

Modulation of Sleep-Wake Cycles in Mice and Rats with Cannabinoids

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Introduction

The main psychoactive ingredient of marijuana Δ^9 tetrahydrocannabinol (Δ^9 THC) and endocannabinoids are involved in the modulation of the sleep-wake cycle. Systemic administration of the CB₁ antagonist/inverse agonist SR141716A dose-dependently induced an increase in wakefulness and a reduction in slow wave sleep (SWS) and rapid eye movement (REM) sleep [1]. In keeping with this observation, intracerebroventricular [2] or direct administration of anandamide into the peduncle pontine tegmental nucleus and medial preoptic area [3], two regions involved in the regulation of REM and non-rapid eye movement (NREM) sleep, increased NREM and REM sleep and reduced wakefulness. This effect was reversed with SR141716A [3, 4] indicating that the effects on sleep are mediated by activation of CB₁ receptors. However, the exact mechanisms by which cannabinoids modulate sleep-wake cycle are not fully understood.

Aim

The aim of the present study was to further our understanding of the endocannabinoid system in the modulation of sleep-wake cycle in freely moving rats and mice using a novel microchip-based data logging system.

Methods

C57BL6 mice (30-40g) (Harlan, UK) and Lister Hooded rats (250-300g) (Harlan, UK) were used in this study with all experiments performed under UK Home Office regulations and in accordance with the Federation of European Laboratory Animal Science Associations (FELASA) guidelines.

During surgery animals were anaesthetised with isoflurane and placed in a stereotaxic frame. Gold screw electrodes were positioned above the medial pre-frontal cortex and parietal cortex overlaying the left and right hippocampus. Further electrodes were located in the parietal and occipital areas as reference and ground. The pins were then assembled and secured to the skull surface using dental cement and tissue glue. Post-surgery, animals were allowed 7 days recovery prior to the start of recordings.

Mice received an acute intraperitoneal (ip.) injection of either the CB₁ agonist WIN55,212-2 (WIN-2), CB₁ antagonist AM251, the neutral CB₁ antagonist ABD459 or vehicle (Triethylene glycol and phosphate buffered saline). Whereas, rats were injected with either i) WIN-2; ii) AM251; iii) WIN-2 + AM251 or iiiii) vehicle. EEG recordings were performed immediately following drug administration with a recording duration of 6 hours. For recording purposes a wireless data Neurologger (New Behavior, Zurich, Switzerland) was used to monitor EEG activity. The Neurologger was attached to the head stage of the animal and allowed recording from up to 4 channels at a sampling rate of 200Hz (high pass filter: 0.1Hz, low pass filter:70Hz). A built-in accelerometer registered the animals movements throughout the experiment. Recordings were downloaded offline to a PC for analysis. Data were transformed using Matlab before being imported to SleepSign for the analysis of vigilant staging (NREM, REM, wake) and EEG spectral power [5].

Results

In mice, a significant reduction in REM sleep was evident following all cannabinoid treatments, combined with shorter REM episodes relative to vehicle controls. The cannabinoids however induced differential effects on wakefulness and NREM sleep; administration of WIN-2 resulted in a marked decrease in wakefulness while

AM251 induced an increase (see Figure 1). The CB₁ agonist also significantly increased NREM sleep whereas no change was observed with either of the antagonists.

In rats, WIN-2 also increased NREM sleep and the length of NREM episodes. Administration of AM251 failed to reverse the WIN-2 effect. However, AM251 did block the overall reduction in EEG spectral power observed during NREM and wakefulness after WIN-2 exposure. Treatment with AM251 alone had no effect on sleep stages but did increase the overall spectral power during NREM sleep.

Conclusions

All cannabinoids disrupted normal sleep patterns to some degree. The findings with WIN-2 and AM251 in this study corroborate previous studies reporting an increase in NREM and a reduction in wakefulness following exposure to anandamide or Δ⁹-THC [2,3,6] and increased wakefulness after treatment with SR141716A [1]. Overall the results are consistent with the endocannabinoid system being involved in modulating sleep and support the possibility of using cannabinoid agonists, neutral antagonists/inverse agonists for the treatment of sleep disorders.

References

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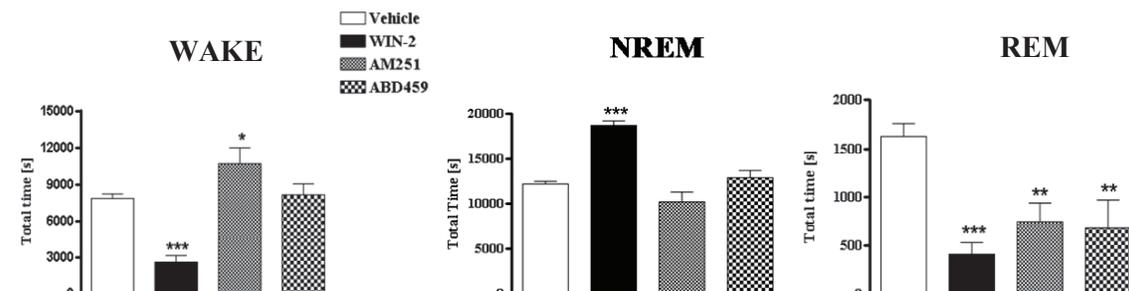


Figure 1. Effect of cannabinoids on time spent in WAKE, NREM and REM stages in mice. Mean + SEM. Asterisks denote reliable differences to Vehicle group (*= p<0.05; **= p<0.01; ***= p<0.001).