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Abstract

The present study was performed to investigate the effects of valerian extract on the sleep-wake cycle using sleep-disturbed model rats. A significant shortening in sleep latency was observed with valerian extract at doses of 1000 and 3000 mg/kg. On the other hand, valerian extract had no significant effects on total times of wakefulness, non-rapid eye movement (non-REM) sleep, or REM sleep, even at a dose of 3000 mg/kg. Valerian extract at doses of 1000 and 3000 mg/kg showed a significant increase in the delta activity during non-REM sleep. In conclusion, valerian extract may be useful as an herbal medicine having not only sleep-inducing effects but also sleep quality-enhancement effects.

KEYWORDS: delta activity, insomnia, sleep latency, sleep quality, sleep-disturbed model, valerian

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Original Article

Effects of Valerian Extract on the Sleep-wake Cycle in Sleep-disturbed Rats

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The present study was performed to investigate the effects of valerian extract on the sleep-wake cycle using sleep-disturbed model rats. A significant shortening in sleep latency was observed with valerian extract at doses of 1000 and 3000 mg/kg. On the other hand, valerian extract had no significant effects on total times of wakefulness, non-rapid eye movement (non-REM) sleep, or REM sleep, even at a dose of 3000 mg/kg. Valerian extract at doses of 1000 and 3000 mg/kg showed a significant increase in the delta activity during non-REM sleep. In conclusion, valerian extract may be useful as an herbal medicine having not only sleep-inducing effects but also sleep quality-enhancement effects.

Key words: delta activity, insomnia, sleep latency, sleep quality, sleep-disturbed model, valerian

Valerian, an herbal product consisting of the root of *Valeriana officinalis*, is used extensively as a traditional medicine for the treatment of insomnia and anxiety. For several decades, the use of valerian has decreased, and benzodiazepine sleep-inducers have taken the place of the herb [1]. However, it is well recognized that benzodiazepines have many unpleasant effects, such as low tolerance by some users, dependence, rebound insomnia, amnesia, and muscle relaxation [2, 3]. Therefore, particular attention should be given to the application of these hypnotics. In contrast, valerian showed no significant adverse effects, even at a dose approximately 20 times that of the recommended therapeutic dose [4].

There have been some reports on the hypnotic effects of valerian in humans [5–7]. In animal experiments,

Sakamoto *et al.* [8] reported that a 30% ethanol extract of valerian root significantly prolonged hexobarbital-induced sleep and decreased spontaneous ambulation and rearing with an open field test in mice. Leuschner *et al.* [9] also revealed that valerian root extract increased the thiopental sleeping-time in mice. However, there have been no reports that the effects of valerian on the sleep-wake cycle were studied in animals. In general, normal animals show a high-baseline sleep time during the day-time, so the hypnotic effect of valerian may be mild. Given these reports, it seems likely that nobody has been able to observe a significant effect of valerian on the sleep-wake cycle in animals.

In a previous study, we developed a sleep-disturbed model that was useful for evaluating hypnotic potencies by placing rats on a grid suspended over water [10, 11]. The hypnotic effects of drugs in rats placed on the grid suspended over water were more potent than that in rats placed on sawdust. In the present study, therefore, we studied the effect of valerian extract on the sleep-wake

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cycle using the sleep-disturbed model.

Materials and Methods

Animals. Wistar male rats weighing 230–310 g (Japan SLC, Shizuoka, Japan) were used. All animals were maintained in an air-conditioned room with controlled temperature ($24 \pm 2^\circ\text{C}$) and humidity ($55 \pm 15\%$). They were housed in aluminium cages with sawdust and kept under a light-dark cycle (lights on from 7:00 to 19:00). The animals were allowed free access to food and water except during the experiments. All procedures involving animals were conducted in accordance with the guidelines of the Animal Care and Use Committee, Faculty of Pharmaceutical Sciences, Okayama University.

Surgery. The animals were anesthetized with pentobarbital sodium (Nembutal®, 35 mg/kg, i.p., Abbott Laboratories, North Chicago, IL, USA) and then fixed in a stereotaxic apparatus (SR-5, Narishige, Tokyo, Japan). For electroencephalogram (EEG) recording, a stainless steel screw electrode was chronically implanted into the right frontal cortex (A: 0.5, L: 3.0) according to the atlas of Paxinos and Watson [12]. To record the electromyogram (EMG), we implanted stainless steel wire electrodes ($200\ \mu\text{m}$) into the dorsal neck muscle. A stainless steel screw fixed in the left frontal bone served as a reference electrode. The electrodes were connected to a miniature receptacle, and the whole assembly was fixed to the skull with dental cement. At least 7 days were allowed for recovery from the surgery.

EEG and EMG recordings. EEG and EMG were recorded with an electroencephalograph (Model EEG 5113, Nihon Kohden, Tokyo, Japan) from 9:00 to 15:00. The recording was carried out according to the method described previously [11, 13]. The signals were amplified and filtered (EEG, 0.5–30 Hz; EMG, 16–128 Hz), then digitized at a sampling rate of 128 Hz and recorded using the data acquisition program SleepSign ver.2.0 (Kissei Comtec, Nagano, Japan). The EEG and EMG were measured in a plastic cage ($30 \times 18 \times 24\ \text{cm}$) that had a grid floor ($29 \times 15 \times 7\ \text{cm}$). The cage was filled with water up to 1 cm below the grid surface. The stainless steel rods of the grid (3 mm wide) were set 2 cm apart from each other. The observation cage was placed in a sound-proof and electrically-shielded box ($70 \times 60 \times 60\ \text{cm}$).

Sleep-wake state analysis. The sleep-wake states were automatically classified by 10-sec epochs as wakefulness, non-rapid eye movement (non-REM), or REM sleep by SleepSign ver.2.0 according to the criteria previously described [10, 14]. As a final step, the defined sleep-wake stages were examined visually and corrected if necessary. Each state was characterized as follows: wakefulness, low-amplitude EEG and high-voltage EMG activities; non-REM sleep, high-amplitude slow or spindle EEG and low-EMG activities; REM sleep, low-voltage EEG and EMG activities.

Calculation for delta activity during non-REM sleep. The delta activity during non-REM sleep was determined using a program of SleepSign ver. 2.0. The power spectrum densities, integrated and averaged, could be divided into 4 frequency areas: delta wave (0.5–4 Hz), theta wave (4–8 Hz), alpha wave (8–13 Hz), and beta wave (13–30 Hz). The data of delta power in non-REM sleep were expressed as a percentage of the average delta activity in non-REM sleep during the entire recording period of each control group.

Drugs. The valerian (*Valeriana officinalis* L.) root extract was provided by Kobayashi Pharmaceutical Co., Ltd. (Osaka, Japan). The root of *Valeriana officinalis* L. was extracted with 70% ethanol at room temperature. The extract was concentrated and spray-dried with 8% maltodextrin and 1% silicon dioxide. The drug was dissolved in water and administered orally at 9:00, and EEG and EMG were measured for 6 h after drug administration. Eight rats were used in each group, and a counterbalanced design for drug dosage was used. Drugs were administered at intervals of 7 days when the same rats were used for repeated experiments. Each rat was subjected to experimentation 4 times.

Data analysis and statistics. Values shown are means \pm S.E.M. Statistical analyses were performed using the software Dr. SPSS II (Nankodo Co., Ltd., Tokyo, Japan). One-way analysis of variance (ANOVA) with Dunnett's test was used for estimating the drug effects. Sleep latency was defined as the time from drug administration up until the first 12 consecutive 10-s epochs of sleep.

Results

Effects on sleep parameters. Valerian extract caused a dose-dependent decrease in sleep latency. A significant shortening of sleep latency was observed at

doses of 1000 and 3000 mg/kg (Fig. 1). On the other hand, valerian extract had no significant effects on the total times of wakefulness, non-REM sleep, or REM sleep, even at a dose of 3000 mg/kg (Fig. 2).

Effects on delta activity during non-REM sleep. At 1–2 h after administration, rats given valerian extract at a dose of 1000 mg/kg showed a significant increase in delta activity during non-REM sleep. A significant increase of delta power was also observed at 1–2 and 2–3 h when a dose of 3000 mg/kg valerian extract

was given (Table 1).

Discussion

In the present study, we studied the effect of valerian extract on sleep using a sleep-disturbed model developed by placing rats on a grid suspended over water. As described in the previous section, valerian extract caused a significant shortening of sleep latency. Balderer and Borbély [5] have reported that valerian extract at doses of 450 and 900 mg reduced sleep latency and awake time after sleep onset in healthy subjects. Francis and Dempster [15] also reported that valerian treatment led to significant reductions in sleep latency or nocturnal time awake and an increase in total sleep time compared to a placebo in children. This is the first finding that valerian is an herbal product having hypnotic potency in rats with a sleep disturbance model.

To clarify whether valerian exerts an effect on sleep quality, we studied the effect of valerian on delta activities during non-REM, which are thought to reflect sleep quality. It is well known that delta power is an indicator of depth within non-REM sleep [16–18]. Valerian extract produced a significant increase of delta activities during non-REM sleep. Schulz *et al.* [6] found that valerian induced an increase of slow wave sleep (SWS) in subjects with low amounts of SWS. Herrera-Arellano *et al.* [19] demonstrated that valerian extract increased delta sleep in patients suffering from insomnia. On the other hand, Vonderheid-Guth *et al.* [20] reported that in healthy volunteers, a valerian and hops extract combination produced a clear power increase in delta frequency at 2 and 4 h after administration. These findings revealed

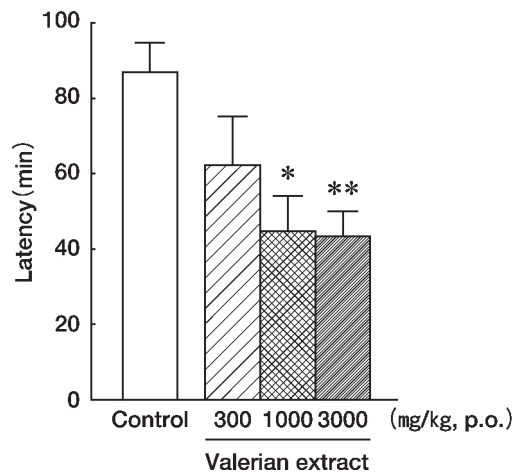


Fig. 1 Effects of valerian extract on sleep latency in sleep-disturbed rats. Columns and vertical bars represent means \pm S.E.M. ($n=8$). Drugs were administered orally. *, **, Significantly different from the control group at $P < 0.05$ and $P < 0.01$, respectively.

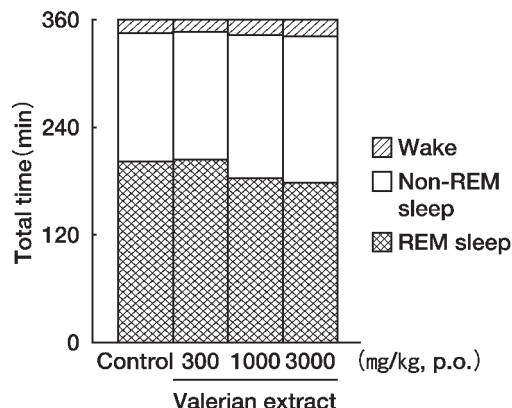


Fig. 2 Effects of valerian extract on total time of each sleep state in sleep-disturbed rats. Columns represent the means of each sleep state ($n=8$). Drugs were administered orally, and EEG and EMG were measured for 6 h.

Table 1 Effects of valerian extract on delta activity during non-REM sleep in sleep-disturbed rats

Hour	Control	Valerian extract (mg/kg, p.o.)		
		300	1000	3000
0–1 h	92.7 \pm 6.6	104.5 \pm 9.4	108.1 \pm 8.9	107.3 \pm 8.0
1–2 h	109.0 \pm 2.1	117.1 \pm 6.3	129.6 \pm 6.5*	131.5 \pm 3.5**
2–3 h	103.6 \pm 3.2	110.2 \pm 2.6	111.2 \pm 6.7	122.8 \pm 5.4*
3–4 h	98.8 \pm 6.8	102.1 \pm 5.3	107.5 \pm 5.1	109.9 \pm 9.9
4–5 h	96.4 \pm 3.9	96.0 \pm 5.3	101.0 \pm 4.6	102.6 \pm 4.0
5–6 h	85.1 \pm 5.2	84.1 \pm 5.9	89.0 \pm 5.5	91.4 \pm 3.6 (%)

Values represent means \pm S.E.M. ($n=8$). Drugs were administered orally. *, **, Significantly different from control group at $P < 0.05$ and $P < 0.01$, respectively.

that valerian extract enhances sleep quality.

The detailed mechanisms involved in the hypnotic effects and the increasing of delta power during non-REM sleep induced by valerian are not yet clear. Santos *et al.* [21, 22] reported that an aqueous extract of valerian inhibits (^3H) γ -aminobutyric acid (GABA) uptake and induces the Ca^{2+} -independent release of (^3H)-GABA. Ortiz *et al.* [23] also reported that *Valeriana officinalis* extract increased both K^+ - and veratridine-stimulated (^3H) GABA release and caused the inhibition of (^3H) GABA uptake. On the other hand, it is well known that GABA_A agonists, such as muscimol and 4, 5, 6, 7-tetrahydroisoxazolo-(5, 4-c) pyridin-3-ol enhanced delta activity during non-REM sleep [17, 24]. From these findings, it seems likely that the extracellular GABA level increased by valerian extract caused the hypnotic effects and the enhancement of delta activity during non-REM sleep through the GABA_A receptor.

In conclusion, valerian extract may be useful as an herbal medicine having not only mild hypnotic effects but also sleep quality-enhancement effects.

References

- Wagner J, Wagner ML and Hening WA: Beyond benzodiazepines: alternative pharmacologic agents for the treatment of insomnia. *Ann Pharmacother* (1998) 32: 680-691.
- Ashton H: Guidelines for the rational use of benzodiazepines; when and what to use. *Drugs* (1994) 48: 25-40.
- Freeman HL: Is there a need for a pure hypnotic? Approaches to the co-diagnosis of insomnia and anxiety. *J Drug Dev Clin Pract* (1996) 7: 289-302.
- Willey LB, Mady SP, Coughlin DJ and Wax PM: Valerian overdose: a case report. *Vet Hum Toxicol* (1995) 37: 364-365.
- Balderer G and Borbély AA: Effect of valerian on human sleep. *Psychopharmacology* (1985) 87: 406-409.
- Schulz H, Stolz C and Müller J: The effect of valerian extract on sleep polygraphy in poor sleepers: a pilot study. *Pharmacopsychiat* (1994) 27: 147-151.
- Stevenson C and Ernst E: Valerian for insomnia: a systematic review of randomized clinical trials. *Sleep Med* (2000) 1: 91-99.
- Sakamoto T, Mitani Y and Nakajima K: Psychotropic effects of Japanese valerian root extract. *Chem Pharm Bull* (1992) 40: 758-761.
- Leuschner J, Müller J and Rudmann M: Characterisation of the central nervous depressant activity of a commercially available valerian root extract. *Arzneimittelforschung* (1993) 43: 638-641.
- Shinomiya K, Shigemoto Y, Okuma C, Mio M and Kamei C: Effects of short-acting hypnotics on sleep latency in rats placed on grid suspended over water. *Eur J Pharmacol* (2003) 460: 139-144.
- Shinomiya K, Shigemoto Y, Omichi J, Utsu Y, Mio M and Kamei C: Effects of three hypnotics on the sleep-wakefulness cycle in sleep-disturbed rats. *Psychopharmacology* (2004) 173: 203-209.
- Paxinos G and Watson C: *The Rat Brain in Stereotaxic Coordinates*, 2nd Ed, Academic Press, San Diego (1986).
- Huang Z-L, Qu W-M, Li W-D, Mochizuki T, Eguchi N, Watanabe T, Urade Y and Hayaishi O: Arousal effect of orexin A depends on activation of the histaminergic system. *Proc Natl Acad Sci* (2001) 98: 9965-9970.
- Shigemoto Y, Shinomiya K, Mio M, Azuma N and Kamei C: Effects of second-generation histamine H₁ receptor antagonists on the sleep-wakefulness cycle in rats. *Eur J Pharmacol* (2004) 494: 161-165.
- Francis AJ and Dempster RJ: Effect of valerian, *Valeriana edulis*, on sleep difficulties in children with intellectual deficits: randomised trial. *Phytomedicine* (2002) 9: 273-279.
- Neckelmann D and Ursin R: Sleep stages and EEG power spectrum in relation to acoustical stimulus arousal threshold in the rat. *Sleep* (1993) 16: 467-477.
- Faulhaber J, Steiger A and Lancel M: The GABA_A agonist THIP produces slow wave sleep and reduces spindling activity in NREM sleep in humans. *Psychopharmacology* (1997) 130: 285-291.
- Lancel M: The GABA_A agonist THIP increases non-REM sleep and enhances non-REM sleep-specific delta activity in the rat during the dark period. *Sleep* (1997) 20: 1099-1104.
- Herrera-Arellano A, Luna-Villegas G, Cuevas-Urtegui Ma.L, Alvarez L, Vargas-Pineda G, Zamila-Alvarez A and Tortoriello J: Polysomnographic evaluation of the hypnotic effect of *valeriana edulis* standardized extract in patients suffering from insomnia. *Planta Med* (2001) 67: 695-699.
- Vonderheid-Guth B, Todorova A, Brattström A and Dimpfel W: Pharmacodynamic effects of valerian and hops extract combination (Ze 91019) on the quantitative-topographical EEG in healthy volunteers. *Eur J Med Res* (2000) 5: 139-144.
- Santos MS, Ferreira F, Cunha AP, Carvalho AP and Macedo T: An aqueous extract of valerian influences the transport of GABA in synaptosomes. *Planta Med* (1994) 60: 278-279.
- Santos MS, Ferreira F, Cunha AP, Carvalho AP, Ribeiro CF and Macedo T: Synaptosomal GABA release as influenced by valerian root extract-involvement of the GABA carrier. *Arch Int Pharmacodyn* (1994) 327: 220-231.
- Ortiz JG, Nieves-Natal J and Chavez P: Effects of *Valeriana Officinalis* extracts on (^3H) flunitrazepam binding, synaptosomal (^3H) GABA uptake, and hippocampal (^3H) GABA release. *Neurochem Res* (1999) 24: 1373-1378.
- Lancel M, Crönlein TAM and Faulhaber J: Role of GABA_A receptors in sleep regulation; differential effects of muscimol and midazolam on sleep in rats. *Neuropsychopharmacology* (1996) 15: 63-74.