Evaluation of Alternative Anesthetics for Electroretinogram (ERG) Recording in Rats

Kelly Tenneson , Christian Li , Mark Vezina
Charles River Montreal

1 Introduction

Electroretinograms are a commonly used endpoint in non-clinical safety studies to evaluate the potential effects of new chemical entities on retinal function. In animal models, this assessment must be done while the animals are anesthetized; therefore, selection of the anesthetic agent and dose is critical to obtain suitable results. Some of the factors that must be taken into consideration are the length of the testing protocol relative to duration of the anesthesia, the anesthetic agent (as some can interfere with or modify ERG response), and inter-animal sensitivity to anesthesia. We characterized the effect of multiple anesthetic agents on ERG responses and to evaluate duration of anesthesia of the effect in rats.

2 Methods

Albino rats (up to 5 males/group; 100 to 300 g and approximately 6 to 12 weeks of age) were dark adapted overnight and administered one of 4 different anesthetic cocktails by intramuscular (IM) or intraperitoneal (IP) injection:

Group	KX↓	KX↑	KXA	KA
N =	3	5	4	4
Route	IM	IP	IM	IP
Ketamine	50 mg/kg	75 mg/kg	40 mg/kg	100 mg/kg
Xylazine	5 mg/kg	7.5 mg/kg	2.5 mg/kg	-
Acepromazine	-	-	0.75 mg/kg	5 mg/kg

Topical anesthetic and mydriatic drops were administered to each eye, and 1% CMC drops applied to maintain contact with the corneal electrode. Each animal was then positioned in a Ganzfeld dome and subjected to the following ERG protocol:

Step	Log scale (dB)	Flash intensity ($cd \cdot s/m^2$)	Number to average	Time between flashes
1	-30	0.0025	5	10 seconds
2	-10	0.25	5	15 seconds
3	0	2.5	2	Minimum 2 minutes

3 Results

Group	KX↓	KX↑	KXA	KA
Duration of anesthesia (minutes)	129 ± 15	79 ± 7	105 ± 15	134 ± 17

Anesthesia duration was significantly shorter with KX↑ given IP (79 ± 7 min), despite the higher dose level when compared to KX↓. This was likely due to a slower biodistribution when given IM. Animals given KX↓, KXA and KA required long recovery times (>1.5 h).

Figure 1. Amplitude A-Wave

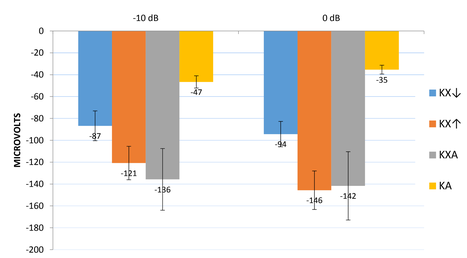


Figure 2. Amplitude B-Wave

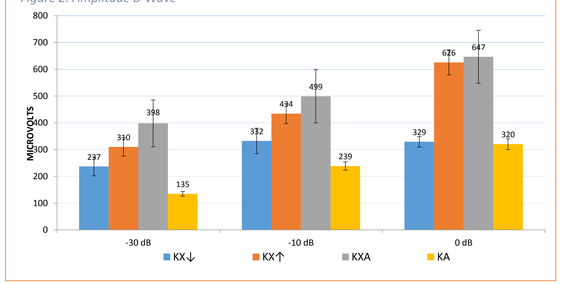


Figure 3. Implicit time A-Wave

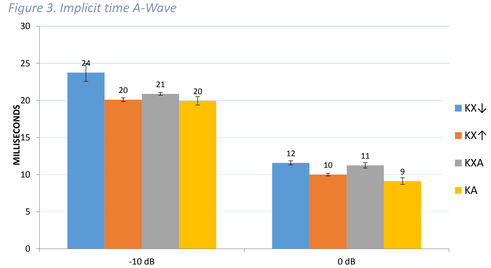
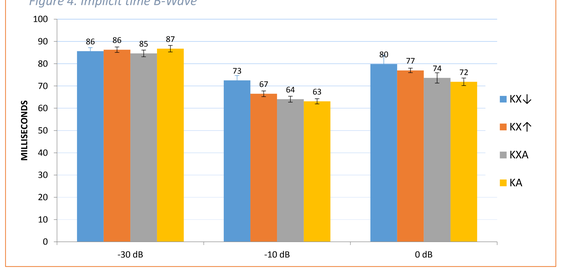


Figure 4. Implicit time B-Wave



The anesthetic used was shown to have a major impact on the amplitude of the ERG responses. KX↑ and KXA scotopic 0 dB b-wave amplitudes (626 and 647 uV, respectively) were approximately 2-fold higher than the response when KX↓ or KA (329 and 320 uV, respectively). A similar difference was observed with all stimulus intensities tested. Latency times were generally unaffected.

4 Conclusion

The anesthetic agent used for ERG recording has a significant impact on the magnitude of the response, which needs to be taken into consideration during the study design phase of nonclinical studies.

- Agents that reduce ERG response can reduce the sensitivity of the test and can mask potential changes.
- The agent used needs to be taken into consideration when comparing between results in different studies. Ideally, comparisons should be made to concurrent controls.
- Anesthesia duration is an important factor: Prolonged sedation periods can result in corneal drying and poor recovery.

In this study, KX↑ ketamine 75mg/kg, xylazine 7.5 mg/kg) given by IP injection provided a good ERG response with an appropriate length of anesthesia.

Acknowledgements
The authors would like to acknowledge the contributions made by the technical staff in the Ocular and Neuroscience department at Charles River Montreal.