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Report

Interfering with Theories of Sleep and Memory: Sleep, Declarative Memory, and Associative Interference

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Summary

Mounting behavioral evidence in humans supports the claim that sleep leads to improvements in recently acquired, nondeclarative memories. Examples include motor-sequence learning [1, 2]; visual-discrimination learning [3]; and perceptual learning of a synthetic language [4]. In contrast, there are limited human data supporting a benefit of sleep for declarative (hippocampus-mediated) memory in humans (for review, see [5]). This is particularly surprising given that animal models (e.g., [6–8]) and neuroimaging studies (e.g., [9]) predict that sleep facilitates hippocampus-based memory consolidation. We hypothesized that we could unmask the benefits of sleep by challenging the declarative memory system with competing information (interference). This is the first study to demonstrate that sleep protects declarative memories from subsequent associative interference, and it has important implications for understanding the neurobiology of memory consolidation.

Results

Participants first learned a list of word-pair associates (A_i-B_i), followed by a 12 hr, off-line, retention period containing sleep or wakefulness. After this retention period, but prior to testing, the sleep and wake groups were each split into interference and no-interference conditions (Figure 1). Subjects in the interference conditions learned a new list of word pairs (A_i-C_i) 12 min prior to testing, whereas the no-interference subjects went directly to testing. For all participants, the primary outcome was mean percent cued recall of the target words (B_i).

We performed pairwise comparisons of individual groups by using two-tailed t tests (assuming unequal variances). Statistical analyses for recall performance were conducted on data (proportion of correct answers) after arcsine transformation of all measures [10]. (Means, standard deviations, and standard errors are presented numerically and graphically in their untransformed form.) In the no-interference conditions, mean recall was marginally higher in the sleep group (mean [M] = 94%, standard deviation [SD] = 7), than in the wake group (M = 82%, SD = 17), $t(18) = 1.97$, $p = .064$. However, in the interference conditions, there was a large and highly significant difference between the sleep-interference group (M = 76%, SD = 17) and wake-interference (M = 32%, SD = 19), $t(22) = 5.34$, $p < .0001$ (Table 1). We also performed a two-way, between-subjects ANOVA (n = 48) demonstrating significant main effects of sleep [$F(1,47) = 26.93$, $p < .0001$] and interference [$F(1,47) = 46.17$, $p < .0001$], as well as a significant sleep-by-interference interaction [$F(1,47) = 5.84$, $p = .02$] (Figure 2).

We also examined a number of outcome variables that do not directly impact our hypothesis but that address a common concern in sleep research: circadian performance. We compared the total number of trials necessary for a subject to learn all the word pairs at different times of day and found no significant differences between training in the morning (a.m.) and evening (p.m.) [A-B lists – p.m.: M = 105, SD = 23; a.m.: M = 118, SD = 42; $t(36) = -1.26$, $p = .22$; and A–C lists – p.m.: M = 99, SD = 23; a.m.: M = 115, SD = 39; $t(18) = -1.2$, $p = 0.23$]. We also compared second-list recall (C of A–C) at 12 min after training and found no significant differences between the morning and evening performance [a.m.: M = 96%, SD = 6; p.m.: M = 94%, SD = 8; $t(21) = 0.71$, $p = 0.49$].

To further address the concern for time-of-day effects, we ran an additional, independent group: 24 hr pm to pm, with interference (24-hr-PM-I). Participants in this group (n = 12) underwent the same screening, training, and testing procedures as those in the two 12 hr interference groups, Sleep-I and Wake-I. However, unlike the Sleep-I and Wake-I groups, this 24-hr-PM-I group was tested and trained at the same time of day (9 p.m.). Performance in this 24-hour-PM-I group (M = 71%, SD = 25) was nearly identical to that in the Sleep-I group, $t(20) = -0.32$, $p = 0.75$, and significantly better than that in the Wake-I group, $t(21) = 4{,}13$, $p < .001$ (Figure 3).

Discussion

This is the first study to demonstrate that sleep protects declarative memories from subsequent, associative interference. Our data show a benefit of sleep for declarative memory and suggest that sleep actively strengthens declarative memories, which it renders resistant to interference. We showed that cued recall of paired

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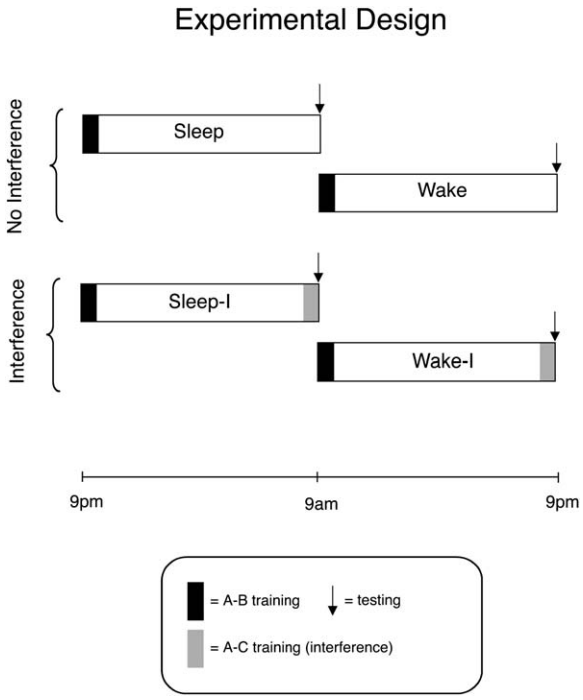

Figure 1. Experimental Design

All 48 participants learned a list of paired associates (A-B). After a 12 hr retention interval that included either sleep or wakefulness, half of the participants were tested on A-B pairs (Sleep and Wake groups); the remaining participants learned an interfering list (A-C) 12 min prior to testing (Sleep-I and Wake-I groups). After completion of these four groups, an additional 24 hr group was run with the interference manipulation (so-called 24-hr-PM-I group, not shown in figure).

Table 1. Mean Percent Recall of the First List and Second List in the Entire Sample

		Recall (SD)		p[a]	Cohen's *d*
Condition	n	B	C[b]	B	B
Wake	12	82 (17)	-	0.064	0.92
Sleep	12	94 (7)	-		
Wake-I	12	32 (19)	94 (8)	<.0001	3.07
Sleep-I	12	76 (17)	96 (6)		

Recall of the first list was recall of B of the A-B pair. Recall of the second list was recall of C of the A-C pair.

[a] Statistical analyses for recall performance were conducted on data after arcsine transformation of all measures, formula = [arcsine(sqrt(accuracy proportion))] [10].

[b] Participants in these no-interference conditions did not undergo associative-interference testing.

associates after 12 hr retention intervals containing sleep or wakefulness yields small differences in recall (13% relative reduction in performance in the Wake group compared to Sleep), a trend consistent with previous studies (e.g., [11]), whereas introducing associative interference after the delay demonstrated robust, large differences (58% reduction). Thus, although a memory trace may appear only modestly improved after similar intervals of sleep compared to wakefulness, the pronounced, beneficial effects of sleep are unmasked by interference testing; after wakefulness, memories remain highly susceptible to associative interference, whereas memories after sleep are resilient to disruption. We propose that sleep plays an active role in consolidating declarative memories and makes them resistant to interference.

It should be noted that the data from the no-interference sleep group approached ceiling, which might have caused us to underestimate the difference in recall of these groups. In addition, it remains unclear whether this sleep benefit is specific for associative processes (i.e., A-B resistant to A-C interference) or is a more general memory effect (i.e., A-B resistant to C-D). Finally, although time-of-day effects are always a concern in sleep studies, we find it unlikely that they account for the findings of this study. There was no difference, at either time of day, in the amount of training needed for subjects to learn all the word pairs, and the training mechanism itself is designed to account for inter-individual differences by providing more learning trials—for those who require it—to reach an equal level of learning. Results of the 24-hr-PM-I group additionally argue against a time-of-day effect because training and testing took place at the same time of day.

In addition to arguing against time-of-day effects, results of the 24-hr-PM-I group demonstrate that sleep provides a benefit that persists throughout the subsequent waking day. Although we did not attempt to control waking mental activities in the wake groups, this was equally true for all groups. If incidental interference (i.e., nonexperimental interference that occurs as a result of waking mental activity) were to account for differences seen in our study between the 12 hr Wake-I and Sleep-I

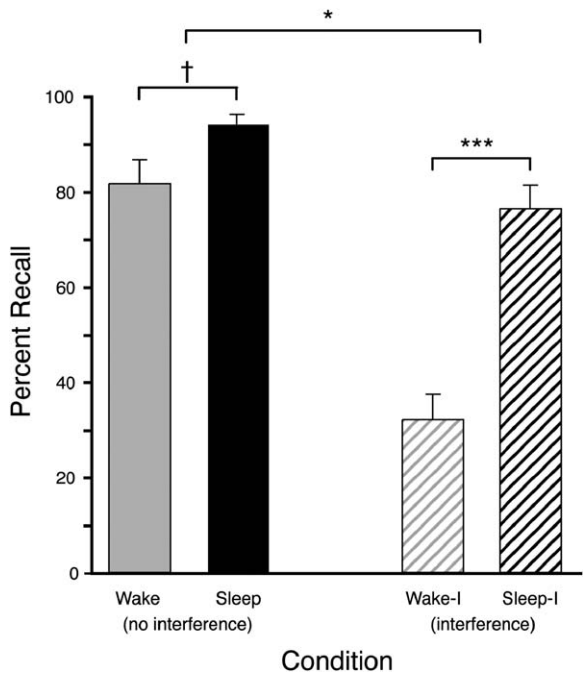

Figure 2. Results

Percent correct recall for B words from the original A-B pair after a 12 hr retention interval is plotted for all four conditions. The bar indicates one standard error of the mean. † = $0.05 \leq p \leq 0.10$; * = $p < 0.05$; *** = $p < 0.001$

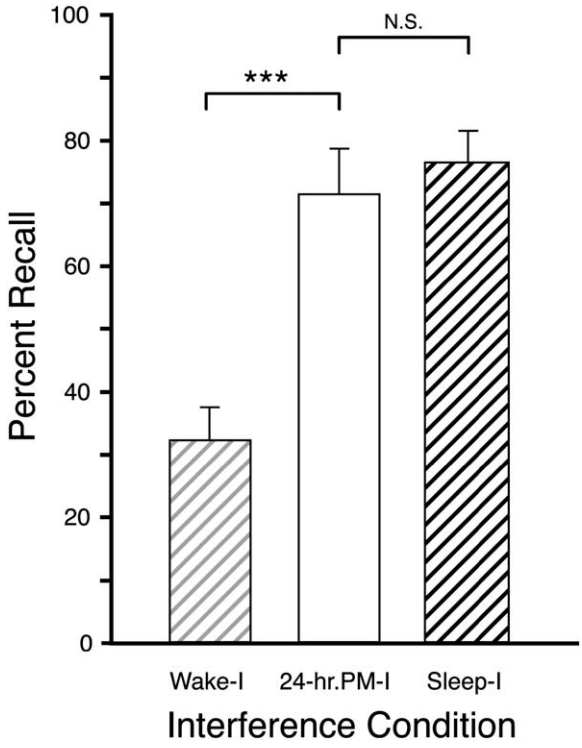

Figure 3. Results of Interference Groups, Including 24-hr-PM-I

Percent correct recall for B words from the original A-B pair is plotted for the 24 hr, PM (i.e., 9 p.m. training and 9 p.m. testing) with interference group (24-hr-PM-I) compared to the 12-hr sleep with interference group (Sleep-I) and 12-hr wakefulness with interference group (Wake-I). The bar indicates one standard error of the mean. (Please note that the Sleep-I and Wake-I groups presented in this figure are the same groups as the Sleep-I and Wake-I groups presented in Figure 2.) *** = $p < 0.001$; N.S.= not significant.

groups, then the 24-hr-PM-I group should perform at least as poorly as the Wake-I because members of this group had more than 12 hr of wakefulness; however, they performed considerably better. These data collectively indicate that the benefit that sleep provides declarative memory is not a passive (i.e., transient) protection against interference while an individual is asleep (a perspective inherited from the work of Jenkins and Dallenbach [12]). Rather, sleep actively stabilizes memories such that they become resistant to interference in the subsequent day.

This study clarifies and extends prior work attempting to understand the role of sleep in declarative-memory consolidation. Many studies have emphasized individual sleep stages, sleep deprivation, or both in their experimental paradigms (for a review of traditional study designs, and exemplary studies, see [13]). Two recent studies extended the work of Barrett et al. [14] and emphasized the role of slow wave sleep (SWS) in declarative-memory consolidation [15, 16]. They showed enhanced performance across the first half of the night (so-called "early sleep," a portion of sleep with relatively large amounts of SWS) compared to a matched period of sleep deprivation. We have extended these findings by examining sleep across an entire night and comparing them to wakefulness across the daytime (i.e., without acute sleep deprivation). Ekstrand [11] demonstrated that cued recall receives a similar benefit of sleep as our no-interference groups. We demonstrated a much larger effect size, and more significant effect of sleep, when participants were tested after associative interference. By tapping a particular sleep-provided benefit—rendering memories resistant to interference—we were able to show a robust benefit of sleep across the entire night and even across 24 hr.

Other studies have examined A-B/A-C learning paradigms and sleep. Norman et al. applied a computational, neural network (the complementary-learning-system model [17, 18]) to study the effects of REM sleep on semantic learning in neocortical architecture [19]. This computational model demonstrates that semantic knowledge, represented by extensive A-B training, is "repaired" by REM sleep epochs that alternate with A-C learning. Ekstrand [11] demonstrated that disruption of A-B learning by immediate A-C learning—all *before* sleep—can be recovered by subsequent sleep (although in a subsequent Ekstrand concluded that the interference effects he observed were not due to sleep [20]). Our findings extend these studies by demonstrating that sleep leads to the protection of episodic memories from subsequent interference; when we sleep after learning new, episodic information, the memories become resistant to disruption by subsequent learning.

Evidence from animal models and human neuroimaging studies predicts the active participation of sleep in hippocampus-mediated memory consolidation. Several animal studies demonstrate that recently acquired, hippocampus-based memories are "replayed" during sleep (e.g.,[6–8]) and that this reverberation is coherent with associated neocortex (e.g., [21, 22]). Recent neuroimaging findings in humans further demonstrate increased hippocampal activity during sleep after spatial learning [9]. Collectively, these studies suggest that hippocampus-dependent memories are repeatedly reactivated during sleep and that coherent networks form within and between appropriate brain regions. This sleep-dependent, reiterative process orchestrates the strengthening of memories and thereby renders them less vulnerable to interference. It is plausible that such biological mechanisms underlie the critical role we observed for sleep in declarative-memory consolidation.

Conclusions

This is the first study to demonstrate that sleep protects declarative memories from associative interference in the subsequent day, and it thereby provides key evidence that sleep does not passively (i.e., transiently) protect declarative memories; rather, sleep plays an active role in declarative-memory consolidation. Our study provides a source of convergence among human behavior, animal research, computational models, and neuroimaging studies for investigations of declarative-memory consolidation. Although further research is needed to define the empirical limits and physiological correlates of this sleep and memory interaction, our study provides a new framework for considering the effects of sleep on human memory: sleep helps consolidate declarative memories and renders them resistant to associative interference.

Experimental Procedures

We sought to challenge the assumption that sleep reduces forgetting merely by passively protecting encoded memories. To achieve this goal, we used the classic A-B, A-C paradigm from Barnes and Underwood [23]. (For a discussion of many studies using this manipulation, see reference [24].) In our adaptation, we experimentally introduced interference after off-line retention periods that contained sleep or wakefulness. Our hypothesis was that if consolidated memories are resistant to interference, and if sleep plays an active role in declarative-memory consolidation, then those memories would be resistant to interference after sleep but susceptible to interference after wakefulness.

Participants

All potential participants completed a screening questionnaire and interview prior to selection. Individuals taking prescription, psychoactive medication or illicit drugs were excluded prior to randomization. All participants were native English speakers. We excluded those with known sleep disorders or abnormal sleep patterns: habitual sleep onset after 2 a.m.; sleep duration less then 6 hr; or pathologic sleepiness (defined by an Epworth Sleepiness Scale score > 10). Sixty participants (ages 18–39; 33 women) were enrolled and successfully completed the study.

Materials

Words were drawn from the 478 nouns in the Toronto Word Pool [25]. We selected a random subset of two-syllable nouns and created a list of 60 words that were matched for imageability, frequency, and concreteness. Words were randomly divided into three groups of 20, forming three lists: A, B, and C. Item assignments to list B or C were counterbalanced across participants. Each word in the A list was paired with one word each from the B and C lists, thereby creating two lists of paired associates: A-B and A-C (e.g., BLANKET-VILLAGE and BLANKET-RUBBER). Word pairs with obvious semantic relationships were re-randomized.

Procedures

Forty-eight participants were randomly assigned to one of four groups: Sleep; Wake; Sleep-I (sleep with associative interference); and Wake-I (wake with associative interference). All groups learned 20 paired associates in two phases: study-only and anticipation-plus-study. Twelve hours later, they were tested for recall. Participants in the interference conditions learned a second word-pair list prior to this testing. Training and testing were administered on a computer with E-prime software (Psychology Software Tools Inc.) in a quiet testing room.

In the study-only phase, paired associates were presented in black and were centered on a white screen, in capital letters. Word pairs were presented sequentially, in a fixed order across subjects and groups, for 7 s each. Immediately afterward, in the second phase of learning, the list was repeatedly shown one pair at a time in the same order, but this time with an anticipation-plus-study procedure similar to that used by Bower, Thompson-Schill, and Tulving [26]. Participants were presented with the first word of each pair and were required to type the second word. The computer provided immediate feedback by displaying the correct pairing for two seconds (e.g., "Correct. The correct pairing is:" or "Incorrect. The correct pairing is:"). After any individual pair was correctly recalled three times, it was removed from the list. Study continued until all word pairs were removed; the learning criterion was thus set to 100% for all participants.

After completing this two-phase learning, participants left the laboratory for 12 hr. Participants in the sleep groups trained at 9 p.m. and returned at 9 a.m. for testing, whereas participants in the Wake groups trained at 9 a.m. and returned at 9 p.m. for testing (Figure 1). Self-reports from the participants in the two sleep groups demonstrated similar post-training quantities of sleep (Sleep: M = 7.16 hr, SD = 0.60; Sleep-I: M = 7.22, SD = 0.67). Participants in the Wake groups were not restricted from any activity, other then napping between the training and testing phases of the experiment.

Upon returning to the laboratory, participants in the no-interference conditions were immediately tested. They were provided a piece of paper containing all 20 A-list stimuli (the A words of the A-B pairs), followed by a blank space, and were instructed to write the word that completed the pair (B of A-B). They had 6 min to complete this cued-recall task. Participants in the interference conditions learned a new list (A-C) prior to testing; the same training procedure was used as with the original (A-B) list. After learning this new list, these participants performed a 12 min finger-tapping task in order to prevent rehearsal during a brief delay between training and testing. After this motor task, participants were asked to recall the paired words from both lists, B and C of A-B and A-C. Although our outcome of interest was recall accuracy of the B list, participants were allowed to record the C words during recall testing. This established manipulation [23] provided the benefit that if both B and C words for any given A cue were remembered, these responses would not be required to compete for the single response slot; rather, subjects could provide both answers. If subjects only remembered one of the two completions, they were to leave the other column blank. If they recalled a completion word but did not remember the source list, they were instructed to write the word in either column and mark it with an asterisk. Morphological errors (e.g., "fathers" instead of "father") were counted as correct. Only those words that were recalled and identified with the correct cue word (A) and placed in the correct list (B column if learned before the delay or C column if learned after the delay) were counted as accurate.

After completion of these four groups (n = 48), an additional group of 12 participants was run in a 24 hr paradigm. These participants underwent the same screening and experimental procedures as the interference groups, except training on A-B was done at 9 p.m. and training on A-C as well as testing was done at 9 p.m. the following day (the so-called 24-hr-PM-I group).

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