

By Elena Moscardo and Chiara Rostello Measuring behavior & physiology

An integrated system for video and radio-telemetric EEG

The combined evaluation of physiology and behaviour allows for a complete and comprehensive preclinical assessment of central nervous system (CNS) functions. An integrated video-telemetric electroencephalography (Video-tEEG) system was developed to enable simultaneous assessment of animal behaviour by video tracking technologies, and electroencephalographic analysis by radio-telemetry in freely moving rats.

MATERIAL AND METHODS

Male Crl:CD (SD rats (Charles River, Italy) were surgically implanted with the three-channel radio-transmitter type TL10M3-F50-EEE (Transoma Medical, US) to obtain both cortical and hippocampal EEG waveforms and the electromyographic (EMG) signal.

Equipment set-up

The Video-tEEG system allows for simultaneous and continuous recording of videos and telemetric traces from up to 6 singly housed rats in the employed set-up (figure 1).

 Radio-telemetric system: 1 Dataquest ART Gold 4.1 (Data Sciences International); 6 receivers RPM-1; 2 synchronisers C12V; 2 Matrix (controlling 3 receivers each and 1 synchroniser C12V). Video system: 6 PhenoTyper cages (L45xW45 cm) with black/white camera top unit; 2 The Observer XT; 2 multi MPEG-4 Encoder software installed on The Observer PC (Noldus Information Technology); 2 Multiplexers Quad system (Panasonic, Model WJ-MS424), receiving images from 3 camera top unit each; 6 lateral black/white cameras (Ikegami, Model ICD-49E).

Data acquisition and analysis

Telemetric signals and videos from implanted animals were recorded during the post-surgery 15-day recovery period. Recordings of parameters were made continuously for 24 hours on Days 1, 6 and 15 after surgery. Video data were video-tracked using EthoVision XT 5.1 software (Noldus Information Technology). The distance moved, elongation, movement and velocity were evaluated. Durations of EEG stages (such as, Weak, Sleep and REM-sleep) were calculated applying the Rodent Sleep Scoring detector (Delta Band 0.5-3.5 Hz; Theta Band 3.5-7.5 Hz; EMG threshold 15-20 μ V) of Neuroscore v.2.0 (Data Science International). A visual review of the quality of telemetric traces was also conducted.

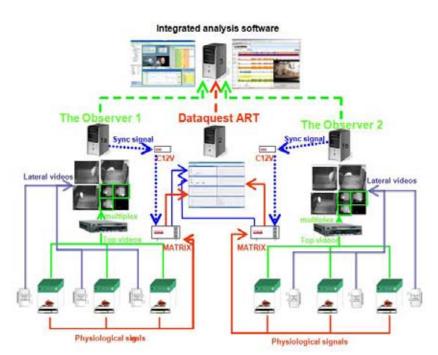


Figure 1. The complete Video-tEEG equipment set-up for the simultaneous and continuous recording of videos and telemetric traces from up to 6 singly housed rats.

RESULTS

No changes were noted in the distance of animal movement across days of the total postsurgery recovery period. Significant (p<0.01) increases of movement duration and velocity were noted comparing Day 15 with both Days 1 and 6. The analysis in time bins of 30 minutes of all the video-tracked parameters showed that the light-dark circadian cycle, with rats less active during the day and active during the night (dark phase from 6:00 pm to 6:00 am), was maintained starting from Day 1 after surgery (figure 2). The stretched position decreased from Day

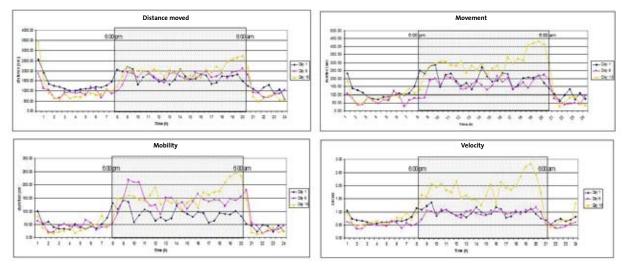


Figure 2. Mean Distance moved, Movement, Mobility and Velocity calculated each 30 minutes in the whole 24 hours period.

1, associated with a significant (p<0.01) increase of the contracted position observed starting from Day 6 (figure 3).

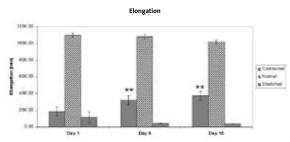


Figure 3. Times spent by the animal in different positions (contracted, normal and stretched) during the whole 24 hours period.** p<0.01 Day 1 vs Day 6 and Day 15 (t-Student for pair-wise comparisons).

The analysis of the EEG stages showed no significant changes among Days 1, 6 and 15. On Day 1 there were clear electrocardiographic (ECG) interferences on the EMG signal (Figure 4). These electrical influences were not present on Day 15 after surgery. The quality of the EEGs signals on Day 15 was satisfactory, both for the cortical EEG and hippocampal EEG, on which the prevalent Theta activity (3.5-7.5 Hz) was clearly identifiable (Figure 5).

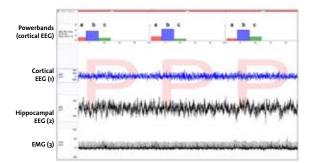


Figure 4. Day 1: example of telemetric traces during REM-Sleep (P) stages; EMG signal (3) with a clear ECG interference; cortical EEG (1), hippocampal EEG (2). Powerbands: a. Delta band 0.5-3.5 Hz; b. Theta band 3.5-7.5 Hz; c. Alpha band >7.5 Hz.

CONCLUSION

The surgical technique that was developed and applied to rats to record simultaneously cortical and hippocampal EEGs required 15 days for the complete recovery of animals, both in terms of health status and quality of telemetric signals. Telemetric waveforms and activities of animals collected with this integrated Video-tEEG system provide useful and powerful insights into the correlations between behavioural and electrical brain's activities. The system allows a reduction of the number of animals used, improving and refining the data quality by maximizing the amount of information gained from each experimental animal. This methodology provides researchers with a number of innovative applications, which can also be applied in routine research areas, such as in convulsion risk assessment, drug-induced sleepwakefulness disruption, environmental conditions and animal welfare evaluations.

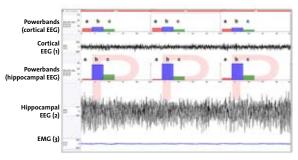


Figure 5. Day 15: example of telemetric traces during REM-Sleep (P) stages; EMG signal (3) without ECG interference; cortical EEG (1), hippocampal EEG (2). Powerbands: a. Delta band 0.5-3.5 Hz; b. Theta band 3.5-7.5 Hz; c. Alpha band >7.5 Hz.

CONTACT INFORMATION

Elena Moscardo - Elena.2.Moscoardo@gsk.com Safety Pharmacology, Safety Assessment Department, GlaxoSmithKline R&D Centre, Via A. Fleming 4, 37135 Verona, Italy