ORIGINAL RESEARCH PAPER

Clinical Research

KEY WORDS: Sleep Disorder, Natural Sleep Supplement, Sleep Quality, Insomnia, Sleep Latency, Functional Food.

A CLINICAL INVESTIGATION ON THE SAFETY AND EFFECTIVENESS OF A NATURAL SLEEP SUPPLEMENT IN TREATING SLEEP DEPRIVATION IN HEALTHY ADULT HUMAN SUBJECTS: A STUDY ON SLEEP DISORDER MANAGEMENT

Dr Manisha Mishra — Medical Advisor, Vedistry Private Limited. 21, Evergreen Industrial estate, Shakti mills lane, Off Dr E Moses Road, Mahalaxmi, Mumbai- 400011, India

Dr Nayan Patel — Medical Director, NovoBliss Research Private Limited, Office# A - 206, 2nd Floor, Shaligram Lakeview, Nr. Vaishnodevi Circle, Khoraj, Gandhinagar – 382421, India

Maheshvari Patel* — Director Operations, NovoBliss Research Private Limited, Office# A - 206, 2nd Floor, Shaligram Lakeview, Nr. Vaishnodevi Circle, Khoraj, Gandhinagar – 382421, India *Corresponding Author

ABSTRACT

Background: Sleep disorders afflict millions globally, affecting overall health. Insomnia, characterized by difficulty falling or staying asleep, is particularly prevalent. Conventional treatments often pose side effects, prompting exploration of alternative approaches. This study investigates the safety and effectiveness of a natural sleep supplement containing Tagar (Valeriana officinalis), a key Ayurvedic herb, in addressing sleep disorders. **Methods:** During a 30-day study period, a cohort of 32 subjects presenting complaints regarding sleep, with a mean age of 39.63±8.37years, were treated and evaluated. The improvement in sleep quality was evaluated using Leeds Sleep Evaluation Questionnaires (LSEQ) and 'sleep diaries' that were provided to the subjects to record their sleeping patterns. Additional relevant endpoints were determined through clinical examinations, blood reports, and hedonic questionnaires for consumer feedback perception. Ethical approval was obtained prior to the commencement of the study, ensuring compliance with established research ethics. The statistical analysis of the acquired data was performed using R software (Version: 4.2.2). **Results:** A significant improvement in the sleep quality of subjects was observed, wherein 100% of the subjects reported facilitation in the onset of sleep, along with fewer nocturnal wakeful periods by Day 15. The ease to fall asleep exhibited a substantial increase of 258.8±76.67% (p-value<0.001), with time period to fall asleep reducing by 237.50±71.84% (p-value<0.001) after 15 days of treatment. Similarly, statistically highly significant improvements with p-value<0.001, were observed in parameters such as the time taken to awaken, the ease of waking up, a heightened sense of freshness and alertness, and diminished disruptions in balance and coordination during the morning hours. The study also identified a sixfold reduction in sleep onset latency, and a 1.3-fold increase in the overall duration of sleep. Importantly, over the course of the 30-day study period, no adverse effects or instances of treatment habituation were observed among the study subjects. **Conclusions:** The natural sleep supplement demonstrated efficacy in enhancing sleep quality, duration, sleep onset latency, and waking-up, while mitigating symptoms of morning fatigue. Notably, no habituation of the therapy or adverse events were observed after 30 days of regular administration. The tested treatment, 'Zzowin' tablet can be considered well-tolerated and efficacious in treating people suffering from sleep deprivation.

INTRODUCTION

Insomnia is the perception or complaint of inadequate or poor quality of sleep because of difficulty in falling asleep, difficulty in maintaining sleep or waking too early in the morning. Poor sleep is a common problem that affects many people worldwide. Chronic Insomnia has been proven to cause severe fatigue, anxiety, depression and lack of concentration. Poor sleep or sleep deprivation have been associated with poor concentration, poor judgment, depression, and even poor decision-making skill.

The test treatment is a tablet unit oral dosage form used to help people sleep faster, allowing a deeper and calm sleep, so they wake up fresh and energised, with a restful, quality sleep. The rationale of the study was to demonstrate the safety and effectiveness of a natural sleep supplement in healthy human adults suffering from sleep deprivation. While conventional treatments are available, they often come with side effects that can be unpleasant. For example, medications such as benzodiazepines and non-benzodiazepine hypnotics, have been shown to have side effects such as dizziness, headaches, and nausea. (Mayo Clinic, 2016)

Herbal medicine offers a natural alternative that can help improve sleep quality without the side effects of conventional treatments. (RootBabes, 2022) Supplements such as Indian valerian have been traditionally used to decrease the time it takes to fall asleep, while improving deep and restful sleep. (Shinjyo, Waddell, & Green, 2020) (Guadagna, Barattini, Rosu, & Ferini-Strambi, 2020)

The test treatment in this study contains Indian Valerian Root (*Tagar*), along with Melatonin, L-tryptophan and Manganese as key ingredients. This formulation consists of a unique blend of sleep-disorder treatments, including natural herbal alternative. The aim of this paper is to provide a better understanding of the benefits of an alternative natural sleep supplement i.e. 'Zzowin' tablets in treating sleep disorders, and to encourage further research in this area.

METHODS

Ethical conduct of the study

The research adhered to all applicable federal government codes, acts and regulations, GCP requirements, and ICH guidance E6 (R2) on 'Good Clinical Practice', ICMR Guidelines, Food Safety and Standards Authority of India (FSSAI) guidelines. Before initiating the study procedures, an independent ethics committee granted approval for the study protocol [version#01 (Final)], informed consent form [version#01 (Final)], case report form [version#01 (Final)], and other essential documents. This clinical study was registered with ClinicalTrials.gov with ID NCT05853757, and with the Clinical Trial Registry of India (CTRI) with CTRI number CTRI/2023/04/051587.

Study design

This was a single-arm, single-blind, single-centre study designed to assess the safety, efficacy, and in-use tolerability of the test herbal supplement in healthy adult human subjects experiencing difficulty falling asleep. The blinding was maintained for subjects, and they remained unaware of the test treatment.

A total of 35 adult human subjects within the age range of 18–65 years (mean age=39.63 years) experiencing difficulty falling asleep were enrolled, with 32 completing the study. Potential participants were screened according to the inclusion and exclusion criteria only after obtaining written informed consent. The study spanned 30 days and comprised four visits: a screening and baseline evaluations visit, followed by three additional visits on Day 01, Day 15, and Day 30 from the initiation of the test treatment. Subjects were advised during screening (prior to enrolment) to abstain from consuming any weight-loss medications or supplementation treatments throughout the study. A subjective assessment, conducted by a general medical practicing physician, was carried out during a 7-day observational period before the initiation of the test treatment to measure the routine sleep patterns of the subjects. Efficacy parameters were assessed before the initiation of the test treatment on Day 01 and compared with assessments after the use of the test treatment on Day 15 (+2 days) and Day 30 (+2 days).

The evaluation of the test treatment included the assessment of changes in sleep quality and duration on Day 01 and Day 07, using the Leeds Sleep Evaluation Questionnaires (LSEQ). The improvement in overall health and well-being was measured from baseline (Day 01) to Day 15 (+2 days), while dependence and withdrawal symptoms caused, if any (such as nausea, vomiting, dizziness, and headache) were examined through a subject treatment perception questionnaire via telephonic follow-up at Day 30 (+2 days). Additionally, the impact of the test treatment on blood biochemical parameters (Complete Blood Count (CBC), Erythrocyte Sedimentation Rate (ESR), Random Blood Sugar (RBS), SGPT, SGOT, Serum Creatinine) was assessed, comparing values from baseline (Day 01) to Day 15 (+2 days).

The study employed comprehensive consumer feedback assessment, utilizing structured questionnaires as a means of capturing the subjects' perceptions. The study staff quantified the effectiveness, likeliness, and overall satisfaction of the participants by employing a 9-point hedonic scale. This systematic approach allowed for a thorough exploration of participants' subjective experiences. Additionally, participants were equipped with 'Sleep Diaries' to monitor and document their adherence to the prescribed test treatment regimen. The person filling out the diary was required to be the same person who consumed the test treatment.

Test Treatment

The test treatment, developed by Charak Pharma, and marketed by Vedistry Private Limited (India), is a natural sleep supplement comprising 5 mg of Melatonin in a unit dosage, along with herbs such as Tagar and other essential nutrients. The product aims to enhance the quality of sleep, promote restfulness, and balance the sleep-awake cycle, all without inducing habit formation. (Table 01)

Table 01: Details of the test treatment.

Details	Test Treatment
Name	Zzowin
Ingredients	Melatonin, Tagar, L-tryptophan, L-theanine, Vitamin B6, Iron, Zinc, Magnesium
Dosage form	Tablet (solid unit dosage form)
Dosage	1 tablet daily half an hour before going to bed for consecutive 30 days
Route of administration	Oral Administration with a glassful (approx. 250mL) of water.
Manufactured by	Charak Pharma Private Limited, India
Marketed by	Vedistry Private Limited, India

Study participants

A total of 35 subjects were initially enrolled, resulting in 32 completed subjects who experienced difficulty falling asleep. During the screening phase, subjects were given a 'Sleep Diary' and were asked to fill out the diary card on a daily basis. After the completion of the 7-day screening period, the subjects were enrolled based on the evaluation of sleep patterns as per the filled daily diary.

Inclusion criteria

The study enrolled healthy, nonpregnant, and nonlactating female and male subjects aged 18 to 65 years who reported difficulties falling asleep due to shift work disturbance, jet lag, or mental stress. Inclusion criteria comprised subjects taking more than 30 minutes to fall asleep, having a subjective total sleep time of 6.5 hours per night for at least three nights per week, experiencing daytime complaints associated with disturbed sleep, and maintaining a habitual night bedtime between 20:30 hours and midnight. For female subjects of childbearing potential, inclusion necessitated a negative pregnancy test report and a commitment to follow an accepted method of birth control throughout the study. Additionally, subjects were required to provide written informed consent and agree to comply with all study procedures.

Exclusion criteria

Subjects with a history of seizures, allergies/sensitivities to the test treatment ingredients, or significant head trauma were excluded from the study. Additionally, individuals with sleep disorders other than primary insomnia (such as restless leg syndrome, sleep apnoea, etc.), those who were substance-dependent or had a history of alcohol abuse in the past year, and subjects using tobacco products during the night were not included. Exclusions extended to subjects who had worked night/rotating shifts in the past 7 days or planned to do so during the study, individuals on any regular medications (except for antihypertensives, antidiabetics, lipid-lowering agents, and drugs for primary cardiovascular prophylaxis), and those who had taken anti-inflammatory drugs or any other drug leading to weight gain (such as corticosteroids) within 5 days or any antihistamines/immunosuppressive agents within 7 days of the first dosing of the test treatment.

Furthermore, subjects who had used any other investigational drug within the past three months or had participated in similar nutraceuticals, food, supplemental, or therapeutic trials within the past four weeks were excluded. Individuals deemed unsuitable for enrolment by the investigator or expert physician were also excluded from the study.

Leeds Sleep Evaluation Questionnaire (LSEQ)

Subjects were asked the Leeds Sleep Evaluation Questionnaire on both Day 01 and Day 15. This questionnaire comprised ten questions pertaining to four consecutive aspects of sleep: getting to sleep (GTS), quality of sleep (QOS), awakening from sleep (AFS), and behaviour following wakefulness (BFW).

Subjects' Sleep Diary

There was 7-day observational period to observe the sleep pattern of the subject. Enrolment occurred for those subjects who completed the sleep diary, and the data collected included information on (i) the time interval to fall asleep, (ii) sleep interruptions, (iii) wakeup time, (iv) naps during wakeup time, (v) feelings, (vi) irritability, (vii) total sleep duration, and (viii) sleep quality. Change in sleep quality was self-evaluated by subjects using the sleep diary on Day 01 compared to Day -07 before test treatment usage, and again on Day 15 (+2 Days) after test treatment usage.

Clinical examination

A clinical examination was conducted before dosing on Day 01 for the baseline, and then on Day 15 (+2 days). This examination included the observation of common symptoms

such as not feeling refreshed, inability to sleep despite being tired, daytime drowsiness, irritability, difficulty in concentrating, impaired ability to perform normal activities, body ache, and heaviness of the body. Subjective parameters such as lack of concentration, heaviness in the head, headache, yawning, mental fatigue, drowsiness, giddiness, and exhaustion were also assessed.

Blood Parameters

During the study, various blood parameters, namely Complete Blood Count (CBC), Erythrocyte Sedimentation Rate (ESR), Random Blood Sugar (RBS), Serum Glutamic Pyruvic Transaminase (SGPT), and Serum Glutamic Oxaloacetic Transaminase (SGOT), were assessed on both Day 01 and Day 15 (+2 days). The investigation aimed to examine potential correlations between sleep patterns and blood glucose levels, given that sleep disturbances can influence the regulation of glucose in the body. The Random Blood Sugar test was utilized to determine the amount of sugar present in the blood at any given point in time, as both excessively low and high blood sugar levels have been noted to impact sleep patterns.

Subjective evaluation

Subjective perceptions regarding the test treatment, encompassing overall health, well-being, and improvements in sleep, were documented by posing questions to the subjects both before treatment at baseline on Day 01 and after treatment on Day 15 (+2 days). Consumer feedback on the treatment's effectiveness, likeliness, and overall satisfaction was collected through hedonic questionnaires. Additionally, the assessment of subjective product perception for dependence and withdrawal symptoms was conducted after the test treatment usage on Day 30 (+2 Days).

Statistics

Continuous variables were characterized using descriptive statistics, including the number of observations (N), mean, standard deviation (SD), median, minimum, and maximum values. Categorical variables were presented using frequency and percentage. For continuous variables, a comparison from baseline to post-product was conducted employing the Paired t-test. The statistical analysis was performed utilizing R software (Version: 4.2.2) with a significance level set at 5%. Data from withdrawn subjects were excluded from the statistical analysis.

RESULTS

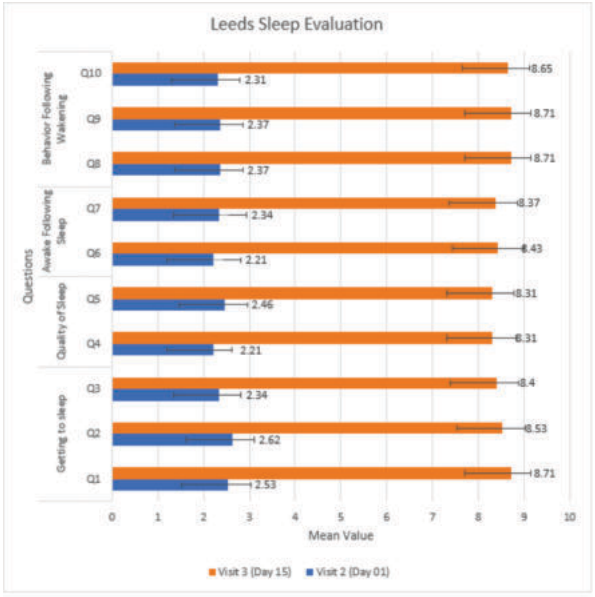
In this study, age of the 32 subjects ranged from 18 to 44 years with mean age 39.6 years. Based on the clinical and statistical analysis, the study showed the following results.

A statistically highly significant improvement, with a p-value < 0.001, was observed from Day 01 to Day 15 in the manner in which subjects fell asleep, encompassing perceptions of difficulty or ease, slowness or quickness, and feeling less sleepy or sleepier after test treatment usage compared to the usual state before test treatment usage. 100% of the subjects exhibited improvement in the way they fell asleep, reporting easier and quicker sleep. The ease to fall asleep, measured on a scale of 10, displayed a substantial mean value increase of 258.8 ± 76.67%, progressing from 2.53 ± 0.5 on Day 01 to 8.71 ± 0.45 on Day 15. Similarly, the sleep onset latency exhibited a significant improvement of 237.50 ± 71.84%, with scores advancing from 2.62 ± 0.49 to 8.53 ± 0.50 during the same period. Additionally, the overall sleepiness, also rated on a scale of 10, demonstrated a remarkable improvement of 271.87 ± 69.87%, rising from 2.34 ± 0.48 on Day 01 to 8.40 ± 0.49 on Day 15.

All 100% of the subjects reported an improvement in the quality of sleep. The participants noted experiencing easier sleep and fewer wakeful periods during their sleep. The paired t-test yielded a p-value of < 0.001, indicating a statistically highly significant difference observed on Day 15 in terms of the quality of sleep. The assessment of restfulness, measured on a scale of 10, revealed a noteworthy improvement of 285.41 ± 64.02%, from 2.21 ± 0.42 on Day 01 to 8.31 ± 0.53 on Day 15. Simultaneously, the evaluation of wakefulness during the day demonstrated a significant enhancement of 251.04 ± 74.89%. Scores progressed from 2.46 ± 0.50 to 8.31 ± 0.47 over the same period. (Figure 01).

All 100% participants in the study, reported improvement in the process of awakening after sleep. Waking up in the morning became easier and required less time following the test treatment. A statistically highly significant difference, with a p-value <0.001, was observed on Day 15 in both the time period and ease associated with waking up. The ease of waking up, quantified on a 10-point scale, demonstrated a substantial increase of 317.19 ± 153.94%, transitioning from a baseline value of 2.21 ± 0.60 on Day 01 to 8.43 ± 0.56 on Day 15. Likewise, the time taken to wake up exhibited a noteworthy improvement of 288.02 ± 139.39%, with scores progressing from 2.34 ± 0.60 to 8.37 ± 0.49 over the same duration.

In assessing the balance and coordination of subjects upon waking on Day 15 in comparison to the baseline, all participants reported heightened alertness and diminished disruption in balance and coordination. The paired t-test revealed a p-value of less than 0.001, indicating a statistically highly significant difference in these parameters. The alertness upon waking, evaluated on a 10-point scale, exhibited a substantial increase of 281.78 ± 75.78%, transitioning from 2.37 ± 0.5 on Day 01 to 8.71 ± 0.45 on Day 15. Furthermore, the assessment of balance and coordination upon waking, also rated on a 10-point scale, demonstrated a noteworthy improvement of 288.02 ± 72.73%, escalating from 2.31 ± 0.47 on Day 01 to 8.65 ± 0.48 on Day 15. (Figure 01).



Q1. 0 = more difficult than usual; 10 = easier than usual
Q2. 0 = slower than usual; 10 = quicker than usual
Q3. 0 = less sleepy than usual; 10 = sleepier than usual
Q4. 0 = more restless than usual; 10 = easier than usual
Q5. 0 = more wakeful period than usual; 10 = less wakeful period than usual
Q6. 0 = difficult to get up; 10 = easier to get up
Q7. 0 = longer time to get up; 10 = shorter time to get up
Q8. 0 = feel tired after wake up; 10 = feel alert after wake up
Q9. 0 = feel tired during day; 10 = feel alert during the day
Q10. 0 = more disrupted balance & co-ordination after waking up; 10 = less disrupted balance & co-ordination after waking up

Figure 01: Change from baseline: Leeds Sleep Evaluation Questionnaire (LSEQ)

Utilizing Pearson's Chi-squared test on the data derived from the sleep diary concerning sleep quality, the outcome indicated a statistically significant difference in sleep quality from Day 01 to Day 15. Following a single dose of the test treatment, 62.5% *(p-value <0.001)* of the subjects reported

experiencing good quality sleep on Day 01, and this percentage increased to 96.9% *(p-value <0.05)* on Day 15 compared to Day 01. Importantly, all subjects demonstrated an improvement in sleep quality, transitioning from poor, mild, or moderate levels to good and very good. (Figure 02).

Figure 02: Quality of Sleep assessment as per Subject Diary.

The time interval required to fall asleep after going to bed exhibited a 6-fold (6x) reduction. Following the use of the test treatment, the average time to fall asleep decreased to 13 minutes, a significant improvement from the baseline average of 80 minutes before test treatment. Prior to taking the test treatment, 88.4% of the subjects took more than 30 minutes to fall asleep, but after the 15-day treatment, 100% of the subjects fell asleep within 30 minutes.

In terms of duration of sleep, a 1.3 times improvement was noted, as reported by the subjects. The average sleep duration increased from 6 hours and 34 minutes before the test treatment to 8 hours and 58 minutes post-test treatment usage. Prior to the test treatment, 80% of subjects felt tired upon morning awakening, while after 15 days of treatment, 100% reported feeling fresh and active. On Day 01, 37% felt no irritability waking-up after a single treatment dose, increasing to 75% by Day 15.

No significant changes were observed in blood parameters, including Complete Blood Count, Erythrocyte Sedimentation Rate, SGPT, SGOT, and Serum Creatinine. However, the test treatment significantly improved sleep quality and may have influenced random blood glucose levels. The paired t-test yielded a p-value < 0.05, indicating a statistically significant 6.32% reduction in random blood sugar levels at Day 15 post-test treatment, with the mean reduced to 105.78±47.03mg/dL from 119.71±74.48 mg/dL. (Table 02) (Figure 03)

Table 02: Change in biochemical parameter – Random Blood Sugar (mg/dL)

Summary Statistics (N=32)	Visit 2 (Day 01)	Visit 3 (Day 15)	CFB	%CFB
n	32	32	32	32
Mean (SD)	119.71 (74.48)	105.78 (47.03)	-13.93 (35.75)	-6.32 (21.21)
Median	98.00	90.50	-7.00	-7.55
Minimum	74.00	77.00	-141.00	-38.68
Maximum	453.00	312.00	63.00	70.00
Within Group p-value*	0.03			

Note: n: Number of non-missing observation
SD: Standard Deviation
*: Paired t-test CFB= Change from Baseline
% CFB = ((Postbaseline - Baseline)/Baseline) *100, visit 01 is considered as baseline

All participants (100%) attested to the efficacy of the test treatment in facilitating faster sleep onset, providing uninterrupted, deep, calm, and restful sleep, aiding in waking up feeling refreshed, reducing anxiety and mental stress, and alleviating difficulty in concentration, and improving general health and well-being. Furthermore, all subjects expressed a favourable response to the taste of the test treatment and encountered no challenges in its oral administration. (Table 03).

Figure 03: Baseline and Post-Baseline Values for Random Blood Glucose (mg/dL)

Table 03: Summary for Subject Perception Questionnaire

Questionnaire	Response	Visit 3 (Day 15)
1. On a scale of 1-9, where will you rank the test treatment in terms of effectiveness in producing faster sleep?	Very Much effective	29 (90.6%)
	Moderately effective	3 (9.4%)
2. 2. On a scale of 1-9, where will you rank the test treatment in terms of effectiveness in providing uninterrupted sleep?	Very Much effective	28 (87.5%)
	Moderately effective	2 (6.25%)
	Extremely effective	2 (6.25%)
3. On a scale of 1-9, where will you rank the test treatment in terms of effectiveness in providing deep and calm sleep?	Very Much effective	22 (68.75%)
	Moderately effective	2 (6.25%)
	Extremely effective	8 (25.0%)
4. On a scale of 1-9, where will you rank the test treatment in terms of waking up fresh in next morning?	Very Much effective	20 (62.5%)
	Moderately effective	1 (3.12%)
	Extremely effective	11 (34.3%)
5. On a scale of 1-9, where will you rank the test treatment in terms of providing restful sleep?	Very Much effective	30 (93.76%)
	Moderately effective	1 (3.12%)
	Extremely effective	1 (3.12%)
6. On a scale of 1-9, where will you rank the test treatment in terms of effectiveness in reducing the anxiety and mental stress?	Very Much effective	23 (71.88%)
	Moderately effective	1 (3.12%)
	Extremely effective	8 (25.0%)
7. On a scale of 1-9, where will you rank the test treatment in terms of effectiveness in reducing difficulty and concentration?	Very Much effective	19 (59.4%)
	Extremely effective	13 (40.6%)
8. On a scale of 1-9, where will you rank the test treatment in terms of packaging?	Liked Extremely	9 (28.2%)
	Liked Very Much	23 (71.8%)

9. On a scale of 1-9, where will you rank the test treatment in terms of taste?	Liked Extremely	9 (28.2%)
	Liked Very Much	22 (68.75%)
	Liked Moderately	1 (3.12%)
10. On a scale of 1-9, where will you rank the test treatment in terms of difficulty in swallowing?	Liked Extremely	2 (6.25%)
	Liked Very Much	29 (90.6%)
	Liked Moderately	1 (3.12%)
11. On a scale of 1-9, where will you rank the test treatment in terms of effectiveness in improving general health and well-being post-usage?	Very Much effective	0 (0.0%)
	Extremely effective	3 (16.67%)
12. On a scale of 1-9, where will you rank the test treatment in terms of overall satisfaction post-usage?	Liked Extremely	15 (46.9%)
	Liked Very Much	17 (53.1%)

A significant reduction was noted in various symptoms, including giddiness, heaviness in the head, headache, exhaustion, yawning, drowsiness, lack of concentration, mental fatigue, and general fatigue. (Table 04)

Table 04: Summary for Overall Health and Well-Being

Parameters	Score	Visit 2 (Day 1)	Visit 3 (Day 15)
Exhaustion	0	0 (0.0%)	16 (50.0%)
	1	6 (18.8%)	16 (50.0%)
	2	11 (34.4%)	0 (0.0%)
	3	15 (46.9%)	0 (0.0%)
	p-value		<0.001
Giddiness	0	28 (87.5%)	31 (96.9%)
	1	2 (6.25%)	1 (3.1%)
	2	2 (6.25%)	0 (0.0%)
	p-value		<0.001
Headache	0	0 (0.0%)	25 (78.1%)
	1	0 (0.0%)	7 (21.9%)
	2	23 (71.9%)	0 (0.0%)
	3	9 (28.1%)	0 (0.0%)
	p-value		<0.001
Heaviness in Head	0	0 (0.0%)	26 (81.2%)
	1	0 (0.0%)	6 (18.8%)
	2	31 (96.9%)	0 (0.0%)
	3	1 (3.1%)	0 (0.0%)
	p-value		<0.001
Yawning	0	0 (0.0%)	29 (90.6%)
	1	0 (0.0%)	3 (9.4%)
	2	30 (93.7%)	0 (0.0%)
	3	2 (6.3%)	0 (0.0%)
	p-value		<0.001
Drowsiness	0	0 (0.0%)	22 (68.7%)
	1	0 (0.0%)	9 (28.1%)
	2	10 (31.3%)	1 (0.2%)
	3	22 (68.7%)	0 (0.0%)
	p-value		<0.001
Lack of Concentration	0	0 (0.0%)	23 (71.8%)
	1	0 (0.0%)	9 (28.2%)
	2	15 (46.8%)	0 (0.0%)
	3	17 (53.2%)	0 (0.0%)
	p-value		<0.001
Mental Fatigue	0	0 (0.0%)	20 (62.5%)
	1	0 (0.0%)	12 (37.5%)
	2	17 (53.2%)	0 (0.0%)
	3	15 (46.8%)	0 (0.0%)
	p-value		<0.001

None of the subjects reported a proclivity for the test treatment or the development of habituation, and no adverse effects such as nausea, vomiting, headache, dizziness, anxiety, or stress were documented following the discontinuation of the test treatment on Day 30. Throughout the study duration, no adverse events were reported by either the subjects or the investigator.

DISCUSSION

Indigenous to the Indian subcontinent, *Valeriana wallichii*, or the Indian valerian *Tagar*, has long been used to induce sleep and relaxation as well as have sedative and anxiolytic effects in Ayurvedic medicine. The plant's root is commonly used in medicine to treat headaches, tension, anxiety, and depression, among other conditions. These therapeutic qualities contribute to the overall enhancement of sleep quality.(Miller, 2023)

A study published in Advances in Therapy investigated the impact of a standardized extract of *Valeriana officinalis* on overall sleep quality in individuals with sleep complaints. Conducted over an 8-week period, the study involved 80 adult subjects. Results demonstrated significant improvements in various sleep parameters within the *Valeriana officinalis* group, including reduced sleep latency, increased actual sleep time, and enhanced sleep efficiency, as evaluated through wrist actigraphy. Moreover, participants in this group reported a decrease in anxiety and daytime drowsiness, coupled with an augmented sensation of waking up feeling refreshed. (Chandra Shekhar, Joshua, & Thomas, 2023)

Melatonin is a hormone that plays a pivotal role in regulating the body's circadian rhythm. This intrinsic rhythm manages physiological changes in response to the day-night cycle, influencing states of sleepiness and wakefulness. Moreover, research indicates a correlation between magnesium deficiency and reduced melatonin levels, indicating the importance of magnesium in the regulation of the fundamental biological cycles. (MediLexicon International, 023)

Another study by **Nutrition & Metabolism** found that co-supplementation with magnesium and melatonin demonstrated favourable impacts on both sleep quality and total testosterone levels. Notably, melatonin supplementation alone was associated with a significant reduction in PSQI (Pittsburgh Sleep Quality Index) score.(Alizadeh, et al., 2021)

Furthermore, studies involving tryptophan supplements, specifically those containing one gram or */more, have revealed their potential to enhance subjective sleepiness and reduce the time to fall asleep, particularly in individuals experiencing mild insomnia.(Zhao, Tuo, Wang, & Zhao, 2020)

The test treatment 'Zzowin' is more than just a melatonin tablet. It is a phyto-nutraceutical supplement that contains 5 mg Melatonin along with herbs like *Tagar* (*Valeriana wallichii*) and other nutrients–each of which has been proven to treat sleep problems. This herbal amalgamation is distinctive for its integration of proven sleep-cycle treatments such as melatonin, magnesium, and L-tryptophan, among others, along with traditional herbal medicine known for its established effectiveness.

The outcomes of our investigation affirm the test treatment as an efficacious intervention for individuals with sleep disorders, showcasing significant enhancements in sleep parameters, including sleep duration, sleep quality, sleep onset and waking time, as well as improvements in -balance, coordination, and morning energy levels. The tablet also proved to be safe in all the subjects. A comparison with conventional treatments of sleep problems such as benzodiazepines, makes this sleep supplement stand out for

its tolerability.

However, it is imperative to acknowledge certain limitations in this study. The research involved a relatively small cohort, necessitating further investigations with larger populations to establish the treatment's efficacy and safety across diverse populations. Additionally, the study lacks a comparative nature with a placebo or similar substances, warranting consideration in future investigations.

CONCLUSION

The study concludes that the test treatment demonstrated efficacy in a cohort of healthy adult subjects within the age range of 18–65 years, suffering from sleep deprivation due to shift work disturbance, jet lag, and/or mental stress. Examination of various parameters revealed a statistically significant improvement in sleep quality, sleep onset latency, and sleep duration, leading to positive changes in symptoms such as giddiness, heaviness in the head, headache, exhaustion, yawning, drowsiness, lack of concentration, mental fatigue, and overall fatigue. Notably, no indications of craving for the test treatment, habituation, or adverse effects such as nausea, vomiting, headache, dizziness, anxiety, or stress were reported upon discontinuation. Throughout the trial period, no clinically significant changes or adverse events were observed. For individuals experiencing sleep disorders stemming from disturbances like shift work, jet lag, or mental stress, the non-habit-forming 'Zzowin' tablet emerges as a potentially effective treatment for restoring balance to the sleep cycle.

Acknowledgements

The authors extend their sincere gratitude to Charak Pharma and Vedistry Private Limited, along with their committed teams, for their collaborative support. Appreciation is conveyed to the NovoBliss Research study team for overseeing the *in-vivo* clinical study, the statistical team for their expertise in data analysis, and the scientific writing team for their invaluable assistance in manuscript preparation. Special acknowledgment is due to Ms. Nistha Jani for her comprehensive study coordination and to Dr. Arnav Purani for his contributions to medical writing. Heartfelt appreciation is expressed to all study participants for their significant and valuable contributions to the research endeavour.

Declarations

Funding: Vedistry Private Limited, India
Conflict of interest: Authors declare there is no conflict of interest.
Ethical approval: The study was approved by the Institutional Ethics Committee.
ClinicalTrials.gov Registration: Yes, NCT05853757
CTRI Registration: Yes, CTRI/2023/04/051587

REFERENCES

1. Alizadeh, M., Karandish, M., Asghari Jafarabadi, M., Heidari, L., Nikbakht, R., Babaahmadi Rezaei, H., & Mousavi, R. (2021). Metabolic and hormonal effects of melatonin and/or magnesium supplementation in women with polycystic ovary syndrome: A randomized, double-blind, placebo-controlled trial. *Nutrition & Metabolism, 18*. doi:10.1186/s12986-021-00586-9
2. Chandra Shekhar, H., Joshua, L., & Thomas, J. V. (2023). Standardized extract of *Valeriana officinalis* improves overall sleep quality in human subjects with sleep complaints: A randomized, double-blind, placebo-controlled, clinical study. *Advances in Therapy*. doi:10.1007/s12325-023-02708-6
3. Guadagna, S., Barattini, D. F., Rosu, S., & Ferini-Strambi, L. (2020). Plant extracts for sleep disturbances: A systematic review. *Evidence-Based Complementary and Alternative Medicine*, 1-9. doi:10.1155/2020/3792390
4. Mayo Clinic. (2016, October 15). *Insomnia*. (Mayo Foundation for Medical Education and Research) Retrieved from Mayo Clinic: https://www.mayoclinic.org/diseases-conditions/insomnia/diagnosis-treatment/drc-20355173
5. MediLexicon International. (023, August 1). *Magnesium for sleep: Benefits and how to take it*. (MediLexicon International) Retrieved from Medical News Today: https://www.medicalnewstoday.com/articles/magnesium-for-sleep
6. Miller, R. (2023, November 6). *Valeriana officinalis vs. Valeriana wallichii*. Retrieved from Zhi Herbals: https://www.zhiherbals.com/blog/valeriana-officinalis-vs-valeriana-wallichii
7. RootBabes. (2022, April 25). *7 Ayurvedic medicine for Sleep Disorders and Quality Rest*. Retrieved from RootBabes: https://rootbabes.com/ayurvedic-medicines-for-sleep-disorders/
8. Shinjyo, N., Waddell, G., & Green, J. (2020). Valerian root in treating sleep problems and associated disorders—a systematic review and meta-analysis. *Journal of Evidence-Based Integrative Medicine, 25*. doi:10.1177/ 2515690x 20967323
9. Telles, S. (2015, May). Ayurvedic doshas as predictors of sleep quality. *Medical Science Monitor* (e-ISSN 1643-3750), 7. doi:10.12659/MSM.893302
10. Zhao, M., Tuo, H., Wang, S., & Zhao, L. (2020). The effects of dietary nutrition on sleep and sleep disorders. *Mediators of Inflammation*, 1-7. doi:10.1155/2020/ 3142874