The Effects of Different Anesthetic Agents on Short Electroretinography Protocol in Dogs

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ABSTRACT. The purpose of this article was to investigate the effects of sedatives and general anesthetics, such as tiletamine-zolazepam, medetomidine, and isoflurane on the short ERG protocol. Six healthy mongrel dogs were assessed by a convenient short ERG protocol with the owners’ consent. The amplitudes of a-wave and b-wave, as well as the implicit time of ERG under different anesthesia statuses, were recorded and analyzed. The amplitudes of ERG waves were not significantly different between tiletamine-zolazepam and medetomidine groups, except in b-wave after 5 min dark adaptation (140 ± 42 μV in tiletamine-zolazepam and 101 ± 32 μV in medetomidine, p<0.01). The amplitude of ERG recorded in isoflurane (5 ± 3 μV of a-wave and 12 ± 6 μV of b-wave under light adaptation; 41 ± 19 μV of b-wave after 1 min dark adaptation; 28 ± 15 μV of a-wave and 58 ± 32 μV of b-wave after 5 min dark adaptation) were significantly different from tiletamine-zolazepam (8 ± 2 μV of a-wave and 24 ± 9 μV of b-wave under light adaptation; 117 ± 44 μV of b-wave after 1 min dark adaptation; 59 ± 18 μV of a-wave and 140 ± 42 μV of b-wave after 5 min dark adaptation), except in a-wave after 1 min dark adaptation (39 ± 13 μV in tiletamine-zolazepam and 34 ± 17 μV in isoflurane). Comment–General anesthesia had significantly lower amplitudes in the dark-adapted group compared with the sedation group. Therefore, tiletamine-zolazepam is a desirable choice for the short ERG protocol in dogs.

KEY WORDS: electroretinogram, isoflurane, medetomidine, tiletamine-zolazepam.


The retina is the most important tissue in the mammalian eye. Lesions in this part of the eye will therefore affect normal vision [15]. Because the retina is in the deepest portion of the eye, it is very difficult to examine without specialized instrumentation. The most widely used method for examination of the retina involves indirect ophthalmoscopy to inspect the morphology of the retina combined with electroretinography (ERG) to assess retinal function [15]. Indirect ophthalmoscopy is easy to perform after administration of mydriatic drops and is a valuable tool for assessing the health of the eye. However, lesions in the fundus are hard to spot, especially in the early stages of diseases, because the fundus is readily obstructed by other ocular diseases such as cataract and others. In addition, assessment of indirect ophthalmoscope images often depends on the subjective interpretation of the clinician.

Electroretinography (ERG) is a more objective technique for assessing the function of the retina. ERG allows for detection and diagnosis of abnormalities in the retina even in the early phase of diseases when there may be no macroscopic changes in the fundus [2]. It can also provide an accurate assessment in patients with severe cataract and whose fundus appears to be without abnormality [13]. A lot of research has been done on the effects of general anesthetics on electroretinography (ERG) in dogs but there are few data in the literature on the effects of sedatives on ERG results [1, 8]. There are 2 recommended ERG protocols, short and long ERG protocol, for use in dogs. The short ERG protocol is usually for quick examination to determine whether a response of retina is present or absent. This technique is usually adapted to quickly evaluate the retinal function; however, generalized photoreceptor disease is not able to be diagnosed because it does not have a complete test for the specific rod and cone functions [10]. The long ERG protocol is generally named a diagnostic ERG protocol since it offers more extensive ability to assess the outer retinal function. This diagnostic ERG protocol is useful for hereditary outer retinal disease [10]. Therefore, the short protocol is typically used for pre-operation before cataract surgery or in the differential diagnosis of specific blinding disorders such as in the acutely blinding disease Sudden Acquired Retinal Degeneration (SARD) in dogs.

For examination of animals using ERG, sedation or general anesthesia is strongly recommended to prevent unwanted movement since ERG is very sensitive to changes in retinal electrical current, up to 10⁻⁶ volts. Several studies have indicated that the use of different anesthetic agents may interfere with ERG examination results [3, 8, 17]. Yanase and Ogawa [17] noticed that the amplitude of the b-wave was decreased in dogs anesthetized with halothane. Chaudhary et al. [3] showed that rats anesthetized with tiletamine-zolazepam or pentobarbital had remarkable differences in ERG examination results. Kommonen et al. [8] demonstrated that dogs anesthetized with a fast injection of propofol had an obvious increase in the amplitude of the b-waves. In recent years, ketamine, xylazine, and medetomidine have been used as anesthetic agents for conducting...
ERG examinations [4, 12].

Tiletamine-zolazepam is an injectable anesthetic and sedative agent. It is a mix of 2 drugs, tiletamine and zolazepam, similar to the combination of ketamine and diazepam. In recent years, tiletamine-zolazepam has been the preferred choice for use in conducting clinical examinations and short duration surgeries due to its rapid induction and short recovery time [6]. To the best of the authors’ knowledge no previous studies have been conducted to investigate whether tiletamine-zolazepam affects the result of ERG examination in veterinary medicine. Furthermore, to date, a comparison among the effects of tiletamine-zolazepam, medetomidine, and isoflurane on ERG examination results has not been previously reported. Before surgical treatment for the clinically mature cataract, it is not possible to evaluate the basic vision of dogs and cats by any ophthalmological examination, except ERG. Therefore, the aim of the current study was to compare the effects of two sedatives and an anesthetic agent on the results of wave forms for the short protocol of ERG examination in a veterinary clinic.

MATERIALS AND METHODS

Two male and four female mongrels (2 to 7 years old; 11.7 ± 3.6 kg) requiring sedation or anesthesia for various therapeutic reasons, such as neutering, inguinal hernia repair, and so on, at National Chung Hsing University’s Veterinary Medical Teaching Hospital, were used in this study. With the owners’ consent, the short ERG examination was performed prior to the therapeutic procedure while sedated or anesthetized. The Slit lamp microscope and indirect ophthalmoscope were used for ophthalmologic examination a week before study for ruling out any underlying ophthalmic diseases. These dogs had the short ERG examination under 3 different anesthetic protocols at intervals of at least 2 weeks (tiletamine-zolazepam group, medetomidine group, and isoflurane group).

The ERG instrument used was a portable Eickemeyer™ ERG (Eickemeyer, Unit 4 Twickenham, Germany), which incorporates a stimulator, amplifier and recorder. We performed the Eickemeyer ERG examination according to the guidelines described by Narfstrom et al. [10]. Mydriatic drops (1% tropicamide, Mydriacyl®, Alcon, Belgium, UK) and atropine 0.04 mg/kg subcutaneous injection were given 30 min and 15 min before anesthesia and the local anesthetic drops (0.5% proparacaine hydrochloride, Alcaine®, Alcon, Belgium, UK) were given 5 min prior to the ERG test. In the isoflurane group, dogs were anesthetized with the standard flash. A complete ERG report has to include a-wave amplitude, b-wave amplitude, a-wave implicit time, and b-wave implicit time. The a-wave amplitude is measured from the baseline to the a-wave trough. The b-wave amplitude is measured from the a-wave trough to the b-wave peak. The a- and b-wave implicit time are measured from the stimulus onset to the a-wave trough and b-wave peak, respectively.

The duration of the procedure was measured from injection of sedatives or propofol injection to the completion of the ERG examination. The mean head-up time (MHT) and mean walking time (MWT) were measured and recorded throughout the procedure. The MHT and MWT were defined as the mean time for dogs showing head-up and walking movement after tiletamine-zolazepam injection, atipamezole injection for the reversal of medetomidine and turning off the isoflurane after finishing the short ERG protocol, respectively.

In this study, the paired t-test was used to analyze the ERG parameters and the non-parametric ANOVA with post hoc test was used to analyze MHT and MWT in three groups. A p value below 0.01 was considered significant.
RESULTS

Comparison of the effects of the two sedatives on ERG test: The ERG examination results revealed that there were no significant differences between tiletamine-zolazepam and medetomidine used as a sedative with regard to light adaptation and 1 min dark adaptation. However, the tiletamine-zolazepam group had the largest wave after 5 min of dark adaptation (Fig. 1). The results of the light adaptation showed that the average amplitude of b-wave was 19 ± 3 µV and 24 ± 9 µV in the medetomidine group and in the tiletamine-zolazepam group, respectively, but this was non-significant (p>0.01). The 1 min dark adaptation recording data indicated that the average amplitude of b-wave was 86 ± 32 µV and 117 ± 44 µV in the medetomidine group and in the tiletamine-zolazepam group, respectively, which was also non-significantly different (p>0.01). However, the 5 min dark adaptation results demonstrated that the average amplitude of b-wave in the medetomidine group was 101 ± 32 µV compared with 140 ± 42 µV in the tiletamine-zolazepam group, which was significantly different (p<0.01) (Fig. 2). In short, the amplitudes of ERG waves were not significantly different between sedation with tiletamine-zolazepam and with medetomidine, while medetomidine had a propensity to depress the ERG b-wave amplitude as seen in the significant difference recorded after 5 min dark adaptation.

The effects of general anesthesia compared with sedation on ERG test: A comparison of ERG results obtained using the sedative tiletamine-zolazepam and the general anesthetic isoflurane revealed significant differences between the two agents for light adaptation as well as for 1 min and 5 min dark adaptation (p<0.01) (Fig. 3). Moreover, our results indicated that the ERG amplitude of the sedative group was greater than that of the anesthesia group (Fig. 3). Under light adaptation, the average amplitude of a-waves in the sedative group and the anesthesia group was 8 ± 2 µV and 5 ± 3 µV, respectively, which was statistically significant (p<0.01). However, at 1 min dark adaptation, the average amplitude of a-waves in the sedative group and in the anesthesia group was 39 ± 13 µV and 34 ± 17 µV, respectively, which was non-significant (p>0.01). Interestingly, there was a significant difference between these two groups after 5 min dark adaptation (p<0.01) (Fig. 4). After 5 min dark adaptation, the average amplitude a-wave of the sedative group was 59 ± 18 µV compared with 28 ± 15 µV in the anesthesia group. Under light adaptation, the average amplitude of b-waves in the sedative group and in the anesthesia group was 24 ± 9 µV and 12 ± 6 µV, respectively, which was significant (p<0.01). The average amplitude of

![Fig. 1. Typical ERG tracings after 5 min dark adaptation recorded in a same dog sedated with medetomidine or tiletamine-zolazepam.](image)

![Fig. 2. The means of amplitude of the b-wave recorded in dogs sedated with medetomidine or tiletamine-zolazepam under different adaptation of light environment situations. The amplitudes of ERG waves were not significantly different between sedation with tiletamine-zolazepam and with medetomidine, except in b-wave recorded after 5 min dark adaptation.](image)
b-waves in the sedative group and in the anesthesia group was 117 ± 44 µV and 41 ± 19 µV under 1 min of dark adaptation, respectively, which was significantly different \((p<0.01)\). Furthermore, the average amplitude of b-waves in the sedative group and the anesthesia group was 140 ± 42 µV and 58 ± 32 µV after 5 min of dark adaptation, respectively, which was statistically significant \((p<0.01)\) (Fig. 4). In short, the amplitudes of ERG waves recorded during general anesthesia with isoflurane were significantly lower than those recorded during sedation with tiletamine-zolazepam, except in a-wave recorded after 1 min dark adaptation.

A comprehensive comparison of the three anesthetics: The ERG flash of the tested dogs in both medetomidine and isoflurane groups was influenced by the eyeball disposition towards ventromedial (reversed Bell’s phenomenon). In these groups, therefore, in all dogs both eyelids were opened with Barraquer wire speculum and received 3 stay suture on the bulbar conjunctiva to ensure cornea was oriented in the central position. Both sedation time of intravenous propofol injection and intravenous tiletamine-zolazepam injection used for the induction of isoflurane general anesthesia were less than 10 sec. But the duration of intramuscular medetomidine injection usually required 8.0 ± 1.5 min. Our results indicated that the duration of procedure using tiletamine-zolazepam was 43.5 ± 8.0 min (Table 1). Meanwhile, our study data showed that all three of the agents used were able to provide enough depth and duration of sedation/anesthesia to perform the short ERG examination. Our results also

Table 1. Comprehensive comparison of three sedative/anesthetic protocols for the short ERG examination

<table>
<thead>
<tr>
<th>Agents</th>
<th>Subconjunctival stay suture</th>
<th>ERG wave form</th>
<th>MHT (min) (mean ± SE)</th>
<th>MWT (min) (mean ± SE)</th>
<th>Recovery</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tiletamine-zolazepam</td>
<td>Not required</td>
<td>Big</td>
<td>43.5 ± 8.0⁹</td>
<td>57.3 ± 4.9⁹</td>
<td>longer</td>
</tr>
<tr>
<td>Medetomidine</td>
<td>Required</td>
<td>Medium</td>
<td>8.7 ± 1.8⁹</td>
<td>9.7 ± 1.4⁹</td>
<td>short after atipamezole</td>
</tr>
<tr>
<td>Isoflurane</td>
<td>Required</td>
<td>Small</td>
<td>3.8 ± 0.9⁹</td>
<td>20.7 ± 2.6⁹</td>
<td>moderate</td>
</tr>
</tbody>
</table>

MHT: mean time for dogs showing head-up after the ERG examination completed.
MWT: mean time for dogs showing walking movement after the ERG completed.
a,b) the different characters mean that there are significantly different between two groups \((p<0.01)\).
revealed that the recovery time of medetomidine after atipamezole injection was shorter than that of isoflurane or tiletamine-zolazepam. From our results, the MHT and MWT of tiletamine-zolazepam were significantly longer than that of medetomidine and isoflurane \( p<0.001 \) (Table 1). However, the MHT and MWT of medetomidine and isoflurane did not reveal any significant differences \( p>0.001 \) (Table 1).

**DISCUSSION**

In this study, we successfully completed the short ERG examination under three sedative/anesthetic protocols in dogs. Each sedative/anesthetic protocol had different features during examination and varying effects on the amplitudes of ERG waves. Our study showed that the amplitudes of ERG waves tended to be depressed in dogs sedated with medetomidine compared to tiletamine-zolazepam. Furthermore, the waves were significantly more depressed in dogs anesthetized with isoflurane than in those sedated with tiletamine-zolazepam. The mechanism and effect of tiletamine-zolazepam (sedation and anesthetic with zolazepam and tiletamine 1:1 mixture) is very similar to that of diazepam and ketamine \[11\]. The ERG usually records activity in the outer retina. However, the mechanism and effect of medetomidine is similar to xylazine, which is an \( \alpha_2 \)-adrenoreceptor agonist \[14\]. The reflect activity of bipolar cells is read as the ERG b-wave. Some study results indicated that the rat and rabbit b-wave could be generated by depolarizing the bipolar cells of both animals. One possible mechanism might be that the activated bipolar cells were able to release the excitatory transmitter glutamate \[8\]. In our study, medetomidine had a greater propensity to depress the ERG b-wave amplitude compared to tiletaminie-zolazepam as a significant difference in b-wave was recorded after 5 min dark adaptation. We postulate that this may be due, in part, to the differing effects of the agents on retinal nerve conduction. The main function of zolazepam is to produce a nerve conduction suppressant (\( \gamma \)-aminobutyric acid, GABA) in the retina \[3\]. The sites of its main inhibitory action are in pyramidal cell (cone cell), double-pole cell (bipolar cell) and ganglion cell (ganglion cell) \[9\]. However, tiletamine exerts its effect mainly by blocking the ion channel of the N-methyl-D-aspartate (NMDA) receptor and increasing the activity of the double-pole cell and ganglion cell \[3\]. Therefore, tiletamine-zolazepam may regard as these two medicines to assemble the response of ERG examines. The large ERG wave is very helpful for providing an accurate assessment of the remaining retinal function in patients with severe cataract, although the magnified wave might be enhanced by tiletamine-zolazepam.

Comparing the results of ERG obtained with isoflurane (general anesthesia group) and tiletamine-zolazepam (sedative group), the sedative group had a higher a-wave amplitude than that of the general anesthesia group after dark adaptation. The sedative group had higher b-wave amplitudes under light adaptation as well as after dark adaptation than those in the general anesthesia group. The reason may be due to the different abilities of the two agents to suppress the central nervous system and to interfere with nerve transmission in the retina \[3, 17\]. The main mechanism of tiletamine-zolazepam in sedative dosage is to inhibit the thalamus limbic system and the function of hypothalamus, to reduce the tone of sympathetic nerves, and to slightly inhibit the action of the central nervous system. However, the main mechanism of isoflurane is to make a reversible inhibitory action in the central nervous system and to cause loss of consciousness by blocking external stimulation \[16\]. Therefore, isoflurane may affect the mechanism of potential action and change the ERG wave. Yanase et al. \[17\] indicated that halothane and sevoflurane were able to depress the b-wave, influence nerve conduction, and create a changed ERG wave.

Dogs are usually anesthetized or sedated to avoid distress and prevent unwanted movement during ERG measurement. As described above, sedation with tiletamine-zolazepam was able to provide larger ERG waves during the short ERG protocol in dogs, compared to sedation with medetomidine and general anesthesia with isoflurane. Furthermore, subconjunctival stay sutures are required in dogs sedated with medetomidine and in dogs anesthetized with isoflurane to fix the eyeball in the central position. However, sedation with tiletamine-zolazepam produced longer duration of recovery in this study. This delayed recovery may be shortened with flumazenil, a benzodiazepine antagonist. Flumazenil should be injected at 45 to 60 min after the administration of tiletamine-zolazepam in dogs \[7\]. Medetomidine provides a quick recovery when antagonized with atipamezole, however, it should be administered to healthy young animals free from cardiovascular disease because it has severe cardiovascular side-effects \[5\]. Isoflurane achieves a safe and stable anesthesia when animals are intubated and anesthesia is maintained with appropriate cardio-respiratory management. Isoflurane may be a viable choice for sick and/or elder animals requiring diagnostic ERG examination, i.e., long ERG protocol. In conclusion, our findings suggest that tiletamine-zolazepam may be an appropriate sedative agent for the canine short ERG examination in a clinical setting.

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**REFERENCES**

4. Chaudieu, G. and Molon-Noblot, S. 2004. Early retinopathy in...