Journal of Pharmacological and Toxicological Methods xxx (2009) xxx–xxx

Contents lists available at ScienceDirect

Journal of Pharmacological and Toxicological Methods

journal homepage: www.elsevier.com/locate/jpharmtox

Original article

Video-electroencephalography in conscious non human primate using radiotelemetry and computerized analysis: Refinement of a safety pharmacology model ☆

S. Authier [a,b,*], D. Paquette [a], D. Gauvin [b], V. Sammut [a], S. Fournier [a], F. Chaurand [a], E. Troncy [b]

[a] LAB Research Inc., 445 Armand Frappier, Laval, Quebec, Canada H7V 4B3
[b] GREPAQ — Department of Veterinary Biomedicine, Faculté de médecine vétérinaire, Université de Montréal, P.O. box 5000, St-Hyacinthe, Quebec, Canada J2S 7C6

ARTICLE INFO

Article history:
Received 10 November 2008
Accepted 23 December 2008
Available online xxxx

Keywords:
Electroencephalography
Monkey
Pharmacology
Safety
Conscious
Seizure
Pentylenetetrazole

ABSTRACT

Introduction: Electroencephalography (EEG) investigations are occasionally required as follow-up studies for safety pharmacology core battery (S7A). Video-EEG monitoring is a standard diagnostic tool in humans but limited data is available on its use in telemetered freely moving macaque monkeys for safety pharmacology investigations. While proconvulsant risk evaluations are routinely conducted in rodents, pharmacological or pharmacokinetic considerations lead to the use of non human primates in toxicology and safety pharmacology in some cases. **Methods:** Cynomolgus monkeys were instrumented with telemetry implants. Placement of EEG electrode was based on the 10–20 system using three derivations (C3-O1, Cz-Oz and C4-O2). EEG trace analysis was carried out using NeuroScore software. After 24 h of continuous video-EEG monitoring, animals received pentylenetetrazole (PTZ, 10 mg/kg/15 min) until convulsions were noted. Convulsions were immediately treated with diazepam (1.0 mg/kg). A seizure detection protocol with a dynamic spike train threshold was used for the entire EEG monitoring period (total of 44 h) including periods when PTZ was administered. Spectral analysis was done to quantify the absolute and relative amplitude of EEG frequency bands (delta, theta, alpha, sigma and beta waves). Sleep stages were quantified and EEGs during seizures were analyzed using fast Fourier transformation (FFT) to assess dominant frequencies. **Results:** Spike trains were detected by computerized analysis in all animals presenting PTZ-induced seizures while paroxysmal activities were systematically predictive (at least 4-min prior to generalized seizures). Beta activity increased with visual stimulation using monkey treats. Characteristics of EEG for all sleep stages (I, II, III and IV) were present in all animals. Delta activity was predominant in normal awake EEG as well as in all sleep stages. Seizure peak frequency was 3–6 Hz on FFT, corresponding to the discharge of the underlying generator. **Discussion:** EEG-video monitoring can be useful when using non human primates to characterize neurological adverse effects with unpredictable onset. Computerized video-EEG analysis was a valuable tool for safety pharmacology investigations including proconvulsant risk assessment, spectral analysis of frequency bands and sleep stage determination.

1. Introduction

Neurological and neuromuscular adverse effects are seen with a number of new drug candidates and characterization of the underlying pathophysiological mechanisms is critical for the future of the drug candidate. A drug that induces life-threatening adverse effects such as generalized seizures or *status epilepticus* may lead to drug development discontinuation. It is critical to differentiate CNS adverse effects from peripheral toxicity. Uncontrolled muscular activities can result from centrally mediated toxicity (*e.g.* seizures) or from neuromuscular transmitter disorder as seen with physostigmine, a cholinesterase inhibitor (Ambrani & Van Woert, 1972). While cholinesterase activity level can be investigated *in vivo* (Thomsen, Kewitz, & Pleul, 1988), it may be difficult to differentiate involuntary skeletal muscle contractions caused by other neuromuscular disorders from simple partial seizures when using neurological examination in safety pharmacology studies. Drug-induced muscular contractions can originate from central or peripheral neurological alterations and classification of adverse events can be a complex and challenging task (Lahunta, Glass & Kent, 2006.) The challenge is even greater considering that the timing of adverse effects is often unpredictable. Electroencephalography (EEG) has been used in safety pharmacology to assess the proconvulsant risk in various species (Danielsson et al., 2006; Durmuller, Guillaume, Lacroix, Porsolt, & Moser, 2007). While

☆ Experimental work was undertaken at: LAB Research Inc., 445 Armand Frappier, Laval, Québec, Canada H7V 4B3.

* Corresponding author. LAB Research Inc., 445 Armand Frappier, Laval, Quebec, Canada H7V 4B3. Tel.: +1 450 973 2240x1403; fax: +1 450 973 2259.
E-mail address: authiers@labresearch.com (S. Authier).

1056-8719/$ – see front matter © 2009 Elsevier Inc. All rights reserved.
doi:10.1016/j.vascn.2008.12.003

proconvulsant risk evaluations are routinely conducted in rodents, pharmacological or pharmacokinetic considerations lead to the use of non human primates in some cases. The use of monkeys is often justified by pharmacokinetic and metabolic profiles that are comparable to humans when rodents are not a suitable model (Oldham et al., 1990). The use of radiotelemetry with single (Pearce, Crofts, Muggleton, & Scott, 1998) and dual (Almirall et al., 1999) EEG channel has been reported in non human primates for neurobehavioral studies. Video-EEG monitoring is a standard diagnostic tool in humans (Asano et al., 2005), but limited data is available on its use in telemetered freely moving macaque monkeys for safety pharmacology investigations. Electrophysiology examinations are occasionally required as follow-up studies for safety pharmacology core battery (U.S. FDA, 2001). The aim of the current study was to qualify a telemetered non human primate model of continuous video-EEG monitoring and computerized EEG analysis including seizure detection. Pentylenetetrazole (PTZ) is a well characterized agent inducing seizures in humans (Danielsen & Ellebjerg, 1966) and a variety of species including macaque monkeys (David & Grewal, 1977) that allowed qualification of this safety pharmacology model.

2. Materials and methods

2.1. Statement on use and care of animals

During the study, care and use of animals were conducted in accordance with principles outlined in the current Guide to the Care and Use of Experimental Animals published by the Canadian Council on Animal Care and the Guide for the Care and Use of Laboratory Animals published by the US National Institutes of Health (NIH Publication No. 85-23, revised 1996). LAB Research Inc.'s facility is AAALAC accredited. All procedures were conducted as per Standard Operating Procedures (SOPs) in place.

2.2. Animal housing and preparation

Three (3) cynomolgus (*Macaca fascicularis*) monkeys were surgically prepared with telemetry transmitters (TL11M2-D70-EEE™, DSI, St-Paul, MN, USA). Monkeys were 3 to 4 years old and weighed between 3.7 and 4.3 kg. The animal room environment was controlled (temperature 21 ± 3 °C, humidity 30–70%, 12 h light, 12 h dark, 10–15 air changes per hour) and temperature and relative humidity were monitored continuously. A standard certified commercial primate chow (Certified Primate Diet 2055C™, Harlan Teklad, Madison, WI, USA) was available to each monkey twice daily. Prophylactic antibiotics (cefazolin 25 mg/kg) were administered by intramuscular (IM) injection at least 30 min prior to surgery and every 4–8 h post-injection for at least 48-h post-surgery. Preemptive analgesia (buprenorphine, Temgesic™, 0.05 mg/kg, Schering-Plough, Welwyn Garden City, Hertfordshire, UK) was administered by IM injection before surgery and every 6 to 12 h for at least 48 h post-surgery. Animals were placed on a heating pad and inhaled a mixture of oxygen (O_2) and isoflurane (AErrane™, Baxter Corporation, Mississauga, ON, CAN) with the O_2 flow meter and the vaporizer set at 1.0 L/min, and 2.0%, respectively. Respiratory rate was maintained between 10 and 13 breaths/min with an inspiratory airway pressure between 15 and 20 cm H_2O using a mechanical ventilator (2002, Hallowell EMC, Pittsfield, MA, USA). During anesthesia, monitoring included heart rate and pulsatile hemoglobin saturation in O_2 (VetOx 4404™ pulse oximeter, Heska, Fribourg, Switzerland). Bipolar centro-occipital and temporal-occipital EEG derivations were selected to minimize electromyographic (EMG) artefacts as previously described with needle electrodes in macaque monkeys (Danielsson et al., 2006). An abdominal midline skin incision was initially done cranial to the umbilicus and a longitudinal incision was done in the middle of the *rectus abdominis* muscle. The telemetry transmitter was placed between the *internal abdominal oblique* muscle and the aponeurosis of the *transversus abdominis* muscle. The *rectus abdominis* was sutured with a simple continuous suture and EEG electrodes were tunnelled subcutaneously to a small skin incision in the neck. The abdominal skin incision was closed with interrupted buried sutures and the animal was placed in sternal recumbency to expose the cranium for the remainder of the surgery. Electrode placement was based on the internationally standardized 10–20 system using three derivations (C3-O1, Cz-Oz and C4-O2) (Sharbrough et al., 1991). Incision and electrode placement sites on the cranium were measured and marked with a surgical skin marker before the surgery to ensure precision. A sagittal incision was done in the occipital region over Oz and allowed visualization of O1 and O2. Then, a transverse incision was done over C3, Cz and C4. The temporal muscle at the level of C3 and C4 was visualised and incised parallel to muscle fibers on each side to expose the cranium. A small groove-shaped hole (approximately 3 mm) was drilled through the skull at the level of C3, Cz, C4, O1, Oz and O2 and the electrodes were inserted in the holes and secured with surgical glue (Vetbond™, 3M, St-Paul, MN, USA). The holes were subsequently filled with polymethyl methacrylate. The temporal muscles and the skin were sutured. Immediately at completion of surgery, a local analgesic (Marcaine® E, 2.5 mg/mL, Hospira, Montreal, QC, CAN) was injected in 6–10 sites (0.2 mL/site) around surgical areas on the skull.

2.3. Experimental methods

The EEG-video monitoring included digital color camera with daylight and infrared night vision connected to a computerized system (IBM Intellistation Z pro, Xeon 3.8 GHz, 3.5 TB hard drive). The video recording was used for observational evaluation of behavioral convulsions both in real-time for diazepam administration and *a posteriori* when EEG traces were interpreted. To maximize telemetry signal quality, two receivers (DSI model RMC-1) were placed in each monkey cage (top and bottom). Acquisition of the telemetry signal was done at sampling rate of 500 Hz with the DSI software (Dataquest A.R.T. 3.01 Gold™) while the frequency range of the telemetry transmitters (D70-EEE, DSI) was 1–100 Hz. Video and EEG were monitored continuously for 24 h to establish baseline prior to subcutaneous injection of PTZ (10 mg/kg, 0.1 mL/kg) approximately every 15 min until seizures were noted. As soon as clonic convulsions were noted on video, a single diazepam injection was administered IV (1.0 mg/kg) and was sufficient to terminate seizure and paroxysmal EEG activity in all animals. Video and EEG were recorded for an additional 20 h after diazepam administration. Food treats were presented to induce a visual stimulation during baseline EEG monitoring to induce CNS stimulation.

2.4. EEG analysis methods

Trace analysis of EEG was carried out using NeuroScore software Version 1.1-2242 (DSI, St-Paul, MN, USA). A seizure detection protocol was created for cynomolgus monkeys with a dynamic spike train threshold which revealed to be the more sensitive and more specific than absolute amplitude threshold for seizure detection in our experimental conditions. The protocol was applied to the entire EEG monitoring period including periods when PTZ was administered. All EEG segments classified as spike trains by the software were evaluated by a trained reviewer and correlated with video for interpretation. All EEG traces after initiation of PTZ administration but before seizure onset were also reviewed to evaluate the presence of any undetected EEG spike train. Spectral analysis was also done at different intervals to quantify the absolute and relative amplitude of EEG frequency bands (delta, theta, alpha, sigma and beta waves). EEG characteristics of various sleep stages were identified (I, II, III or IV) and combined with behavior evaluations (video) and spectral analysis for sleep stage classification. Sleep stages III and IV also known as slow-wave sleep (SWS) Finally, EEG during seizures was analyzed using fast Fourier

Please cite this article as: Authier, S., et al., Video-electroencephalography in conscious non human primate using radiotelemetry and computerized analysis: Refinement of..., *Journal of Pharmacological and Toxicological Methods* (2009), doi:10.1016/j.vascn.2008.12.003

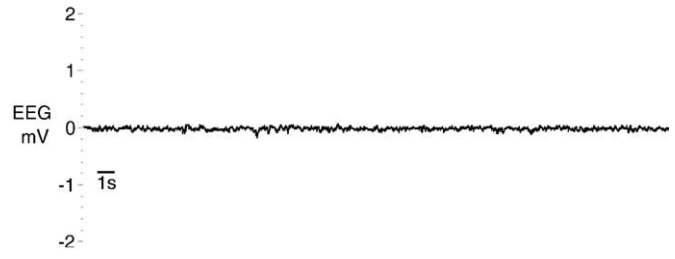

Fig. 1. Electroencephalogram in Cz-Oz derivation from a normal awake freely moving cynomolgus monkey.

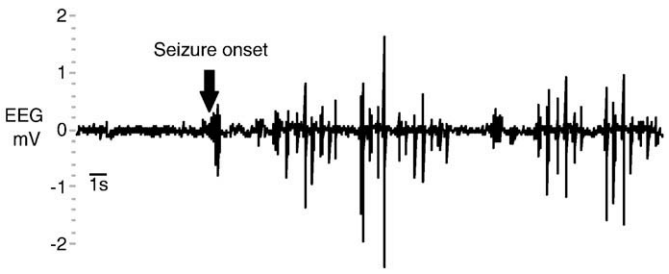

Fig. 3. Spike train detected using seizure detection module of the NeuroScore software in freely moving telemetered cynomolgus monkeys obtained from EEG Cz-Oz derivation.

transformation (FFT) and auto-regression spectrum to assess dominant frequencies. When applicable, data is presented as mean ± standard deviation.

3. Results

Telemetry implants and EEG electrodes were well tolerated in all animals and the quality of EEG traces was adequate throughout the monitoring period (Fig. 1). The most common artefact noted during behavioral evaluations from video analysis was EMG activity. Telemetry signal strength was above 25 MKUs throughout the monitoring period, which was considered optimal to minimize artifacts. When present, EMG activity was most important for derivations C3-O1 and C4-O2 while Cz-Oz (Fig. 1) presented significantly less EMG activity. Paroxysmal activity (Fig. 2) was noted at least 4 min before general seizure in all animals. Behavioral clonic convulsions were observed in all animals (average cumulative PTZ dose of 70 mg/kg ± 17 mg/kg) and correlated with EEG spike trains. Animals presented jerky movements before onset of generalized clonic convulsions corresponding to paroxysmal activity. The NeuroScore software seizure detection module identified spike trains during seizure in all animals (Fig. 3). *A posteriori*, video was used to differentiate jerky movements and clonic convulsion from artefacts such as EMG which may present similitude with EEG paroxysmal activity. Software sensitivity to detect spike trains was high (EEG review confirmed that all spike trains had been identified) while specificity was low (large number of EEG segments identified as spike trains by the software that were excluded following video and EEG review). Computer analysis significantly reduced the amount of EEG traces to review for spike trains (less than 5 min per period of 24 h). Transient background attenuation characteristic of the post-ictal EEG was noted (Fig. 4) in all animals and resulted in increased delta wave amplitude and attenuation of higher EEG frequencies (Fig. 5C). Spectral analysis of EEG throughout the monitoring period revealed that delta activity was predominant in all periods in the cynomolgus monkey (Fig. 5A to E). Visual stimulation resulted in a transient increase in beta activity (Fig. 5B). All sleep stages (I, II, III and IV) were present in all animals. A relative increase in beta activity with decreased alpha activity was noted during stage I sleep (Fig. 5D) while the presence of sleep spindles and K complexes (Fig. 6) were characteristic of stage II sleep. Increased delta activity (slow-wave sleep) was present in stages III (>20% of epoch) and IV (>50% of epoch) sleep stages (Figs. 5E, 7). During seizure, FFT revealed peak amplitude of 3–6 Hz corresponding to the discharge frequency of the underlying generator. Several peaks at integer multiples were also present but the higher harmonics could be distinguished from the fundamental frequency (Fig. 8).

4. Discussion

A trend to develop an increasing number of biologic drug candidates (Hughes, 2008) may justify the use of non human primate for safety investigations for various reasons including the presence of drug target in this species not found in rodents or other animal species. Some drugs administered orally have a relatively slow absorption rate and several hours may elapse between drug administration and the onset of adverse effects. In these cases, it may not be possible or ethical to restrain monkeys for EEG monitoring until an adverse event is noted. Similarly to clinical diagnostic, EEG-video monitoring can be useful when using non human primates to characterize neurological clinical signs with unpredictable onset. Continuous EEG monitoring using telemetry generates an important amount of tracings which may render manual EEG review impractical. The use of computerized EEG analysis is essential to optimize data processing and facilitate evaluation.

In the current study, PTZ was administered at 15 min interval until clonic convulsions were noted to confirm the method of computerized seizure detection. The software accurately identified spike trains in all animals from 44 h of EEG tracings which facilitated data processing. It remains that correlation of spike trains with behavioral activity recorded on video by a trained reviewer was required for interpretation of EEG traces. Dürmüller et al. presented paroxysmal activity as an adequate endpoint to assess the proconvulsant risk of drug candidates in telemetered dogs with PTZ infusion (Durmuller et al., 2007). Dürmüller reported that the onset of paroxysmal EEG activity was

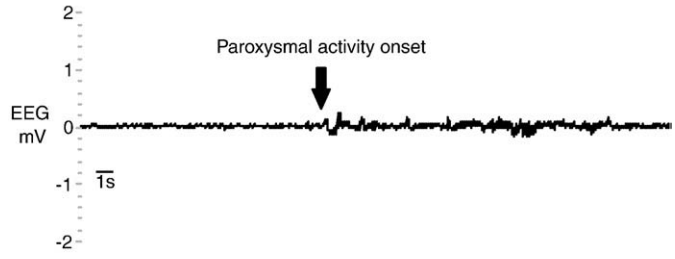

Fig. 2. Illustration of paroxysmal EEG activity in telemetered cynomolgus monkey.

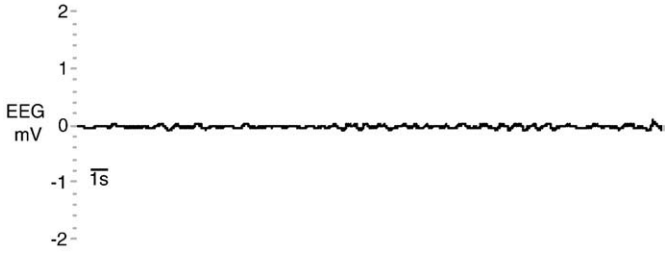

Fig. 4. Post-ictal EEG with background attenuation in Cz-Oz derivation following diazepam injection at onset of seizures in cynomolgus monkey.

Please cite this article as: Authier, S., et al., Video-electroencephalography in conscious non human primate using radiotelemetry and computerized analysis: Refinement of..., *Journal of Pharmacological and Toxicological Methods* (2009), doi:10.1016/j.vascn.2008.12.003

Fig. 5. Spectral analysis of EEG in cynomolgus monkey in different states: A) normal awake, B) increased beta activity during visual stimulation C) post-ictal EEG background attenuation with increased delta activity, and attenuation of higher EEG frequencies, D) stage I sleep with enhanced beta activity, E) stages III and IV sleep with increased delta activity and attenuation of higher EEG frequencies.

during the 60 s preceding convulsions. The delay between paroxysmal activity and clonic convulsion is critical to use this early EEG sign for evaluation of proconvulsant risk during real-time monitoring. Occasional EMG artefacts may require careful EEG tracing assessment to confirm the presence of paroxysmal activity. The current study allowed identification of paroxysmal activity at least 4 min before

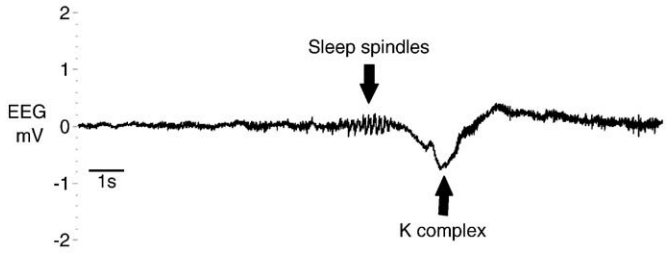

Fig. 6. Electroencephalogram from Cz-Oz derivation during stage II sleep in a cynomolgus monkey.

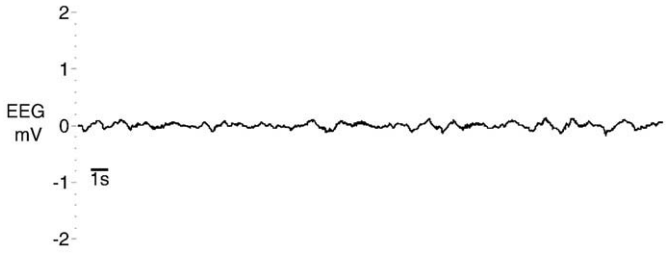

Fig. 7. Electroencephalogram from Cz-Oz derivation during deep sleep (Stage IV) presenting increased delta wave (>75 μV) activity in a cynomolgus monkey.

Please cite this article as: Authier, S., et al., Video-electroencephalography in conscious non human primate using radiotelemetry and computerized analysis: Refinement of..., *Journal of Pharmacological and Toxicological Methods* (2009), doi:10.1016/j.vascn.2008.12.003

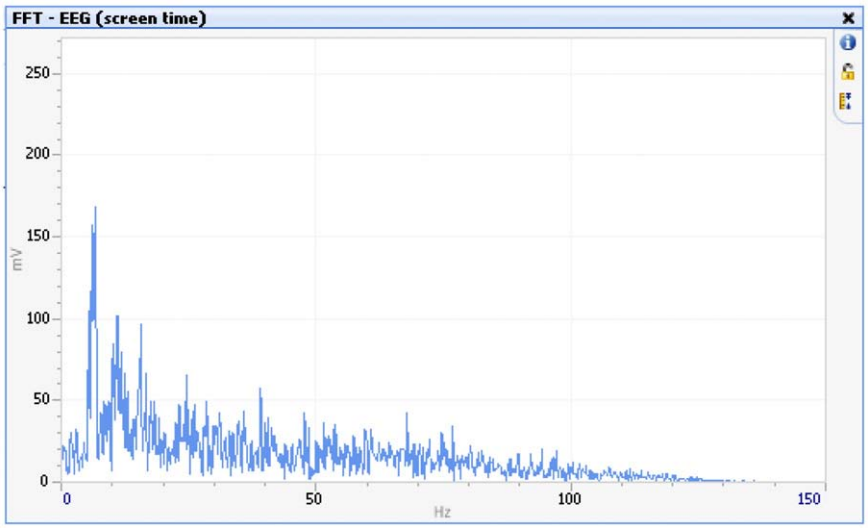

Fig. 8. Fast Fourier Transform of EEG during seizure with peak amplitude at a frequency of 6 Hz.

generalized seizures. The delay between paroxysmal activity onset and generalized convulsions when using SC injection at 15 min intervals may facilitate the use of this early sign as an indication of the proconvulsant risk while minimizing the overall duration of physical restraining. In contrast, the repeated PTZ administration at fixed doses SC decreases the precision and sensitivity of the proconvulsant model when compared with continuous IV infusion where the PTZ administration can be terminated and the total dose calculated. The current model could be further refined with continuous IV infusion in unrestrained non human primates. As seen in humans (Tilz et al., 2006) and other species (Cherubini, Gonella, Mancia, Mecarelli, & Tassinari, 1981), post-ictal EEG was characterized by background attenuation. A single diazepam IV administration (1.0 mg/kg) was sufficient to control and terminate seizure and paroxysmal EEG activity in all animals. Based on immediate resolution of paroxysmal activity, it is unlikely that brain trauma had occurred.

Alpha activity is predominant in normal awake EEG in adult humans while lower frequencies are predominant during rapid eye movement (REM) sleep (Morisson et al., 1998). Delta activity was predominant in awake as well as all sleep stages in our experimental conditions. While earlier report suggested predominant sigma activity in awake cynomolgus monkey (Jurko & Andy, 1967), recent spectral analysis in normal awake cynomolgus monkeys supports our observation that delta activity is predominant in this species (Lallement et al., 1998). In human, an increase in beta EEG activity during performance of a mental task is well described (Poupard, Sartene, & Wallet, 2001). The same observation was present in the monkeys of our study with visual stimulation (food treats). Several marketed drugs including benzodiazepines (Bastien, Leblanc, Carrier, & Morin, 2003), anti-convulsants (Wu & Xiao, 1996), anti-depressants (Feige et al., 2002) and Alzheimer's disease treatments (Gianotti et al., 2008) have known effects on EEG spectral analysis. Video-EEG monitoring can be a useful tool to further characterize drug candidates with central effects. The peak EEG frequencies (3–6 Hz) observed during PTZ-induced seizures were similar to reported values in dogs (4–5 Hz) (Durmuller et al., 2007) and rats (1–3 Hz) (Mirski, Tsai, Rossell, Thakor, & Sherman, 2003) and humans (3 Hz) (Rowan & Tolunsky, 2003).

In conclusion, video-EEG with computerized analysis was a valuable tool for safety pharmacology investigations including proconvulsant risk assessment, spectral analysis of frequency bands and sleep stage determination. In spite of considerable increase in efficiency for data processing, EEG review by a trained observer remains essential for definitive interpretations of traces. When evaluating drug candidate proconvulsant risks, species characteristics must be carefully considered. While most studies have used rats and mice, pharmacological and pharmacokinetic considerations may support the use of non human primates in some cases and this pilot study establishes standards of video-EEG with computerized analysis in cynomolgus monkeys.

Acknowledgements

We thank Caroline Charette for her technical assistance.

Simon Authier is a Ph.D. student funded by the Natural Sciences and Engineering Research Council of Canada Graduate Scholarships and the Faculty of Graduate and Postgraduate Studies of Université de Montréal, Québec, Canada.

Eric Troncy is a member of a New Emerging Team program (#108291) of the Canadian Institutes of Health Research/Heart and Stroke Foundation of Canada.

References

Ambrani, L. M., & Van Woert, M. H. (1972). Modification of the tremorigenic activity of physostigmine. *British Journal of Pharmacology*, *46*, 344—347.

Almirall, H., Pigarev, I., De la Calzada, M. D., Pigareva, M., Herrero, M. T., & Sagales, T. (1999). Nocturnal sleep structure and temperature slope in MPTP treated monkeys. *Journal of Neural Transmission*, *106*, 1125—1134.

Asano, E., Pawlak, C., Shah, A., Shah, J., Luat, A. F., Ahn-Ewing, J., et al. (2005). The diagnostic value of initial video-EEG monitoring in children-review of 1000 cases. *Epilepsy Research*, *66*, 129—135.

Bastien, C. H., Leblanc, M., Carrier, J., & Morin, C. M. (2003). Sleep EEG power spectra, insomnia, and chronic use of benzodiazepines. *Sleep*, *26*, 313—317.

Cherubini, E., Gonella, J., Mancia, D., Mecarelli, O., & Tassinari, C. A. (1981). Effects of PTZ-induced generalized epilepsy on intestinal activity in cats. *Epilepsia*, *22*, 309—314.

Danielsen, J., & Ellebjerg, J. (1966). Comparable EEG activation by megimide and metrazol. *Epilepsia*, *7*, 228—232.

Danielsson, I., Gasior, M., Stevenson, G. W., Folk, J. E., Rice, K. C., & Negus, S. S. (2006). Electroencephalographic and convulsant effects of the delta opioid agonist SNC80 in rhesus monkeys. *Pharmacology, Biochemistry and Behavior*, *85*, 428—434.

David, J., & Grewal, R. S. (1977). Induction of seizures with pentylenetetrazol in *Macaca mulatta*. Photic and pentylenetetrazol-induced seizure susceptibility in *Macaca mulatta*. *Journal of Medical Primatology*, *6*, 337—343.

Durmuller, N., Guillaume, P., Lacroix, P., Porsolt, R. D., & Moser, P. (2007). The use of the dog electroencephalogram (EEG) in safety pharmacology to evaluate proconvulsant risk. *Journal Pharmacological and Toxicological Methods*, *56*, 234—238.

Feige, B., Voderholzer, U., Riemann, D., Dittmann, R., Hohagen, F., & Berger, M. (2002). Fluoxetine and sleep EEG: Effects of a single dose, subchronic treatment, and discontinuation in healthy subjects. *Neuropsychopharmacology*, *26*, 246—258.

Gianotti, L. R., Kunig, G., Faber, P. L., Lehmann, D., Pascual-Marqui, R. D., Kochi, K., et al. (2008). Rivastigmine effects on EEG spectra and three-dimensional LORETA functional imaging in Alzheimer's disease. *Psychopharmacology (Berl)*, *198*, 323—332.

Please cite this article as: Authier, S., et al., Video-electroencephalography in conscious non human primate using radiotelemetry and computerized analysis: Refinement of..., *Journal of Pharmacological and Toxicological Methods* (2009), doi:10.1016/j.vascn.2008.12.003

Hughes, B. (2008). FDA drug approvals: A year of flux.*Nature Reviews Drug Discovery*, *7*, 107−109 2007.

Jurko, M. F., & Andy, O. J. (1967). Comparative EEG frequencies in rhesus, stumptail, and cynomolgus monkeys. *Electroencephalography Clinical Neurophysiology*, *23*, 270−272.

Lahunta, A., Glass, E. N., & Kent, M. (2006). Classifying involuntary muscle contractions. *Compendium Continuous Education Practicing Veterinarians*, *28*, 516−530.

Lallement, G., Clarencon, D., Masqueliez, C., Baubichon, D., Galonnier, M., Burckhart, M. F., et al. (1998). Nerve agent poisoning in primates: Antilethal, anti-epileptic and neuroprotective effects of GK-11. *Archives of Toxicology*, *72*, 84−92.

Mirski, M. A., Tsai, Y. C., Rossell, L. A., Thakor, N. V., & Sherman, D. L. (2003). Anterior thalamic mediation of experimental seizures: Selective EEG spectral coherence. *Epilepsia*, *44*, 355−365.

Morisson, F., Lavigne, G., Petit, D., Nielsen, T., Malo, J., & Montplaisir, J. (1998). Spectral analysis of wakefulness and REM sleep EEG in patients with sleep apnoea syndrome. *The European Respiratory Journal*, *11*, 1135−1140.

Oldham, H. G., Standring, P., Norman, S. J., Blake, T. J., Beattie, I., Cox, P. J., et al. (1990). Metabolism of temelastine (SK&F 93944) in hepatocytes from rat, dog, cynomolgus monkey and man. *Drug Metabolism and Disposition*, *18*, 146−152.

Poupard, L., Sartene, R., & Wallet, J. C. (2001). Scaling behavior in beta-wave amplitude modulation and its relationship to alertness. *Biological Cybernetics*, *85*, 19−26.

Pearce, P. C., Crofts, H. S., Muggleton, N. G., & Scott, E. A. (1998). Concurrent monitoring of EEG and performance in the common marmoset: A methodological approach. *Physiology & Behavior*, *15*, 591−599.

Rowan, J. A., & Tolunsky, E. (2003). *Primer of EEG.* Philadelphia, PA: Butterworth Heinemann.

Sharbrough, F., Chatrian, G. E., Lesser, R. P., Luders, H., Nuwer, M., & Picton, T. W. (1991). American Electroencephalographic Society guidelines for standard electrode position nomenclature. *Journal of Clinical Neurophysiology*, *8*, 200−202.

Thomsen, T., Kewitz, H., & Pleul, O. (1988). Estimation of cholinesterase activity (EC 3.1.1.7; 3.1.1.8) in undiluted plasma and erythrocytes as a tool for measuring *in vivo* effects of reversible inhibitors. *Journal of Clinical Chemistry and Clinical Biochemistry*, *26*, 469−475.

Tilz, C., Stefan, H., Hopfengaertner, R., Kerling, F., Genow, A., & Wang-Tilz, Y. (2006). Influence of levetiracetame on ictal and postictal EEG in patients with partial seizures. *European Journal of Neurology*, *13*, 1352−1358.

U.S. Food and Drug Administration. (2001). Guidance for industry: Safety pharmacology studies for human pharmaceuticals (S7A). http://www.fda.gov/cber/gdlns/ichs7a071201.htm

Wu, X., & Xiao, C. H. (1996). Quantitative pharmaco-EEG of carbamazepine in volunteers and epileptics. *Clinical Electroencephalography*, *27*, 40−45.

Please cite this article as: Authier, S., et al., Video-electroencephalography in conscious non human primate using radiotelemetry and computerized analysis: Refinement of..., *Journal of Pharmacological and Toxicological Methods* (2009), doi:10.1016/j.vascn.2008.12.003