 20 min, respectively after dark adaptation, designated S1, S2, S3, S4, and S5, respectively on the horizontal axis. S-ST and S-H depict scotopic standard intensity and scotopic higher intensity responses obtained at  $3$  and  $10 \text{ cd}\cdot\text{s}/\text{m}^2$  for HMSeERG and  $2.5$  and  $7.9 \text{ cd}\cdot\text{s}/\text{m}^2$  for RETIport, respectively. Photopic ERGs consisted of a photopic single flash (P) response, and photopic 30 Hz flicker (P-FL) responses for the HMSeERG and RETIport, respectively, depicted as P and P-FL at  $3 \text{ cd}\cdot\text{s}/\text{m}^2$  for the HMSeERG and  $2.5 \text{ cd}\cdot\text{s}/\text{m}^2$  for the RETIport after 10 min of light adaptation using  $30$  and  $25 \text{ cd}/\text{m}^2$  of background light for the HMSeERG and the RETIport, respectively.

the distance between the recorded eye and the light stimulator; the use of signal averaging (which was not used for the RETIport); and the light stimulus intensity and the background illumination used. Therefore, it appears reasonable to compare the configuration of the ERG waveforms and the changes in ERG parameters obtained using a graphical representation of the 90% reference range during the recording sessions for each of the equipments, rather than to directly compare the specific ERG values recorded by each instrument. Further, by illustrating the results by using the median and the 90% reference range, differences could be clearly visualized.

As illustrated in Fig. 2, the waveforms of the ERG recordings obtained using the HMSeERG was similar to those obtained by the RETIport in most respects. Furthermore, the ERG results recorded in the present study were very similar to those of a previous study, detailing procedures for obtaining standardized ERGs in dogs [26]. The ERG waveforms obtained in the present study demonstrate that any one of the two ERG systems used can be utilized in evaluat-

ing retinal function in dogs.

For the scotopic low intensity responses, the amplitudes of the b-waves increased dramatically during the process of dark adaptation using both ERG units, while the b-wave implicit times increased slightly during the 20 min of dark adaptation (Fig. 3c and 3d). With increasing time in the dark, there was thus an increase in amplitudes and implicit times of b-waves for the scotopic responses, which reflects the process of dark-adaptation of the retina [34]. Both ERG systems therefore could record electrical activity of the rod photoreceptors under scotopic conditions and under dim light stimulation, and monitor the process of dark-adaptation. For the scotopic higher intensity responses, which are mixed responses for both rods and cones, an unexpected finding was that the a- and b-wave amplitudes for the RETIport were higher than those of the HMSeERG. This was found even though the light intensity for the HMSeERG was higher than that used for the RETIport for the specific responses. The differences in the scotopic higher intensity amplitudes found are likely due to ERG

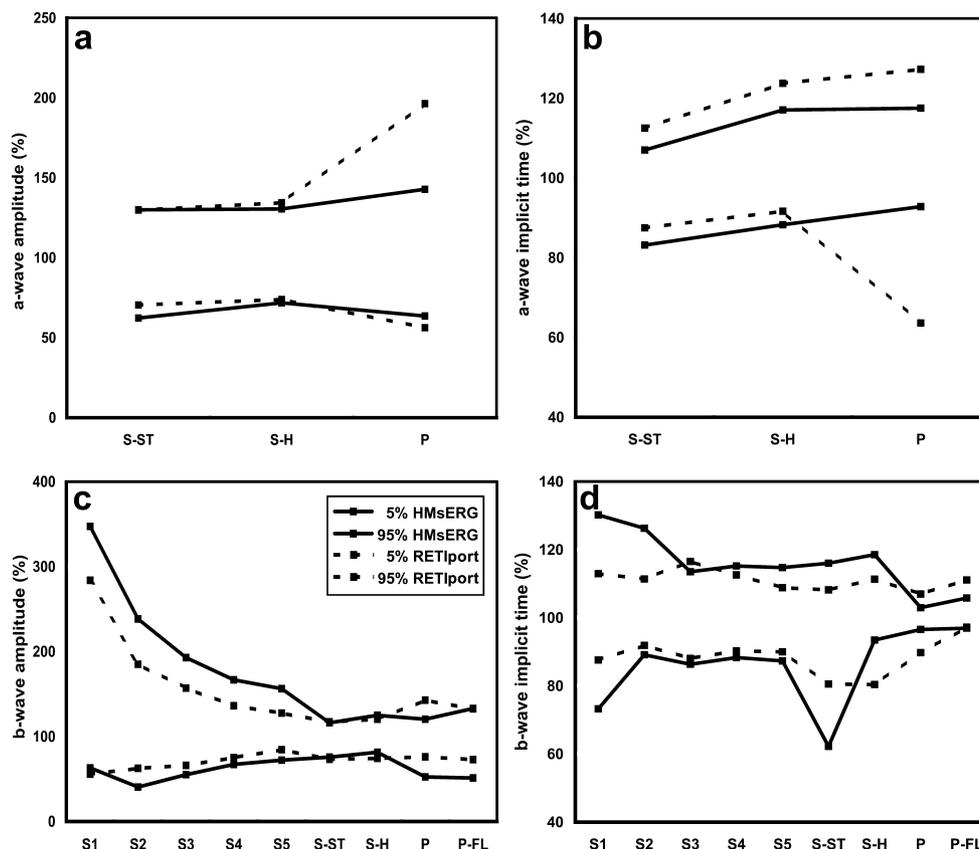


Fig. 4. Graphical illustration of the reference range depicts ERG differences between the HMsERG (a solid line) and the RETIport (a dotted line) using the 5th and 95th percentiles of the MoM distribution, which consist the 90% reference range. Amplitude and implicit time values on the vertical axis for the a- and b-waves, respectively, are shown as percentages of the values obtained by the two ERG units where 100% represents the median of these values. S1 to S5 depicts scotopic low intensity responses obtained using  $0.001 \text{ cd}\cdot\text{s}/\text{m}^2$  for HMsERG and  $0.025 \text{ cd}\cdot\text{s}/\text{m}^2$  for RETIport of light stimulation 4, 8, 12, 16, and 20 min, respectively after dark adaptation, designated S1, S2, S3, S4, and S5, respectively on the horizontal axis. S-ST and S-H depict scotopic standard intensity and scotopic higher intensity responses obtained at 3 and  $10 \text{ cd}\cdot\text{s}/\text{m}^2$  for HMsERG and 2.5 and  $7.9 \text{ cd}\cdot\text{s}/\text{m}^2$  for RETIport, respectively. Photopic ERGs consisted of a photopic single flash (P) response, and photopic 30 Hz flicker (P-FL) responses for the HMsERG and RETIport, respectively, depicted as P and P-FL at  $3 \text{ cd}\cdot\text{s}/\text{m}^2$  for the HMsERG and  $2.5 \text{ cd}\cdot\text{s}/\text{m}^2$  for the RETIport after 10 min of light adaptation using 30 and  $25 \text{ cd}/\text{m}^2$  of background light for the HMsERG and the RETIport, respectively.

equipment-related factors, some of which have been mentioned earlier.

Among the many ERG equipment-related factors affecting the results of the present study, the difference in the type of active electrodes used is probably of greatest importance. Previous studies in humans, dogs, and rodents have shown major ERG recorded variations in ERG parameters depending on type of active electrodes used [2, 19, 23, 35]. In the present study, the two active electrodes were very different: the Jet lens was a thin plastic, transparent corneal lens including a thin gold ring, while the Kooijman-Damhof contact lens was an opaque area covering the cornea attached to a clear corneal ring consisting of a circular silver wire, the latter acting as an active electrode. Further, the position of the eyelids was

different when using the two electrodes. The former was used together with a lid speculum, with the Jet lens positioned in the central part of the cornea, while the latter was centrally positioned on the cornea with the eyelids enclosing the electrode/photostimulator. Due to these differences less light reached the fundus prior to the ERG procedure using the RETIport when the active electrode/photostimulator was positioned on the eye. Therefore, the eye became more dark-adapted in comparison to the situation when using the HMsERG and the Jet lens. Thus, as shown in Fig. 3c, the RETIport started out with amplitudes that were higher in comparison to those recorded by the HMsERG. It is clear, however, that the light stimulation using low light intensity resulted in higher amplitude responses for the RETIport compared to the

HMsERG. These differences could be due to the shorter distance between the light stimulator and the retina and therefore a comparably more effective stimulation obtained for the former instrument.

When depicting the ERG results as a graphical representation using 90% reference ranges of the MoM, the change in ERG parameters obtained during the recording sessions using the HMsERG instrument was very similar to those recorded using the RETIport instrument for most ERG responses, except for a few (Fig. 4). Because some ERG parameters, such as b-wave amplitudes, are usually not distributed in a normal bell-shaped curve, calculations of the mean and standard deviation may be misleading. In order not to make such a statistical error, the ISCEV and the ECVO have recommended that the median and the 90% reference range be used to describe normal ranges of ERG parameters [22, 26]. Moreover, the ERG results obtained in the present study cannot be compared directly due to differences in equipment-related factors. Therefore, in order to show how the variability of the two ERG units might affect clinical recordings, the ERG results from the eye tested were plotted in figures and presented with medians and estimated limits of normality. As illustrated in Fig. 4, some major differences between the two units were shown. It is clear that if only one reference range for both ERG units was used, this would result in misdiagnosis of a retinal disease processes. For example, the b-wave amplitude for the HMsERG would be considered supernormal if the normative values for the RETIport (100  $\mu$ V) were used at S2. It therefore appears critical to establish ERG system-specific reference ranges, with limits of normality for each laboratory. Further, as previously described, a graphical representation of ERG results is very simple to apply for clinical purposes after data from groups of clinically normal animals of the same age and breed are obtained in order to differentiate between clinically normal and affected animals, such as those with hereditary retinal degenerative diseases [7, 25].

The differences between the photostimulators of the two ERG units could result in the different ERG results obtained. LEDs, incorporated in the Kooijman-Damhof contact lens electrode, are used as a light source for both stimulus and background lights (Fig. 1b) [16]. The light stimulator part contains four built-in high luminance white diodes. The contact lens part embraces a corneal ring electrode, in which the opalescent part of the lens (diameter=12 mm) has been made to work as a light diffusing lens. In theory, the combination of the LED array and the opaque corneal lens makes the distribution of the light rays which reach the cornea equal to the distribution of a Ganzfeld dome [12, 16, 21]. There is also a report that the Kooijman-Damhof contact lens under a Ganzfeld dome can illuminate the retina nearly even regardless of the pupil size [15]. Some researchers have, however, questioned the homogeneity of the retinal illumination when using such a LED-electrode [4, 23]. Additionally, Marmor *et al.* stated that it is more difficult to measure the extent and intensity of retinal illumina-

tion when using ocular diffusers other than Ganzfeld stimulators [22].

In general, it is not easy to directly compare ERG parameters recorded by different equipments in different clinics and laboratories, because various physiological and equipment-related factors affect the ERG recordings. Therefore, it is considered more reasonable to directly compare changes in ERG parameters obtained, which can be performed by developing ranges for normality in each species, breed and age group, using a graphical representation of results for each animal tested. This study demonstrates that the ERG results recorded by the HMsERG are mainly comparable to those of the RETIport for routine clinical use, except for a few parameters observed through a graphical representation of the median and the 90% reference range. As a result, it is recommended that reference ranges be developed for every type of ERG system, using the median and 90% reference range, and that methods for performing ERGs are standardized for each clinic and laboratory.

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