summarized as follows. Within the range of D, there is a wide 1:1 phase-locking region. In the outside of this region, the phase-locking regions with n:m appear via tangent bifurcations. The tangent bifurcation is caused by the map nearly tangential to the diagonal line (e.g. see the map for $D = 0.40$). Through this bifurcation, a fixed point with a long period abruptly appears. Interestingly, peculiar to the map of the two process model, the difference of the entrainment ratio $|n - m|$ changes through the tangent bifurcation. Unstable behavior is found in the region where an entrainment ratio changes.

DISCUSSION

If the two process model could be regarded as a combination of the different oscillator, the thresholds and the exponential switching processes, changing D would be equivalent to modifying interactions between them. Here, as a function of D, we have shown that the model has the different types of the mutual entrainment regions which are intervened by the tangent bifurcation. Such a dynamical system could provide the possible mechanisms underlying the diverse behavior of human circadian system. In addition to the internal desynchronizations investigated in a previous study,[3] the phase trapping observed on the way to the complete desynchronization[4] could be understood based on the bifurcation properties obtained here; that is, the circle map with a period n fixed point could exhibit the behavior that the sleep-onset phase comes back to its original position every n cycles where n corresponds to the period of the phase trapping. This is realized by an appropriate $D < 0.415$. Considering these results, our study uniquely provides the biological significance to the bifurcation properties of the two process model.

Bifurcations induced by other parameters such as amplitudes of the thresholds and periods of threshold processes remain unknown, which will be the subject of future study.

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Sleep–wake Regulation

Influence of cedar essence on spontaneous activity and sleep of rats and human daytime nap

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Abstract We investigated whether exposure to the odor of extracted cedar essence (CE) has (i) an influence on spontaneous activity and sleep–wake states of rats and (ii) a sleep-promoting effect on human daytime nap after taking an ordinary night's sleep. In rats exposed to CE, spontaneous activities and amount of wake were significantly decreased, while the amount of non-rapid eye movement (NREM) sleep was significantly increased. In human daytime nap, NREM sleep stage 2 latency was significantly shortened after exposure to CE.

Key words cedar essence, human daytime nap, odor, spontaneous activity and sleep of rats.

INTRODUCTION

It is commonly acknowledged that some odors extracted from plants have, among other things, sedative influences on mood, vigilance states and emotions. However, there are few studies that demonstrate their influences physiologically. The olfactory systems have anatomical connections with sites in the central nervous system, such as the limbic system, hypothalamus or mesencephalic reticular formation, which are well known to be involved in vigilance states or emotional states regulation.[1] Therefore, we investigated whether exposure to the odors of essence extracted from cedar (CE) has (i) an influence on

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spontaneous activity and sleep–wake states of rats and (ii) a sleep-promoting effect on human daytime nap after taking an ordinary night's sleep.

EXPERIMENT 1

We investigated spontaneous activities and sleep–wake states of rats with and without the exposure to the odor of CE.

Subjects and methods

Five male rats (3–5 months old), weighing 350–480 g were used for the spontaneous activity measurement, and five male rats (5–7 months old) were used for the sleep–wake measurement. The animals were housed under 12-h light and 12-h dark conditions (lights on 06:00 h), with *ad libitum* access to food and water. After 24-h habituation to the apparatus, locomotor activity, body movements without locomotion (such as grooming, shivering, postural displacement, etc. and rearing activity were automatically measured with an animal movement analyzing system (Scanet MV-10, MATYS, Toyama, Japan) over 2 days for the baseline activity and then 2 consecutive days for the activity under the exposure to the odor of CE in each animal. For polygraphic recordings, the rats were implanted with electrodes for electroencephalogram (EEG) and electromyogram (EMG) under pentobarbital anesthesia (50 mg/kg, i.p.). After recovery from surgery (for at least 2 weeks), the rats were habituated to the recording procedure for 24 h. Then, 2 day baseline sleep data were obtained, followed by 2 day sleep recordings under exposure to CE. For the present analyses, data from the second baseline and the second cedar day were used, taking account of some time to allow the atmosphere of the chamber to be saturated with CE odor. There was no significant difference between the first and second baseline data. Twenty-four hour EEG were scored visually for 15-s epoch as wake, non-rapid eye movement (NREM) sleep and rapid eye movement (REM) sleep according to standard criteria. The EEG were subjected to a 256-point fast Fourier transformation routine. Power spectra were calculated per sleep stage. Results were analyzed using the Wilcoxon signed-ranks test and the paired *t*-test for pairwise comparisons to baseline.

Results

Exposure to the odor of CE significantly decreased locomotor activities and body movements without the locomotion (Fig. 1) in the active (dark) phase of rats ($P < 0.05$), but no difference was seen in the rearing activities. Fig. 2 shows the average amount of time spent in wake, NREM and REM sleep over baseline 24 h and cedar 24 h. Amounts of wake significantly declined under the exposure to CE ($P < 0.05$). In contrast, exposure to CE significantly increased amount of time spent in NREM sleep ($P < 0.05$). The amount of REM sleep showed no difference. No significant difference was observed between delta power of NREM sleep in the baseline day and that in the cedar day.

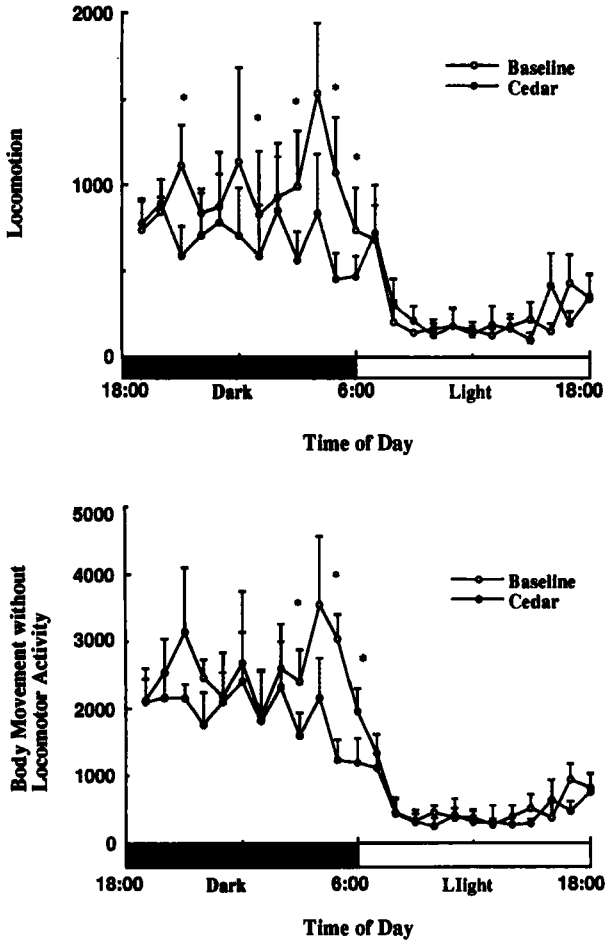

Figure 1. Effect of cedar (–●–) on total time spent in various stages of sleep over 24 h. Values are expressed as mean ± SEM. $^*P < 0.05$ (paired *t*-test). –○–, baseline.

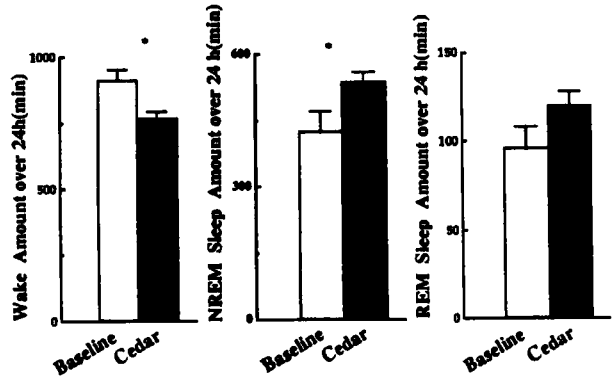

Figure 2. Twenty-four hour profiles of spontaneous activity on baseline (□) *vs* cedar (■) days. Values are expressed as mean ± SEM of arbitrary units provided by an animal movement analyzing system. $^*P < 0.05$ (Wilcoxon signed-ranks test).

Discussion

In the present study, the inhibitory effect of CE on spontaneous activities was probably not due to an increase of impassive states to a stimulus, such as deeply sleeping states, since there were no differences in the rearing activities that are

usually induced following the orientation to an unfamiliar stimulus in rodents. However, a sleep-facilitating effect was observed in the active (dark) phase of rats. In addition, delta power of NREM sleep, which is commonly believed to be an index of deep sleep, was not significantly different on the baseline and the cedar day. Taken together, it is suggested that a sleep-facilitating effect of CE in our results is exerted on the early phases of sleep. Our previous study has suggested that suppression of sympathetic activity may play an important role for the transition from waking to sleep.[2] Although we have no data with the effects of CE on sympathetic activity, Miyazaki *et al.* reported that blood pressure and coefficient of variation of R-R intervals decreased after inhalation of Taiwan Hinoki oil.[3] Therefore, it is plausible to assume that a sleep-facilitating effect of CE, in part, is mediated by sedation or suppression of sympathetic activity.

EXPERIMENT 2

We investigated whether odor exposure of CE has a sleep-facilitating effect on human daytime nap after taking an ordinary night's sleep, under the double-blind, crossover design between two groups with and without the odor.

Subjects and methods

Ten healthy university students, with no complaints of insomnia or olfactory disorders, were studied. The mean age of the subjects was 23.4 ± 1.3 years. Prior to the study, using sleep questionnaire we confirmed no difference in the sleep habit between the two groups. The polysomnograms (EEG, EMG, EOG, ECG and respiration) were monitored to score sleep stages by the manual of Rechtschaffen and Kales.[4] We measured sleep latency to NREM sleep stages 1 (S1) and 2 (S2) according to the guidelines[5] of the multiple sleep latency test (MSLT) twice a day on two consecutive days during the morning (09:30–11:30 h) and the afternoon (13:30–14:30 h). For CE odor exposure, we used a small electrically operated apparatus with a built-in plastic bottle for emitting the odor in the bedroom and placed it under the footboard of the bed. One hour before the experiment it was turned on with an electric switch and it continued to emit the odor throughout the experiment. In the control experiment without the odor, we put tap water, instead of CE oil, in the plastic bottle. Non-parametric Wilcoxon signed-ranks test was used to compare the sleep latency between the two groups.

Results

Table 1 shows the results of sleep latency with and without the CE odor exposure in the daytime nap study. For CE odor exposure, S1 latency shortened in 6/10 subjects (60%) in both morning and afternoon naps. This shortening of S1 latency was not statistically significant in comparison with the control. S2 latency shortened in 8/10 subjects (80%) in the morning nap, while it shortened in 5/10 (50%) in the afternoon nap. This shortening of S2 latency in the morning nap was significant ($P < 0.05$). No side effects were observed during or after CE odor exposure.

Table 1. *Sleep latency with and without cedar essence odor exposure in the daytime nap*

	S1 latency (min)		S2 latency (min)	
	Morning nap	Afternoon nap	Morning nap	Afternoon nap
With cedar essence (mean ± SD)	3.0 ± 2.9	3.1 ± 2.1	13.9 ± 4.8*	11.1 ± 5.1
Without cedar essence (mean ± SD; control)	4.9 ± 4.7	4.6 ± 5.8	16.4 ± 5.8	11.1 ± 5.2

*A significant difference ($P < 0.05$) between sleep latency with and without cedar essence.

DISCUSSION

Sleep latency is more prolonged in the daytime nap after taking an ordinary night's sleep than in the night's sleep, and especially more prolonged in the morning nap than in the afternoon nap. Therefore, in this study, significant shortening of S2 latency in the morning nap for CE odor exposure suggests that CE odor has a sleep-facilitating effect. This sleep facilitating mechanism is unknown, but it is possible that some factor of CE odor works in the process of sleep from arousal. Taken together with the results from experiment 1 in rats, there may be some components in CE odor which cause sedation and suppression of sympathetic activity and also which are important for the transition from waking to sleep.[2]

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