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ARTICLE



Napping between scylla and charybdis of N1 and N3: latency to N2 in a brief afternoon nap can be reduced by binaural beating

Dmitry E. Shumov^a, Irina A. Yakovenko^a, Vladimir B. Dorokhov^a, Dmitry S. Sveshnikov^b, Elena B. Yakunina^b, Zarina V. Bakaeva^b, Aleksey V. Vinokurov^c and Arcady A. Putilov ^b

^aLaboratory of Sleep/Wake Neurobiology, Institute of Higher Nervous Activity and Neurophysiology, Russian Academy of Sciences, Moscow, Russia; ^bDepartment of Normal Physiology, Medical institute of the People's Friendship University of Russia, Moscow, Russia; ^cResearch Department, "Mind Technology" LLC, Moscow, Russia

ABSTRACT

Afternoon nap is regarded as a potent behavioral strategy minimizing sleepiness and fatigue. The benefits of afternoon naps require the accumulation of, at least, 3 min of stage 2 sleep. However, there are practical disadvantages of nap longer than 10–15 min, such as greater length of time consumed by the nap, appearance of slow wave sleep causing greater sleep inertia right after the nap, and possible detrimental impact of such nap on subsequent nocturnal sleep. We previously found that a binaural beat treatment that is a dichotic presentation of two almost equivalent pure tones with slightly different frequencies led to a reduction of latency to stage 2 sleep. To replicate this result and to examine whether such reduction leads, in turn, to the earlier appearance of slow wave sleep, we asked 23 and 21 healthy volunteers to nap in the afternoon for 30 and 20 min, respectively. Almost half of volunteers showed latency to stage 2 longer than 17 min, but most of them responded to the treatment by its reduction. The following occurrence of slow wave sleep reduced level of alertness self-assessed right after the nap. We concluded that latency to stage 2 sleep might be experimentally challenged by binaural beating.

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Acoustic stimulation; falling asleep; fatigue; sleepiness; alertness; afternoon deep

Introduction

A relatively brief afternoon nap may mitigate alertness and performance deficits (Blanchfield et al. 2018; Waterhouse et al. 2007). Therefore, such a nap has been regarded a potent behavioral strategy minimizing sleepiness, fatigue, and impairments of cognitive and physical functioning (Takahashi et al. 2004; Caldwell et al. 2009; Keramidas et al. 2018). The question of nap's brevity and architecture is of both practical and theoretical importance. The efficacy of napping as a sleepiness and fatigue counter-measure was related to the nap duration (Tietzel and Lack 2001, 2002; Hayashi et al. 2005). Moreover, the sleep-wake regulating mechanisms underlying the beneficial action of the nap on levels of alertness and performance might be

CONTACT Arcady A. Putilov I putilov@ngs.ru Daboratory of Sleep/Wake Neurobiology, Institute of Higher Nervous Activity and Neurophysiology, Russian Academy of Sciences, 11, Nipkowstr., Berlin, 12489 Germany © 2019 Informa UK Limited, trading as Taylor & Francis Group

uncovered by experimental manipulating with the nap duration and architecture (Brooks and Lack 2006; Ushimi and Hayashi 2008).

Although longer naps may be recuperative, any benefit of such naps must be weighed against their three practical disadvantages: (1) greater length of time consumed by the nap, (2) greater sleep inertia right after the nap, and ((3) possibility of detrimental impact on subsequent nocturnal sleep (Brooks and Lack 2006). Particularly, it was shown that the 30-min nap after a night with restricted sleep duration produced a period of impaired alertness and performance immediately after napping, indicative of experiencing sleep inertia right upon awakening from the nap (Tietzel and Lack 2001). The 20-min nap following restricted nocturnal sleep produced no significant benefits right after the nap, suggesting some immediate sleep inertia, while the 10-min nap produced a pronounced increase in alertness and performance immediately after napping (Brooks and Lack 2006). Since sleep inertia appears to be directly related to the duration of slow-wave sleep during a sleep episode, it follows that this negative effect on alertness and performance levels will be greater following longer naps that typically contain more slow-wave activity than shorter naps (Dinges et al. 1987; Muzet et al. 1995; Rosekind et al. 1995; Jewett et al. 1999; Tassi and Muzet 2000; Signal et al. 2012). Ushimi and Hayashi (2008) examined more than 100 polysomnograms of university students and found that slow wave sleep did not appear during a nap of less than 15-minute duration, but did appear at least 9 min after the first sleep spindle that is associated with entering into stage 2 sleep.

There is no evidence suggesting that the napping benefits arose either from the nap with stage 1 sleep or from the nap including only the onset of stage 2 (first sleep spindle). Hayashi et al. (2005) concluded that the napping benefits might be derived from the accumulated 3 min of stage 2 sleep per se. Similarly, Tietzel and Lack (2002) found that the 30- and 90-s naps produced no significant post-nap benefits and they concluded that the onset of stage 1 sleep did not appear to be the mechanism underlying the benefits of brief naps. It was hypothesized that the benefits of brief naps following mild nocturnal sleep restriction may require the accumulation of a brief fixed amount of stage 2 sleep or a brief fixed period of total sleep of any stage between 90 s and 10 min in duration (Brooks and Lack 2006). However, further investigation is needed to clarify the mechanisms underpinning the immediate recuperative benefits of brief nap gained on the interval between entering into stage 2 and entering into stage 3 sleep.

Sleep-improving effects were suggested for a binaural beat treatment that is a dichotic presentation of two almost equivalent pure tones with slightly different frequencies leading to virtual beat perception by the brain (Abeln et al. 2014; Jirakittayakorn and Wongsawat 2018). In the previous study of 14 healthy volunteers napping in the afternoon for 30 min we showed that latency to stage2 sleep might be shortened by treatment with binaural beats in combination with monotonous sound as compared to intervention with monaural beats of the same spectrum combined with monotonous sound (Shumov et al. 2017). This observation opens the possibility of experimental manipulations with latency and duration of stage 2 sleep. Such manipulations might be, in particular, used for addressing questions about the role playing by this stage in beneficial effects of a short afternoon nap on sleepiness, fatigue, motivation, performance, memory formation, etc.

Consequently, our two new experimental studies of 20- and 30-min nap architecture in two (with and without binaural beat treatment) conditions were aimed at (1)

examination of replicability of our previous result suggesting that a binaural beat treatment leads to shortening latency to stage 2 sleep, the stage associated with beneficial effects of nap, and (2) determination of whether such a shortening leads, in turn, to increase in amount of stage 3 (slow wave sleep), the stage associated with sleep inertia. We anticipated a significant reduction of latency to stage 2 sleep in the nap of 30- and 20-min duration and, in turn, an earlier occurrence of stage 3 sleep in response to a binaural beat treatment.

Methods

Two experimental studies of afternoon napping attempts of either 30- or 20-min duration were performed in accordance with the ethical standards laid down in the Declaration of Helsinki. Their protocols were approved by the Ethics Committee of the Institute of Higher Nervous Activity and Neurophysiology. Each participant was studied individually in two different conditions, one was with/without and another was without/ with a binaural beat treatment. Participants were informed in detail about the experimental procedures, and written consent was obtained from each participant.

Participants of the first (30-min nap) study were males (n = 15) and females (n = 8) aged between 19 and 32 years (average ± standard deviation: 23.3 ± 4.5). Participants of the second (20-min nap) study were young males (n = 13) and females (n = 8) with ages from 18 to 22 years (average ± standard deviation: 20.1 ± 0.7). They were randomly assigned to the first napping experiment either with or without binaural beat treatment in the afternoon (between 1 p.m. and 4 p.m.). At the next day or later (but not later than in three weeks) they napped again at the same hour of the day either without or with the treatment.

The two experimental studies were slightly different in active treatments counterbalanced with either control stimulation or untreated condition. In the first study, the control stimulation was monotonous sound with pink noise background delivered via stereo headphones (Bose QC-25) for the first 15 min of 30-min napping attempt, and the active treatment was the same monotonous sound combined with binaural beating (frequencies of 0.5, 4, and 2 Hz) for the first 15 min of 30-min napping attempt. No sound was delivered via stereo headphones in the following 15 min of napping attempt. In the second study, participants were left without treatment (i.e. no any sound was delivered via stereo headphones) for the whole interval of 20-min napping attempt, while, in the later or previous napping attempt, they were treated for 20-min by binaural beating (frequencies of 2 and 4 Hz) combined with relaxing music. Sound intensity was adjusted individually, mostly in the interval between 55 and 57 dB SPL (Sound Pressure Level).

During the application of electrodes and polysomnographic recordings, the participants were lying in bed in the sleep laboratory under dim light. After switching off the light, they were recorded in resting condition for 3 min, then they were asked to sleep until hearing the awakening signal from the nursing staff, and they were recorded again for three more minutes in resting condition.

The recordings were performed via a 16-channel wireless system ("Neuropolygraph 24", Neurotech, Taganrog, Russia). A standard monitoring montage was used for polysomnographic recordings including 1 chin electromyogram channel, 2 electro-oculogram channels, and 13 EEG channels (all electrodes were placed in accord with the International 10–20 system of electrode placement). The recorded signals were

Polysomnographic measure, min				Latency to stage				Amount of stage	
Duration				N1		N2		N3	
Sub-sample	of nap	n	Treatment	Mean	SEM	Mean	SEM	Mean	SEM
Whole	30	23	Without	6.565	1.229	12.870	1.451	5.174	1.147
			With	4.935	0.759	10.696	1.114	6.391	1.201
	20	21	Without	4.571	1.287	12.310	1.519	0.810	1.201
			With	4.286	0.794	10.095	1.166	1.333	1.257
Stage 2	30	13	Without	2.846	1.073	7.423	0.863	9.154	1.355
latency			With	3.577	0.723	8.038	0.985	9.115	1.612
<17	20	12	Without	2.000	1.117	6.875	0.899	1.417	1.410
min			With	3.708	0.752	8.292	1.025	1.417	1.678
Stage 2	30	6	Without	10.417	1.580	19.917	1.271	0.000	1.994
latency			With	3.833	1.064	10.25	1.450	4.667	2.373
>17	20	6	Without	10.333	1.580	19.500	1.271	0.000	1.994
min			With	4.417	1.064	8.750	1.450	1.833	2.373

Table 1	. Pol	ysomnogi	raphic	characteristics	of na	apping	attempts i	n two	conditions

N1, N2, and N3: Stage 1, stage 2, and slow wave sleep; Stage 2 latency <17 or >17 min: Two of three Subsamples of the Whole sample showing latency to N2 shorter than 17 min in both conditions (Without and With binaural beat treatment) or longer than 17 min in only one of two conditions (Without treatment); Duration of nap: The first or second experiments with napping attempt of either 30- or 20-min duration, respectively; *n*: Number of study participants in Whole sample and Subsamples; Treatment: Two (Without and With treatment) conditions in each of two experimental studies; SEM: Standard Error of Mean. For calculation of Mean, any latency longer than 20 min was reduced to 20.0 min.

conditioned by the high-pass, low-pass, and notch filters (0.5, 35, and 50 Hz, respectively), sampled and stored on a hard disc with a frequency of 500 Hz.

Conventional scoring (lber et al. 2007) of the 20- and 30-min naps was performed on 30-sec epochs of the polysomnographic records. The epochs were classified into stages including three stages of NREM (Non-Rapid Eye Movement) sleep, stage 1 sleep or N1, stage 2 sleep or N2, and slow-wave sleep or N3 (stage 3). In the present report, the three major polysomnographic characteristics of short nap were analyzed: latencies to N1 (Table 1) and N2 (Table 1 and Figure 1), and amount of N3 (Table 1).

The potential beneficial effects of an afternoon nap on the levels of well-being, alertness, and mood were evaluated with the so-called "WAM test" (Doskin et al. 1973). This test was administered before and after a napping attempt (Figure 2). The test consists of three 10-item sub-scales named Well-being, Alertness, and Mood. Each sub-scale includes 10-word pairs with a 7-point response scale printed between each pair of words (Doskin et al. 1973).

This 30-item list can be exemplified by the first six pairs of words (Doskin et al. 1973):

- (1) Good health 3210123 Bad health
- (2) Feel strong 3210123 Feel weak
- (3) Passive 3210123 Active
- (4) Sedentary 3210123 Agile
- (5) Gleeful 3210123 Sad
- (6) Good mood 3210123 Bad mood

These six pairs belong to the sub-scales of Well-being, Alertness, and Mood (pairs 1 and 2, 3 and 4, and 5 and 6). Each of 30 response scales ("3210123") was transformed, depending upon its polarity, into either "3210–1–2–3" or "-3–2–1023" response scale, and then 10 responses were averaged to obtain raw scores on Well-being, Alertness, and Mood scales



Figure 1. Latency to stage 2 during the 30- or 20-min napping attempts in two conditions.

Only the first 20 min of either 30- or 20-min napping attempt are shown (a or b, respectively). The horizontal lines depict 17-min latency to stage 2 that results in spending, at least, 3 min in this stage during the first 20-min of a napping attempt (this was supposed to be the minimal duration required for the napping benefits; Hayashi et al. 2005). Without or With: Napping attempt in one of two conditions, either Without or With binaural beat treatment.

each ranging from -3 to 3. Negative scores mean feeling bad, sleepy, and sad, and positive score means feeling good, alert, and cheerful. For reducing the influence of individual differences in self-rating that increased both pre- and post-napping variation in sample-averaged score (Figure 2(a)), scores of each study participant were additionally normalized by transforming each raw score relative to the participant's average pre-napping raw score for both – with and without treatment – conditions (Figure 2(b)).

All statistical comparisons were performed with the Statistical Package for the Social Sciences (SPSS), version 22.0 (IBM, Armonk, NY, USA). A Student's paired *t*-test and one-, two- or three-way repeated measure ANOVAs (rANOVAs) with none, one or two independent

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Figure 2. Alertness score before and after napping attempts in two conditions.

A or B: Either raw alertness score or relative alertness score expressed as a deviation from mean score obtained Before treatment in two conditions. Without and With: 20-min napping attempt in one of two conditions, Without and With binaural beat treatment. Before and After: Self-assessment obtained right Before and right After a napping attempt. Subsample, N2 latency <17 and Subsample, with N3: Subsample of participants showing latency to stage 2 sleep shorter than 17 minutes in both conditions but without further entering into slow wave sleep and Subsample of participants who either showed longer than 17-min latency in, at least, one of two conditions (Without treatment) or entered into slow wave sleep (n = 12 and 32 or 11 and 33 in condition either With or Without treatment, respectively); SEM: Standard Error of Mean score.

factors were employed for such comparisons. For each rANOVA, Mauchly's test was conducted to assess the sphericity and, if necessary, the Greenhouse-Geiser correction was used to adjust the degrees of freedom.

Results

As predicted, paired *t*-test revealed significant reduction of latency to stage 2 sleep in the treatment condition, whereas latency to stage 1 and amount of slow wave sleep did not change ($t_{43} = -2.369$, p < 0.05, $t_{43} = -1.285$, p > 0.05, and $t_{43} = 1.145$, p > 0.05, respectively). Similarly, significant main effects and significant interaction were not yielded by two-way

rANOVA of latency to stage 1 sleep with repeated measure "Condition" (without or with binaural beat treatment) and independent factor "Nap duration" (30- or 20-min napping attempt of the first or secondexperimental study, respectively). As predicted, the same rANOVA of latency to stage 2 sleep yielded significant main effect of "Condition" ($F_{1,42} = 5.475$, p < 0.05), and the same rANOVA of the amount of stage 3 sleep yielded significant main effect of "Nap duration" ($F_{1,42} = 9.745$, p < 0.01). The results suggested that latency to stage 2 sleep was reduced by treatment with binaural beats irrespective of nap duration and that enlargement of stage 3 sleep occurred with lengthening of nap duration (from 20 to 30 min) irrespective of conditions (Table 1).

As it is illustrated in Figure 1, the minority of study participants (4 of 23 and 3 of 21 in the 30- and 20-min napping attempts, respectively) showed latency to stage 2 sleep longer than 17 min suggesting that did not get a chance to spend, at least, 3 min in this stage during the first 20 min of their napping attempt. In contrast, more than half of the participants (13 and 12, respectively) showed latency to stage 2 sleep shorter than 17 min in both (without and with treatment) conditions. The remaining 12 participants (6 and 6, respectively) showed latency to stage 2 sleep shorter than 17 min only when treated with binaural beats.

The characteristics of two latter subsamples of the whole sample (25 and 12 participants) are given in Table 1 in addition to the characteristics of the whole sample. As indicated by twoway rANOVAs of the last subsample with repeated measure "Condition" and independent factor "Nap duration", the effect of binaural beat treatment was significant for both latencies and tended to reach a significant level for the amount of stage 3 sleep ($F_{1.10} = 18.607$, p < 0.01, $F_{1.10} = 74.765$, p < 0.001, $F_{1.10} = 4.557$, p = 0.059 for latencies to N1 and N2, and amount of N3, respectively). In contrast, the same rANOVAs of the subsample with shorter than 17-min latency to stage 2 sleep in both conditions revealed significant lengthening of latency to stage 1 sleep in treatment condition ($F_{1,23}$ = 4.333, p < 0.05), but condition did not have any significant impact on latency to stage 2 sleep and amount of slow wave sleep (Table 1 and Figure 1). Significant main effect of independent factor "Nap duration" was revealed only for the amount of slow wave sleep and only in the subsample of participants showing shorter than 17-min latency to stage 2 sleep in both conditions ($F_{1,23} = 13.632$, p = 0.001). In two of six participants in the subsample showing longer than 17-min latency to stage 2 sleep under control stimulation (30-min nap), the amount of slow wave sleep under treatment was as high as 11.5 and 16 min. In three of six participants in this subsample who showed longer than 17min latency to stage 2 sleep in the untreated condition (20-min nap), the amount of stage 3 in treatment condition was as high as 2.5, 3.0, and 5.0 min.

Subjective self-reports did not differ in conditions without and with active treatment. For instance, self-reported duration of night sleep before nap was similar for two conditions (e.g. average duration \pm standard deviation were 345.6 \pm 146.4 and 355.9 \pm 141.6 min, respectively, paired $t_{43} = -0.368$, p > 0.05).

Similarly, Alertness score reported before and after napping attempt did not differ (Figure 2). Consequently, significant main effects and significant interaction were not yielded by two-way rANOVA of Alertness score in either 20-min or 30-min nap. In the subsample of participants with latency shorter than 17 min in both treated and untreated conditions, only a tendency for a significant effect of a nap was revealed by two-way rANOVA with repeated measures "Condition" and "Nap effect" (e.g. before and after 20-min nap: F1,11 = 3.927, p = 0.073). Even after reduction of individual variation in

pre-nap alertness level (Figure 2(a)) by calculating a relative score, Alertness score did not show a profound increase after the nap (Figure 2(b)).

As compared to changes in Alertness score caused by the naps, changes in Well-being and Mood scores were even less pronounced and, therefore, like changes in Alertness score, they failed to reach a statistically significant level (data not shown).

Comparison of individual differences and differences between naps with different architecture suggested that, as expected, Alertness score after nap did not show a linear increase with rising latency to stage 2 sleep. Although a shorter rather than longer latency was, in general, associated with higher Alertness score after napping, the effect was found to be dependent upon the presence of slow wave sleep. As it is illustrated in Figure 2, the post-napping level of alertness was somewhat higher than the pre-napping level for a shorter latency to stage 2 sleep, but the post-napping level decreased rather than increased relative to the pre-napping level if, during further napping, a participant entered into stage 3 sleep (Figure 2). Significant interaction between "Nap effect" and independent factor "Subsample" (with or without slow wave sleep) was yielded by three-way rANOVA with repeated measures "Nap effect" and "Condition" ($F_{142} = 6.720$, p < 0.05).

Discussion

A relatively brief afternoon nap has been regarded a potent behavioral strategy minimizing perceived sleepiness and fatigue, cognitive, and physical performance impairments, deficit of motivation, etc. (Blanchfield et al. 2018; Takahashi et al. 2004; Waterhouse et al. 2007; Caldwell et al. 2009; Keramidas et al. 2018). The experimental nap studies led to a suggestion that the benefits of afternoon naps require the accumulation of, at least, 3 min of stage 2 sleep (Hayashi et al. 2005), and, the necessity of further prolongation of nap must be weighed against such its practical disadvantages as greater length of time consumed by the nap, entering into slow wave sleep causing greater sleep inertia, and possible detrimental impact of such nap on subsequent nocturnal sleep (Brooks and Lack 2006). We previously showed that a binaural beat treatment produced reduction of latency to stage 2 sleep (Shumov et al. 2017). Healthy volunteers napped twice in the afternoon in two new experiments with 30- or 20-min duration of napping attempt (n = 23 and 21, respectively) for 2) testing replicability of this result and 2) examining whether such a reduction can, in turn, lead to increase in the amount of slow wave sleep. We found that the majority of those volunteers who showed a relatively long latency to stage 2 sleep (>17 minutes) when napping under control stimulation or in untreated condition (10 and 9, respectively) responded to the binaural beat treatment (1) by reduction of this latency (6 and 6, respectively) and (2) following increase of amount of slow wave sleep.

In overall, these results of two experimental studies confirmed our previous finding indicating responsiveness of latency to stage 2 sleep to binaural beating (Shumov et al. 2017) and they showed the expected relationship between this latency and amount of the following slow wave sleep in a napping attempt longer than 17 min. It seems that, sometimes, a study participant spent in slow wave sleep more than 10 min of 30-min napping attempt due to a relatively rapid entering into stage 2 sleep. The results of the treatment studies provided a possibility to address the questions about the contribution of stage 2 sleep to the beneficial effects of a short afternoon nap on sleepiness, fatigue, motivation, performance, memory formation, etc., by using this treatment for experimental manipulations with latency and duration of this stage.

Several limitations of our experimental studies require acknowledgment. Although sample sizes were big enough to confirm the significance of the effects of binaural beating on nap architecture, they were too small to describe in more detail the pattern of relationship between such treatment and latency to stage 2 sleep. It seems that the treatment cannot cause further reduction of the latency when it is already relatively short under control simulation/untreated condition, but the treatment promote the reduction of the latency more effectively when it is relatively long in such condition.

Moreover, the samples were not big enough to describe in more detail the nonlinear pattern of relationship between reduction of latency to stage 2 sleep and subjective self-assessments. It seems that the results on such relationship are in agreement with the previously reported findings (Tietzel and Lack 2001, 2002; Hayashi et al. 2005; Brooks and Lack 2006). They suggested that, (1) when a total duration of NREM sleep states is relatively long, a nap might be disadvantageous for immediate alertness level due to appearance of slow-wave sleep that is regarded to be the cause of sleep inertia (Dinges et al. 1987; Muzet et al. 1995; Rosekind et al. 1995; Jewett et al. 1999; Tassi and Muzet 2000; Signal et al. 2012, (2) that a nap without stage 2 sleep might not be advantageous for this level because only the start of stage 2 might be regarded as a reliable boundary between wakefulness and sleep rather than the start of stage 1 (see, e.g. De Gennaro et al. 2001), and (3) that only a nap with a relatively short total duration of NREM sleep states lacking stage of slow wave sleep might be advantageous for this level (Hayashi et al. 2005; Ushimi and Hayashi 2008).

Another limitation of our experimental studies is the implication of subjective rather than objective assessments for evaluation of the association between benefits of nap and its polysomnographic architecture. Applying objective performance tests right before and right after afternoon naps would be necessary for future studies for documenting possible benefits of such naps for cognitive and physical functioning.

Conclusions

The binaural beat treatments of 23 or 21 participants of two experimental nap studies were able to cause reduction of latency to stage 2 sleep in the majority of those participants who were characterized by a relatively long latency to this stage either under control stimulation or in untreated condition of either 30- or 20-min napping attempts (6 of 10 or 6 of 9, respectively). As expected, a significant decrease in alertness level right after napping was seen in naps with slow wave sleep irrespective of stage 2 latency. Therefore, the binaural beat treatment provides a possibility to experimentally challenge latency and duration of stage 2 sleep for deepening knowledge about the contribution of this stage to the beneficial effects of a short afternoon nap on sleepiness, fatigue, motivation, performance, memory formation, etc.

Disclosure statement

No potential conflict of interest was reported by the authors.

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ORCID

Arcady A. Putilov in http://orcid.org/0000-0003-2779-9046

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