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Sleep Improving Effects of a Single Dose Administration of a Valerian/Hops Fluid Extract

A Double Blind, Randomized, Placebo-controlled Sleep-EEG Study in a Parallel Design Using Electrohypnograms*

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Abstract
Repetitive administrations of valerian/hops combinations have been widely used for self-administered therapy of sleep disturbances. This investigation focuses on the question if a single administration can be an effective sleep aid. Two parallel groups of n = 20 (verum) and n = 22 (placebo) were tested. Each subject spent two consecutive nights in the lab (reference night and medication night).

Medication consisted in giving verum or placebo to poor sleepers identified by a validated sleep questionnaire (Schlaffragebogen SF-B). Two ml of the liquid extract or similar smelling placebo were diluted in 50 ml water (flavoured with honey) and administered 15 minutes before EEG recording during the medication night.

The data analysis is based on the electrohypnogram – a method derived from a validated computer assisted automatic analysis for depth of sleep. Differences between the reference nights and medication nights were evaluated and tested for significance. Time spent in sleep (values of the sleep frequency index "SFx" of the electrohypnogram of 74% or lower) was significantly higher for the verum group in comparison to the placebo group ($p<0.01$). The difference with respect to time spent in deeper sleep (i.e. 68% and lower or 62% and lower) between reference and medication night was also statistically significant at $p<0.01$. This parameter correlated with the difference in quality of sleep between the two consecutive nights as derived from the sleep inventory SF-A sub-score (subjects evaluation) with $r = 0.48$ at $p<0.0001$.

The EEG derived parameter "sleep quantity" as calculated from the electrohypnogram proved superiority of the valerian/hops combination over placebo. Thus, the present investigation has shown evidence that a valerian/hops fluid extract can be used successfully using a single administration.
Key words: Valerian, Hops, EEG, Sleep, Electrohypnogram

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Introduction

Insomnia affects approximately one-third of the adult population and contributes to increased rates of absenteeism, health care use, and social disability. Available evidence according to a recent review suggests that valerian might improve sleep quality without producing side effects (Bent et al. 2006). But double-blind, placebo controlled, randomized clinical studies are rare and partially contradictory (Stevinson and Ernst 2000). A recent search in literature did not provide evidence for valerian as a sleep aid (Taibi et al. 2007). There may be different reasons for this. Primarily, sleep disturbances are very heterogeneous. Therefore, the present study was conducted in subjects reporting on poor sleep but who were not suffering from any organic disease. This was assured by a neurological examination, blood analysis, recording of ECG and EEG during the screening procedure. We call them bad sleepers who can be regarded as a more homogeneous but important population.

Secondly, plant derived preparations contain quite a number of different active constituents possibly leading to opposite effects at higher dosages of the extract. Thus, dosage might be critical. Thirdly, there might be a big difference between fresh fluid extracts and dry extracts due to degradation or loss of active ingredients during the preparation of dry extracts. Last not least, different methods of sleep analysis might contribute to contradictory results. The great number of surrogate parameters derived from polysomnographic recordings shows a great deal of uncertainty with respect to their interpretation i.e. with respect to the restful value of sleep. The present study was conducted in order to prove the acute effectiveness of a fresh fluid extract of a mixture of two herbs traditionally known to have a positive influence on sleep by using a highly sensitive validated automatic spectral frequency based EEG analysis published earlier (Dimpfel et al. 1998) leading to what is now called an electrohypnogram.

Material and Methods

The herbal preparation (Dormeasan®, Bioforce AG, Roggwil, Switzerland) consisted of a combination of

460 mg of valeriana officinalis: radix rec. tinct. 1:10 plus 460 mg of Humulus lupulus: strobulus rec. tinct. 1:12 dissolved in 61% ethanol. Two ml were further dissolved in 50 ml of honey flavoured water and administered orally. The placebo preparation was developed specially for this study and was nearly undistinguishable from verum. Since the preparation was not available in Germany, no one could know which one of the strongly smelling preparations was verum.

Male and female otherwise healthy volunteers (30 to 70 years old, mean age of males 48.2 years, of females 50.2 years) reporting on having poor sleep were recruited by asking them to fill out a validated questionnaire on their sleep experience for the last two weeks (Sleep questionnaire SF-B according to Görtelmeyer, 1996). Inclusion criteria consisted in the existence of sleep disturbances without neurological complications. No alcohol or caffeine containing beverages were allowed on trial days and nights. Excluded were subjects with acute or chronic disease, sleep apnea (when detected during the reference night), sleep parasomnia, pathological EEG, pregnancy, allergic diseases, drug dependence, alcohol intake at experimental days or intake of medication within the last 5 days before administration of trial medication (physical exam and medical history during screening visit). Participation was limited to body weight between 50 and 115 kg. The study was performed in a double blind, randomized, placebo controlled manner with parallel groups. Subjects underwent a reference EEG recording starting with 10 min "eyes open" and 10 min "eyes closed" in order to have a reference recording before going to sleep. Recording was performed during two consecutive nights, the first one as reference and the second one under drug condition (either placebo or verum in parallel groups). On the evening of the second night patients got the trial medication (verum or placebo) 15 min before the lights were turned off at 22 h. Recording of EEG and polygraphic parameters was continued for the next 8 hours. With this design each individual was taken as its own control. The difference between these two consecutive nights was taken to compare the parallel groups (placebo or verum).

Depth of sleep during the night varies considerably. Cyclic phases of a duration of about 90 minutes show increases and decreases as already seen using the classical subjective evaluation according to historical criteria (Rechtschaffen and Kales 1968). After validation of the spectral sleep frequency index SFx against these criteria it became obvious that these phases show continuous transitions from waking to deep sleep and back to light sleep within a period of about 90 minutes. The resulting time curve is called an electrohypnogram.

In order to obtain electrohypnograms the EEG was recorded bipolarly from 17 surface electrodes according to the international 10/20 system with Cz as a physical reference electrode (**C**omputer **a**ided **t**opographical **e**lectro-**e**ncephalo-**m**etry: CATEEM® from MediSyst GmbH, 35440 Linden, Germany), using an electrocap. The raw signals were amplified, digitized (2048 Hz/12 bit) and transmitted via fiber optical devices to the computer. The automatic artefact rejection of the CATEEM®-System, which eradicates EEG-alterations caused by eyeblinks, swallows, respiration etc. during the recording was automatically controlled and individually adjusted by the investigator. ECG and EOG were recorded in one channel each in order to facilitate detection of those signals superposing the EEG. The artefact rejection set-up was observed for about 3 minutes prior to the start of the recording to ensure, that all artefacts are correctly eliminated from further evaluation. For safety purposes the original raw data was saved on optical disk in order to allow re-evaluation of the artefact rejection mode if necessary. In these cases the experimental session was re-examined offline in total with a new adapted rejection mode. The amount of rejected data is determined automatically and given in percent of total recording time. Nevertheless the entire recording and the computer-based automatic artefact rejection were continuously supervised and adjusted by a trained technician like reported earlier for day time studies (Dimpfel et al. 1992).

The signals of all 17 electrode positions undergo Fast Fourier Transformation (FFT) based on 4-second sweeps of data epochs (Hanning window). Data are analysed from 0.86 to 35 Hz using the CATEEM® software. The resulting frequency spectra are divided into six frequency bands: delta (1.25 - 4.50 Hz), theta (4.75 - 6.75 Hz), alpha1 (7.00 - 9.50 Hz), alpha2 (9.75 - 12.50 Hz), beta1 (12.75 - 18.50 Hz) and beta2 (18.75 - 35.00 Hz). This frequency analysis is based on absolute spectral power values.

The electrohypnogram was calculated offline using a patented algorithm involving theta and beta power at central electrodes. The formula basically involved the addition of spectral power of theta + beta1 range divided by the power in the beta2 frequency range. For definition of frequency ranges see above. The result of this algorithm has been termed spectral or **S**leep **F**requency inde**x** (SFx) and has been validated against the criteria of Rechtschaffen and Kales (Dimpfel et al., 1998).

The parameter "sleep quantity" has been taken as confirmatory parameter in advance for statistical analysis after calculation of a bootstrap for estimation of number of needed subjects based on an earlier study comparing healthy subjects with patients suffering from insomnia, snoring or obstructive sleep apnea (Hammer et al. 2001). Sleep was evaluated also by a sleep questionnaire (Sleep questionnaire SF-A, CIPS, Görtelmeyer 1996) after the reference and the medication night, the possibility of a correlation analysis was used to relate this surrogate parameter from the EEG to the subcore "sleep quality" of the questionaire.

According to the parallel design of the study Wilcoxon, Mann & Whitney U-test was used for all comparisons. This nonparametric test was chosen due to the fact that normal distribution of EEG data can not be assumed despite the fact that the data were log-transformed. As parameter for the analysis, time spent beyond a particular depth of sleep as determined using the spectral frequency index SFx was taken. The evaluation was based on the comparison of the differences between the first reference night and the consecutive medication night with respect to the parallel placebo and verum groups. Thus, each subject served as its own control. The study protocol was approved by an ethic vote of the "Landesärztekammer Hessen", Germany.

RESULTS

Fig. 1 and Fig. 2 document a representative example of electrohypnograms from a single subject having problems to get to sleep with respect to the first (reference) and second (medication) night. A more or less regular pattern of periodic changes is visible. For quantitative evaluation of sleep, the amount of time was determined, which was spent beyond a particular depth of sleep, i.e. beyond depth of 74% according to the SFx. A transition from wakefulness to sleep has been observed with this method starting at about 80%. Thus, sleep depth of 74% signalizes definite sleep. This parameter is used for evaluation of drug induced changes in comparison to the reference night.

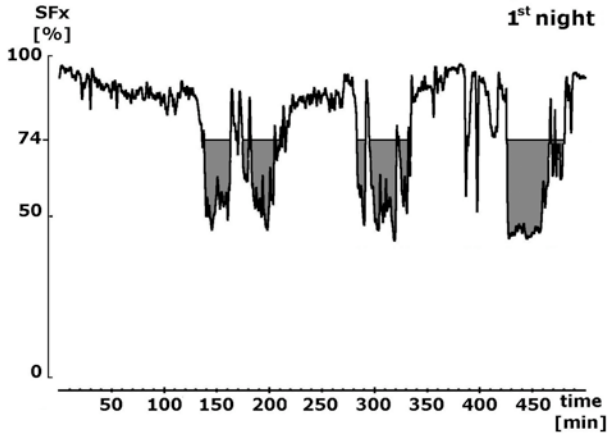

Fig. 1. Schematic view of the electrohypnogram as derived from SFx – values (**S**pectral **F**requency inde**x**) representing the depth of sleep in % of maximal wakefulness (ordinate). The time is given on the abscissa. **Sleep quantity** is defined as the time spent beyond a particular value of the SFx like 74%. A representative example of a **baseline** night is shown.

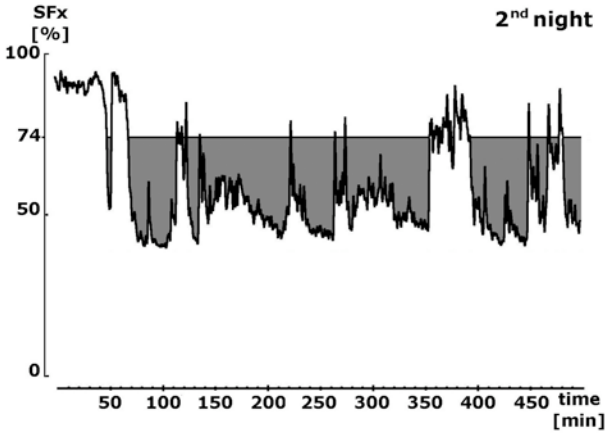

Fig. 2. Schematic view of the electrohypnogram as derived from SFx – values (**S**pectral **F**requency inde**x**) representing the depth of sleep in % of maximal wakefulness (ordinate). The time is given on the abscissa. **Sleep quantity** is defined as the time spent beyond a particular value of the SFx like 74%. A representative example of a **medication** night is shown (same subject as in Fig. 1).

These single sleep periods of about 90 minutes follow such a rigid pattern that it is possible to average the SFx values (representing depth of sleep) of a whole night for a larger group of volunteers. The periodic increases and decreases of depth of sleep are visible despite the average process. The data of the poorly sleeping volunteers of the present study - verum (n = 20) or placebo (n = 22) – were also averaged to give their median of one minute values (green and red values for verum and placebo, respectively as depicted in Fig. 3). It is quite obvious that the periodic changes are still visible also for the placebo sleepers but their sleep does no reach maximal depth. Subjects treated with the valerian/hops combination generally reach deeper sleep

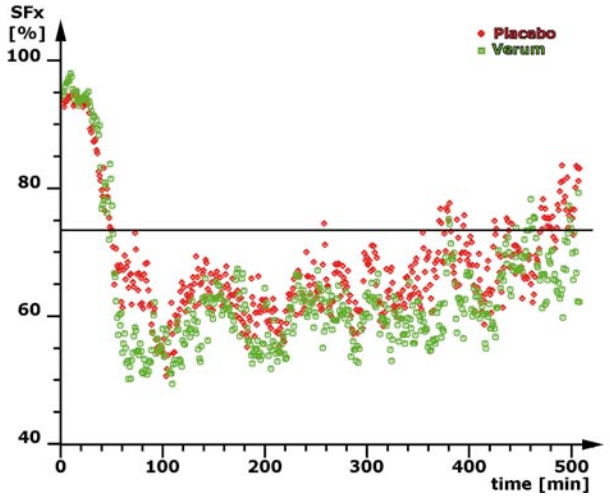

Fig. 3. Median values of depth of sleep as depicted for SFx values recorded every minute throughout the night. Values are depicted for verum and placebo.

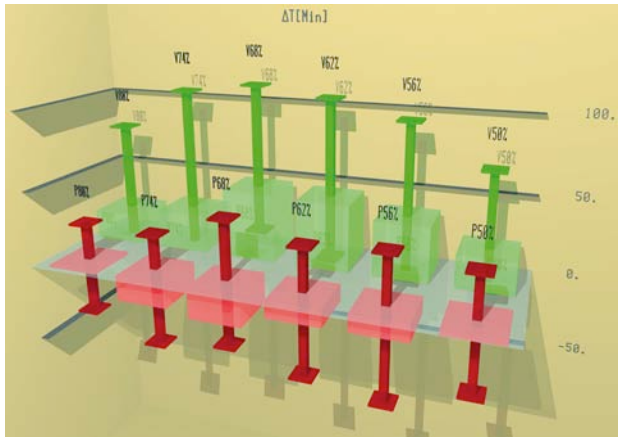

Fig. 4. Bar graph showing the difference between the first and second night with respect to **time** spent beyond a SFx – value (depth of sleep) from <74% (drowsiness – first sleep) down to <50% (deep sleep) throughout the night in jumps of 6%. Median values are given with quantiles. Green columns refer to verum, red columns to placebo. All verum values < 74% were statistically significant different from placebo values with an error probability of $p<0.01$, tested two-sided by use of Wilcoxon-Mann-Whitney statistics.

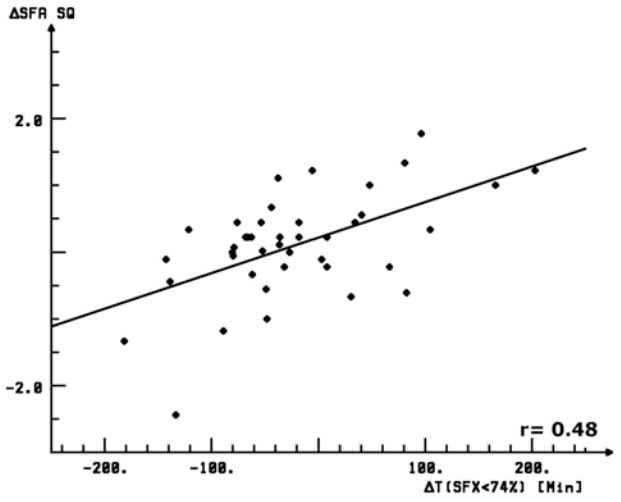

Fig. 5. Correlation analysis on the base of sleep quantity as defined from the SFx- value of 74% and "sleep quality" as defined as sub-score within the sleep questionnaire SF-A from Collegium Internationale Psychiatricum (CIPS). Difference in **Sleep quantity** (ΔT) between the two consecutive nights is given on the abscissa, difference in sub-score "sleep quality" (ΔSFA) on the ordinate.

during these periods than the participants of the placebo group (Fig. 3).

Regarding now the time of sleep spent beyond a given SFx value (termed "sleep quantity" for a particular depth of sleep and lower) it turns out that values for the active treatment group are higher in comparison to the values in the placebo group becoming significant for values of SFx = 74% and lower at the $p<0.01$ level of error probability. Fig. 4 documents these values in steps of 6 %. The valerian/hops combination shifts the time spent in sleep significantly to deeper sleep (depth beyond 74%). This effect is most pronounced at deeper levels of sleep (<68% or <62%. Thus valerian/hops significantly led to deeper sleep.

Results from the questionnaire were related to "sleep quantity". As can be seen in Fig. 5 the sub-score "quality of sleep" correlated significantly with the parameter "sleep quantity" at a particular depth of sleep of 74%. For "quality of sleep" a Pearson correlation coefficient of 0.48 was reached at $p<0.0001$. Thus, a positive conclusion can be drawn from the quantitative EEG data with respect to improvement of sleep also with respect to restfulness by means of a single administration of the fluid valerian/hops combination.

Discussion

Automatic quantitative analysis of the spectral frequency content of the EEG during the night has resulted in the development and validation of a new index of depth of sleep called the sleep spectral frequency index (Dimpfel et al. 1998). This led to the quantitative description of the periodic changes of depth of sleep by means of the electrohypnogram, which depicts these periodic changes in a continuous manner. This kind of analysis has proven to be able to discriminate between healthy subjects, patients suffering from insomnia, snoring or apnea (Hammer et al. 2001). Whereas the partition into 4 artificially defined sleep stages according to Rechtschaffen and Kales (1968) did not succeed in objective description of the quality of sleep, this automatic computer assisted methodology results in a more reliable surrogate parameter for the evaluation of sleep. Furthermore, this analysis based on spectral frequency content of the EEG does not rely on delta frequencies which do not increase necessarily for example during sleep in the presence of anaesthetic drugs. Instead of this, power in theta and beta frequencies are used to define depth of sleep, whereby power in beta2 frequencies decrease during healthy sleep. Prominent increases of spectral theta power have been observed in the rat following strongly sedative and sleep inducing drugs like medetomidine (Dimpfel and Schober 2001). Theta activity in the waking EEG has been shown to be a marker of sleep propensity in the rat (Vyazovskiy and Tobler 2005) and humans (Finelli et al. 2000). Spectral beta1 power increases in the presence of benzodiazepines like diazepam or midazolam prescribed for induction of sleep worldwide. Thus, this algorithm basically defined as theta plus beta1 power divided by beta2 power (SFx) seems to be sufficient to describe depth of sleep in form of the electrohypnogram replacing the hypnogram of Rechtschaffen and Kales (1968). The parameter "sleep quantity" related to a particular depth of sleep derived from the SFx obviously is more suited to describe differences between normal healthy sleep and non restful sleep in patients (Hammer et al. 2001). In addition, using the same parameter SFx as measurement of depth of sleep, vegetatively undisturbed anaesthesia (no cardiovascular reaction of the patient during particular provocations) could be shown to take place beyond a value of 68% (Renz et al. 1999).

Since interpretation of frequency changes of the EEG might be difficult without a golden standard one has to rely on correlations. In the present investigation such a correlation was found with respect to the subscore "sleep quality" of the sleep questionnaire SF-A which was taken in the morning after the reference and medication night as a subjective assessment. Even if the correlation with $r = 0.48$ is not very strong, the statistical error probability of $p<0.0001$ allows to interpret the amount of sleep depth of 74% SFx and lower designed as "sleep quantity" as an indicator of quality of sleep.

First scientific indication of a positive influence on sleep disturbed by noise using a valerian/hops combination was reported already during the seventies of the last century (Müller-Limmroth and Ehrenstein 1977). Successful repetitive administration of a valerian/hops combination as measured by several questionnaires (among them SF-A) was reported (Schmitz and Jackel 1998). These authors found the effect equal to the administration of a benzodiazepine. Evidence for the efficacy of this herbal combination using repetitive administrations comes from studies using polysomnographic standard examinations (Füssel et al. 2000). First evidence for an effect of this combination at day time on the frequency content of the EEG was published in the same year (Vonderheid-Guth et al.

2000). These results on day time spectral frequencies of the EEG were also reproduced meanwhile in rats (Dimpfel et al. 2006). A large multi-centre drug trial on 184 adults (randomized, placebo-controlled, parallel-group study) with mild insomnia revealed modest hypnotic effects of valerian-hops combination and diphenhydramine as examined by questionnaires (Morin et al. 2005). Even though the amount of drug in 2 ml Dormeasan® is below the daily recommendations of the actual European Scientific Cooperative (ESCOP) monographs on valeriana and hops and is also significantly lower than for example in the formulation used in the study of Morin, a statistically significant acute effect for Dormeasan® in comparison to placebo could be found now using a special parallel design, where each subject served as its own control. Reason for the good efficacy could be the amount of essential oils which is much higher in a valerian/hops tincture like Dormeasan® made from fresh plants in comparison to usual preparations made from dried plants.

In order to replace widely abused synthetic sleep aids with their known side effects, acute efficacy of herbal medicines must be proven by appropriate clinical studies based on good clinical practice (GCP). The present study followed these guidelines and provided the first evidence for an acute sleep improving effect of a fluid valerian/hops combination. Therefore this investigation can help doctors and patients in their decision of making a first choice for sleep aids.

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