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New evidence for the necessity of a silent plastic period during sleep for a memory benefit of targeted memory reactivation

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The presentation of learning-related cues during sleep, a procedure known as targeted memory reactivation (TMR), is a promising approach to bias sleep-dependent memory consolidation. Recent evidence suggests that for the TMR procedure to be effective, an undisturbed short plastic period is needed after cueing. In this study, we investigated the impact on memory consolidation of verbatim or interfering auditory presentation of previously learned material during wakefulness or non-rapid eye movement (NREM) sleep. Participants learned a list of 40 word pairs (A–B) presented both visually and aurally. During a 90-min consolidation interval spent awake or asleep (diurnal nap), the first word of half of the pairs was aurally presented, then followed by its correct (A–B) or incorrect (A–C) association. Memory for all pairs was then tested. In the Wake conditions, memory was improved after correct (verbatim) cueing but impaired after the presentation of an incorrect (interference) association, in comparison with uncued word pairs. In the Nap conditions, the TMR procedure had no beneficial or detrimental effects on memory consolidation. Time–frequency analyses indicate that the presentation of the first word during NREM sleep is followed by increased spindle-related sigma activity, suggesting that reactivation of the associated memory content was initiated. However, immediate presentation of the second word, either correct or incorrect, annihilated sigma activity. These results bring additional evidence on the necessity for a silent and undisturbed plastic period during sleep after cueing in a TMR procedure to achieve successful reactivation and the ensuing gain in memory consolidation.

Keywords: declarative memory; sleep; targeted memory reactivation; spindles; active system consolidation; plastic period

HIGHLIGHTS

- Verbatim auditory cueing during sleep does not boost memory consolidation
- Cueing during a wake period allows biasing memories
- The increase in sigma power following sleep-cueing is annihilated by a following cue
- A silent undisturbed period during cueing is crucial for the memory benefits

INTRODUCTION

Compelling evidence fosters the idea that sleep is an active player in the offline processes of memory consolidation. For instance, learning-related neural activity is re-expressed during sleep and associated with overnight memory improvement (Axmacher, Elger, and Fell, 2008; Dupret, O'Neill, Pleydell-Bouverie, and Csicsvari, 2010; Peigneux et al., 2004), in agreement with the hippocampo-cortical dialogue model (Buzsaki, 1989). This model proposes that during non-rapid eye movement (NREM) sleep, hippocampus-dependent novel and fragile memory traces are spontaneously reactivated and progressively transferred toward neocortical regions for enduring storage. The transfer mechanism would be mediated by coordinated activity between hippocampal sharp wave ripples, thalamo-cortical sleep spindles, and slow oscillations (Sirota and Buzsáki, 2005; Marshall and Born, 2007; Born and Wilhelm, 2012).

In the last decade, novel paradigms have been proposed to further promote on-going memory consolidation processes during sleep using learning-related stimulations, a procedure known as targeted memory reactivation (TMR). During NREM sleep, providing olfactory or auditory reminders associated with the newly learned material selectively enhances memory consolidation (for reviews, see Oudiette and Paller, 2013; Farthouat and Peigneux, 2015; Schouten, Pereira, Tops, and Louzada, 2016; Schreiner and Rasch, 2016). Diffusing an odour previously presented as a background during the learning phase was found to improve post-sleep memory accuracy for locations of objects (Rasch, Buchel, Gais, and Born, 2007; Diekelmann, Büchel, Born, and Rasch, 2011; Rihm and Rasch, 2014). Similarly, delivering learning-related sounds during NREM sleep eventually resulted in a better recall for the location of objects

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(Rudoy, Voss, Westerberg, and Paller, 2009). This latter finding indicates that TMR-related effects do not merely reflect a global improvement, but rather are specific to the cued material. TMR was also found beneficial for vocabulary learning (Schreiner and Rasch, 2015a). Participants learned German translations of unknown foreign vocabulary (i.e., Dutch words). Although re-exposure to the Dutch words was not beneficial when cues were presented during post-learning wakefulness (Schreiner and Rasch, 2015b), the performance significantly improved when the cues were delivered during NREM sleep (Schreiner and Rasch, 2015a). Furthermore, replay of memory-related cues during sleep activates the hippocampal formation (Rasch et al., 2007; Diekelmann et al., 2011; van Dongen et al., 2012) and auditory replay of task-related cues biased (re)activation in hippocampal neurons in rodents (Bendor and Wilson, 2012). In addition, consistent evidence suggests that NREM sleep slow waves and spindles frequency-related oscillations play an important role in the processing of cued memories (for a review, see Schreiner and Rasch, 2016). Altogether, these data suggest that TMR effectively promotes memory consolidation for declarative memory material by reactivating and stabilizing hippocampal memory traces (Diekelmann et al., 2011), a process putatively mediated by sleep slow oscillations and spindles according to the active system consolidation theory (Born and Wilhelm, 2012). It was proposed that reactivation-induced plasticity might actually represent the accelerated counterpart of the spontaneous memory reactivation that takes place in the hippocampo-cortical dialogue (Diekelmann et al., 2011), thus providing support to the hypothesis of a causal role between memory reactivation and overnight consolidation processes.

Consequently, cueing memories during sleep is a procedure that gained popularity over the last decade and could possibly be useful for clinical applications. For instance, a cueing procedure was used to extinguish fear memory (He et al., 2015; Rihm and Rasch, 2014; Rolls et al., 2013; Oudiette, Antony, and Paller, 2014), with interesting outcomes for the management of post-traumatic stress disorders. However, Rihm, Sollberger, Soravia, and Rasch (2016) failed to demonstrate a beneficial impact of cueing on therapy outcomes, while cueing in rodent is likely to strengthen the associated fear memories (Barnes and Wilson, 2014). Nonetheless, because the exact mechanisms underlying TMR and memory consolidation are not well known, TMR should be carefully manipulated and there is a need to draw the limits and conditions for successful memory reactivation and stabilization. For instance, most studies focused on cueing material with associated sounds/odours, but actually did not investigate the impact of cueing *verbatim*, the learned material. In the framework of the reconsolidation theory (Nader, 2009), however, one could expect that reactivating the whole learned memory content would even reinforce better the memory trace. In a variant of their initial study (i.e., in which participants learned a new foreign vocabulary; Schreiner and Rasch, 2015a), Schreiner, Lehmann, and Rasch (2015) found a beneficial effect of cueing during sleep when the auditory cue (i.e., the Dutch word) was followed 1,500 ms later by its correct feedback (i.e., the German translation), a benefit associated with changes in theta and sigma frequency power. It is worth noticing here that the memory benefit using both cue and feedback presentations was not superior to a cue presentation alone condition. Nonetheless, the TMR memory benefits and the associated neurophysiological responses were abolished when the feedback rapidly followed the cue (i.e., 200 ms after). To account for this negative finding, the authors proposed that an undisturbed plasticity period of duration of at least 1 s following the presentation of the cue is of crucial importance to achieve successful memory reactivation and to allow the development of the associated neurophysiological responses.

In this study, we further investigated the limits of sleep TMR in a declarative memory task in which participants learned pairs of semantically unrelated words. After learning, they were aurally delivered the two words constituting each pair either during post-training NREM sleep (diurnal nap; Nap conditions) or during wakefulness (Wake conditions). The interval between the first (cue) and second (associate) word of the pair varied between 1,000 and 1,500 ms, an interval supposedly sufficient according to Schreiner et al. (2015) and Schreiner and Rasch (2016). In addition, we investigated the specificity of TMR by displaying an incorrect feedback after the cue, i.e., by creating an interfering association with the learned cue. Indeed, cueing during sleep is thought to stabilize memories and to protect them from future interferences, while cueing during wakefulness has the opposite effect (Diekelmann et al., 2011). Schreiner et al. (2015) previously investigated the presentation of a false feedback in a short interstimulus interval (ISI, 200 ms) condition, i.e., in a condition in which the benefits of memory reactivation were abolished. This effect remains to be tested at longer ISI in which benefits of TMR were evidenced, since nowadays, the consequences of directly presenting interfering associations during sleep remain uncertain.

METHODS

Participants

Ninety-six healthy young adults participated in this study were approved by the Faculty Ethics Committee of the Université libre de Bruxelles. Participants in the Wake conditions ($N = 41$) participated in exchange for course credits. Participants in the sleep (Nap) conditions ($N = 55$) received monetary compensation. All participants were native French speakers, free of medication known to influence sleep quality and/or mood, and reported not having suffered from neurological or psychiatric disorders. Both in the Wake and Nap conditions, participants were randomly assigned to the Reactivation or Interference group. In the Nap conditions, 26 out of the 55 participants slept enough during the nap to allow at least one auditory replay of each to-be-cued word pairs (see below). The other participants ($N = 29$) were *a posteriori* assigned to the No-reactivation group. In the Wake conditions, 13 participants were excluded from the analyses because they did not reach the performance threshold (immediate recall score was at least 60%) in the pre-TMR recall session. One participant was excluded in the Nap conditions for the same reason. The following descriptions and analyses are restricted to the valid participants in the Nap-Reactivation ($N = 14$), Nap-Interference

($N = 11$), Nap-No-reactivation ($N = 29$), Wake-Reactivation ($N = 14$), and Wake-Interference ($N = 14$) groups.

Participants in the Nap conditions (18 males; mean age ± standard deviation: 22.4 ± 3.1 years) had neutral to moderate evening or morning chronotype (mean Morningness–Eveningness Questionnaire score: 48.2 ± 7.5, range: 33–63; Horne and Ostberg, 1976) and vocabulary levels within normative values (mean Mill-Hill Vocabulary Scale score: 33.4/44 ± 3.8; Deltour, 1993). They were asked to maintain a regular sleep–wake cycle during the entire experiment. Compliance was verified by means of a sleep agenda to be completed each morning for the 2 days before the learning session.

Participants in the Wake conditions (9 males; mean age ± standard deviation: 20.21 ± 2.33 years) had neutral to moderate evening or morning chronotype (mean Morningness–Eveningness Questionnaire score: 46.96 ± 7.5, range: 31–59; Horne and Ostberg, 1976) and vocabulary levels within normative values (mean Mill-Hill Vocabulary Scale score: 31.30/44 ± 5.9; Deltour, 1993). Participants in the Nap and Wake conditions did not differ in chronotype or in vocabulary levels (Mann–Whitney U tests, $p > .1$).

Materials

One hundred and twenty French bi-syllabic and emotionally neutral words were selected using the BRULEX database (Content, Mousty, and Radeau, 1990) and assigned to lists A, B, and C. A–B pairs constituted the word pairs to be learned and A–C pairs constituted the interfering list (word lists are available in the supplementary material). The words constituting a pair never started nor ended with the same phoneme and were semantically unrelated (according to the three authors). A male experimenter aurally recorded each word (sampling frequency: 44,000 Hz, duration range 382–883 ms, and average and standard deviation: 585 ± 120 ms) using a stereo headset (Logitech 981-000350 H150). The average duration of the words was similar between lists A, B, and C [one-way ANOVA with between factor Lists (A vs. B vs. C), $F(2, 117) = 1.93$, $p > .1$]. Surrounding noise was removed using the Praat software (Boersma, 2002).

Procedure

The experimental setting for the Nap conditions is summarized in *Fig. 1*. On the experimental day, participants arrived at the sleep laboratory at 12:15 to be prepared for polysomnographic (PSG) recording (see below). Thirty minutes later, vigilance and sleepiness were assessed using the 10-min version of the Psychomotor Vigilance Task (PVT; Dinges and Powell, 1985) and the Karolinska Sleepiness Scale (KSS; Åkerstedt and Gillberg, 1990), respectively. In the Wake conditions, participants were tested during the day (between 9:00 and 18:00). Sleepiness was assessed using the KSS (Åkerstedt and Gillberg, 1990). Word presentation during the learning and recall sessions and during sleep or wakefulness TMR was performed using the Psychtoolbox-3 software (Brainard, 1997) implemented in Matlab 2011 (MathWorks Inc., Natick, MA, USA).

Word-pair learning and pre-TMR recall test. The word-pair learning session was identical for the Wake and Nap conditions (see *Fig. 1*). Participants were informed that a list of 40 word pairs will be visually and aurally presented, and that they must memorize all word pairs for later recall. The 40 word pairs were displayed one by one simultaneously visually on a computer screen for 3,000 ms and aurally through loudspeakers. A fixation cross was displayed in the center of the screen for 2,000 ms before the appearance of the following pair. The list of word pairs was presented

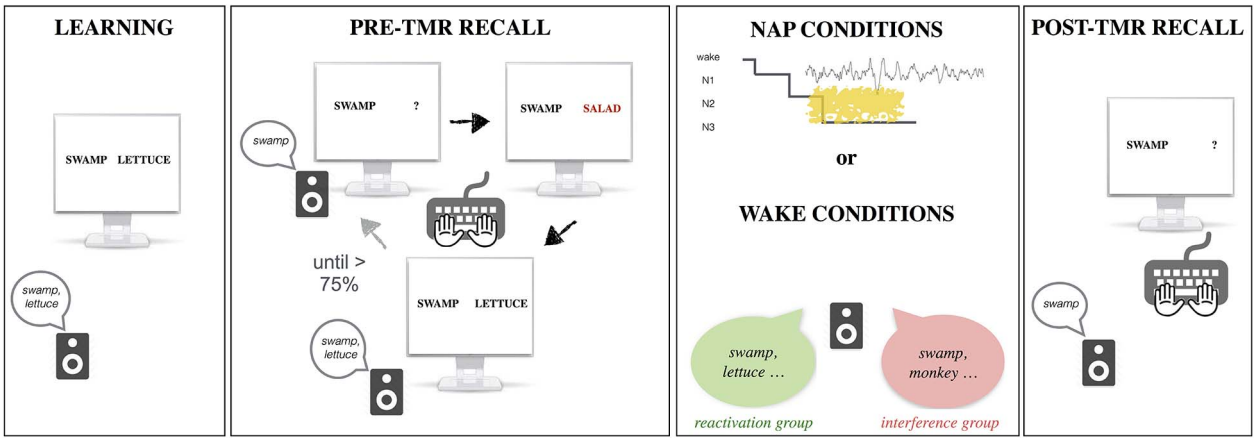

Fig. 1. Experimental protocol. Learning session and pre-TMR recall: At learning, 40 word pairs (A–B) are presented twice simultaneously both in visual and oral modalities. Pre-TMR recall: During the pre-TMR cued recall test, the first word of each pair is presented (both visually and aurally). Participants have to type the associated word on a keyboard, then receive a feedback displaying the correct response to favor error-free consolidation. If the recall score is <75% accuracy, incorrectly learned word pairs are presented again. TMR session: In the Nap conditions, half of the word pairs are aurally displayed during NREM sleep. In the Wake conditions, word pairs are replayed during passive, quiet wakefulness. Word pairs are either identical (A–B, Reactivation group) or interfering (A–C, Interference group). Post-TMR recall: After the memory reactivation procedure, a delayed cued recall test is administered. The test is identical to the pre-TMR recall test, except that no feedback is provided about the correctness of the response and no performance criteria is set

twice in randomized order. The encoding session lasted approximately 10 min.

After encoding, participants were guided to an immediate cued recall test. The first word of each pair was presented both visually on the screen and aurally through the loudspeakers, followed by a question mark on the screen. Participants had then to type the associated word on a keyboard. Whatever the correctness of the participant's answer, a visual feedback was given and the correct response was presented both visually (in green font color for correct responses and in red for incorrect responses) and aurally to favor error-free consolidation in memory. If the recall score was below the 75% of correct responses threshold, incorrectly recalled word pairs were presented again for learning and a new cued recall test was administered with only the incorrectly learned word pairs, to avoid over-consolidation effects. The presentation of the word pairs was randomized at each presentation to prevent serial learning. Each word pair was presented five times maximum, after what performance was calculated even if below the 75% of correct responses criteria.

Targeted memory reactivation (TMR)

Nap conditions. After the learning and immediate cued recall sessions (13:30–13:45), participants in the Nap conditions were allowed a 90-min napping opportunity. The nap took place in an isolated, quiet room. Participants were instructed that a background brown noise would be continuously displayed at low intensity (~30 dB) through loudspeakers during the nap. The brown noise was used to habituate participants sleeping in a noisy environment and decrease the probability of arousals triggered by the presentation of the words during sleep. For each participant, sound intensity was individually adjusted to ensure that it was not disturbing, and 20 out of the 40 word pairs were randomly selected for TMR stimulation. Participants were not informed that the learned material would be replayed during their sleep. According to the experimental group, word pairs selected for stimulation were either identical to the learning set (list A–B, Reactivation group) or interfering (list A–C, Interference group). The stimulation procedure was identical for both groups.

Throughout the nap, PSG (EEG, EOG, and EMG) was checked online. Auditory stimulations were only delivered after a minimum of 10 consecutive 30-s epochs (5 min) of stable and unequivocal N2 sleep. During the stimulation period, the selected list of word pairs was replayed aurally several times, as much as the sleep conditions were met (see below). The interval between the onsets of the first and second words of a pair was fixed to 1,883 ms (i.e., 1,000 ms + the duration of the audio file set to the longest word duration 883 ms). However, since words in the audio files had an actual duration ranging 383–883 ms (see Materials section), it results in variable ISIs (i.e., the interval between the end of the presentation of the first word of the pair and the onset of the second one), actually ranging 1,000–1,500 ms. Each pair was separated from another by a 5,000-ms interval. The brown noise was continuously displayed both during the actual presentation of the words and during the intervals between words or pairs. Sound intensity monitored using a digital sound level meter (Wensn®) varied from one participant to another, and was adjusted online according to the depth of sleep and the electrophysiological responses to the auditory stimulations. A trigger was added to the PSG recording at the onset of each pair of words to allow later analyses on cues-related electrophysiological responses. Word pairs were continuously delivered aurally as long as no (micro-) arousal or beginning of a rapid eye movement (REM) sleep episode was detected. In the case of such an event, the experimenter directly stopped the auditory stimulation and the background brown noise alone continued to be displayed. The presentation of the word pairs was then resumed after a minimum of 10 consecutive epochs of N2 sleep. At the end of the nap, participants were awakened and asked to report on their sleep quality during the nap. They were also asked if they heard any specific sounds during their nap. At the end of the nap, if participants were delivered less than one auditory replay of each to-be-cued word pairs (<20 word pairs) due to sleep conditions, they were arbitrarily assigned to the No-reactivation group.

Wake conditions. For the TMR procedure, participants in the Wake conditions were asked to sit quietly in the experimental room (no PSG recording). Word pairs were delivered via headphones. Participants were asked to pay attention to the pairs but not explicitly asked to memorize the word pairs again. The TMR procedure was identical to the Nap conditions, except that each of the 20 word pairs was replayed four times. The TMR procedure lasted around 15 min.

Post-TMR recall. In the Nap conditions, the PSG montage was removed after final awakening, and participants were allowed to wash their hair. This time period allowed them to fully wake up and dissipate sleep inertia (Hofer-Tinguely et al., 2005). Vigilance and sleepiness were assessed again using the 10-min PVT and KSS, respectively.

In the Nap and Wake conditions, a cued recall test was administered to assess memory both for cued and uncued word pairs. The task was identical to the pre-TMR recall test, except that no feedback was provided about the correctness of the responses and no performance criteria was set.

Polysomnography (PSG)

PSG (BioSemi B.V., Amsterdam) included five electroencephalographic (EEG) channels (FP1, FP2, C3, C4, and O1) referenced to the averaged mastoids (M1 and M2) and two bipolar electrooculograms (EOGs) and chin electromyograms (EMGs), positioned according to the 10–20 electrodes placement system (Jasper, 1958). Silver/silver-chloride (Ag/AgCl) electrodes were fitted with collodion, and impedance was kept below 10 kΩ. EEG data were recorded at a sampling frequency of 2,048 Hz. High-pass and low-pass frequency filters were set at 0.30 and 45 Hz for EEG and EOG recordings, respectively. EMG high-pass and low-pass frequency filters were set at 10 and 100 Hz, respectively. Sleep staging was performed *a posteriori* based on 30-s epochs differentiating Wakefulness, NREM stage 1 (N1), N2, N3, or REM sleep stage according to the AASM criteria (Iber, Ancoi-Israel, Chesson, and Quan, 2007) using the FASST toolbox (http://www.montefiore.ulg.ac.be/~phillips/FASST.html) implemented in Matlab 2011 (MathWorks Inc., Natick, MA, USA).

Data analysis

Behavioral analyses. Performance was computed as the proportion of correctly recalled word pairs from pre-TMR to post-TMR sessions. A retention score was calculated as the ratio between performances at the pre-TMR session relative to post-TMR recall, expressed in percentage (i.e., 100 × post-/pre-TMR recall performance). Because the experimental conditions were slightly different between conditions, analyses were conducted separately in the Wake and Nap conditions. For the Wake conditions, a 2 × 2 repeated measure ANOVA was computed with between factor Group (Reactivation vs. Interference) and within factor TMR (Cued vs. Uncued). For the Nap conditions, a 3 × 2 repeated measure ANOVA was computed with between factor Group (Reactivation vs. Interference vs. No-reactivation) and within factor TMR (Cued vs. Uncued).

Based on the assumption that the benefits of reactivation may be presented only at a long, 1,500 ms ISI (Schreiner et al., 2015; Schreiner and Rasch, 2016), and considering the fact that ISIs actually ranged from 1,000 to 1,500 ms in the present experiment, we performed post-hoc analyses to investigate in detail the impact of ISI duration on the TMR effect in the Nap conditions. To do so, word pairs were separated into three sets of items of similar size, based on the distribution of ISIs (see *Fig.* 2; as a reminder, ISI here is the time elapsed between the end of the presentation of the first word of the pair and the onset of the second word). These three sets of word pairs were defined as short ISI (n = 13 word pairs; 1,000–1,290 ms), medium ISI (n = 13 word pairs; 1,290–1,373 ms), and long ISI (14 word pairs; 1,373–1,500 ms). Separate 2 × 2 repeated measures ANOVA were computed for the short, medium, and long ISI sets on retention scores, with between-subject factor Condition (Reactivation vs. Interference) and within-subject factor TMR (Cued vs. Uncued). Of note, data inspection shows that, respectively, 6.04 ± 1.43, 7.16 ± 1.54, and 6.80 ± 1.35 (mean ± standard deviation) short, medium, and long ISI word pairs were presented during sleep, whereas respectively 6.96 ± 1.43, 5.84 ± 1.54, and 7.20 ± 1.35 short, medium, and long ISI word pairs were not presented during subsequent sleep.

Electrophysiological analyses

PSG analysis. Stages (Wake, N1, N2, N3, and REM) duration, total sleep time, and recording duration were calculated and averaged for each group. To assess sleep differences between conditions, separate one-way ANOVAs were performed on averaged stages duration with between factor Group (No-reactivation vs. Reactivation vs. Interference) separately for each stage.

Spectral power analyses. Spectral power analyses were computed to investigate auditory stimulation-related effects on neurophysiological activity during sleep and the electrophysiological correlates of cueing-dependent memory benefits, using the FieldTrip toolbox (Oostenveld, Fries, Maris, and Schoffelen, 2011).

For time–frequency analyses, data were segmented from 1,500 ms before the onset of the first word of the pair until 4,500 ms after. Segments were then detrended (to remove slow low-frequency drift) and demeaned (i.e., we subtracted the overall mean amplitude to avoid spectral leakage), and selected trials visually inspected to exclude segments containing muscle or eye movement-related activity or technical artifacts (data were band-pass filtered for visualization purposes in the frequency band 0.1–20 Hz). Morlet wavelet analyses (Morlet parameter = 7, i.e., the number of cycles per time window, fixed for all frequencies) were computed on trials with a 50-ms sliding time window. Time–frequency analyses were computed in the 4–22 Hz range (analyses in the delta band <4 Hz were not possible considering the too short 1-s time window). Because of edge effects, 500 ms was discarded at the beginning and end of each segment. For the purpose of visualization, data are plotted with respect to the absolute baseline [−1,000, 0] ms. Please note that similar results were obtained when used a [−0.3, −0.1] ms baseline as done in a previous study (Schreiner et al., 2015). For statistical analyses (computed on data without baseline correction), we defined two sets of windows of interest. The first window of interest featured the 1-s period preceding the onset of the first word of the pair (defined as the baseline), the 1-s during the presentation of the first word of the pair, and the 1-s during the presentation of the second word of the pair. The second window of interest featured the

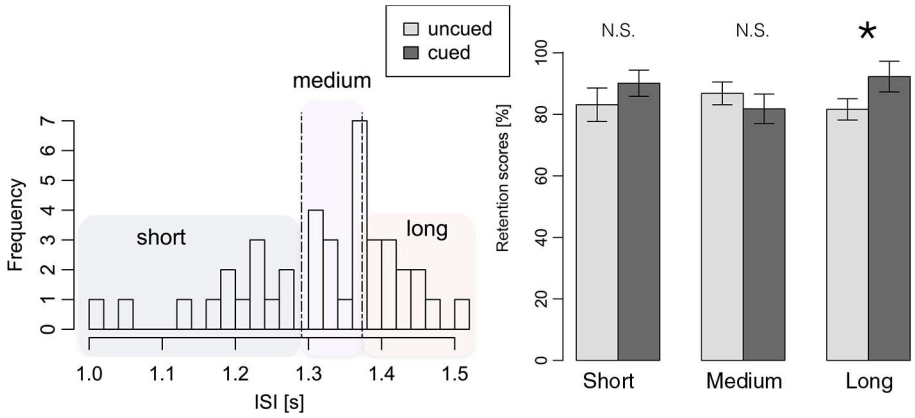

Fig. 2. ISI-related TMR effects on memory retention. (Left) Interstimulus interval (ISI) distribution, ranging 1,000–1,500 ms and divided into three sets of items: short, medium, and long ISI. (Right) Retention scores in the Nap conditions. Retention scores are higher for cued (light gray) than uncued (dark gray) word pairs in the long ISI (1,373 < ISI< 1,500 ms) set only, similar in the Reactivation and Interference groups. Bar plots are mean retention scores and error bars are the standard error. *p < .05, N.S: non-significant

1-s period preceding the onset of the first word of the pair, i.e., the baseline, the 1-s following the presentation of the first word of the pair, and the 1-s following the presentation of the second word of the pair. Based on prior findings (Schreiner and Rasch, 2015a; Schreiner et al., 2015), we also defined two power bands of interest in the theta (5–7.9 Hz) and sigma (11–16 Hz) frequency range. Theta power and sigma power were averaged across the time windows of interest and across frequencies within the respective frequency bands. Finally, power values were averaged between frontal (FP1 and FP2) and central (C3–C4) electrodes separately. Data were log-transformed because they were not normally distributed (Shapiro–Wilk test, $p < .05$). To evaluate the effect of cueing on EEG power for the two sets of windows of interest, spectral power analyses were conducted using the repeated measure ANOVA with within-subject factors Location (central vs. frontal electrodes) and Window (baseline vs. first word vs. second word) separately in the theta and sigma bands.

In addition, global delta power (0.5–4 Hz) and sigma power (11–16 Hz) were calculated over N3 and N2 sleep stages, respectively, using 5-s artifact-free sliding windows (50% overlapping). Power was computed at the two frontal electrodes using a Hanning taper Fast Fourier Transform in the delta frequency range (frequency resolution: 0.2 Hz). Power was averaged between electrodes FP1–FP2 and C3–C4. Based on previous findings (Creery, Oudiette, Antony, and Paller, 2015), analyses were performed on frontal electrodes for delta power and on central electrodes for sigma power.

Additional analyses on event-related potentials evoked by word pairs are available in the supplementary material.

Statistical analyses. All statistical analyses are performed using R (R Core Team, 2014) or JASP (JASP Team, 2016). All correlations are Pearson's correlation (unless otherwise indicated). Tukey post-hoc analyses were conducted when appropriate. All tests were two-tailed ($\alpha = 0.05$). To assess differences between slopes, the ZPF statistic (Raghunathan, Rosenthal, and Rubin, 1996) was calculated in the situation, where there is a variable in common between two correlations (e.g., delta power correlations with performance for both uncued and cued word pairs), using the excel worksheet calculator available at http://www.prostatservices.com/?p=73&option=com_wordpress&Itemid=56.

RESULTS

Sleep parameters derived from PSG recordings are reported in *Table 1*. In the Nap conditions, the average number of auditory stimulations was, by definition, larger for the Reactivation (220.5 ± 117.8) and Interference (235.8 ± 132.5) groups than for the No-reactivation group (4.7 ± 8.3), but did not differ between the Reactivation and Interference groups ($t = 0.5$, $p > .1$). As a reminder, the number of auditory stimulations was fixed by design to 80 (4 × 20 word pairs) in the Wake conditions. Sleep stage durations were the same for the Reactivation and Interference groups, except for the REM duration which was longer in the Reactivation group ($t = -2.9$, $p < .05$; see *Table 1*).

Sleepiness and vigilance in the Nap conditions

No significant effects (all $p > .15$) were disclosed in the repeated measure ANOVA conducted on subjective sleepiness scores with within-subject factor Moment (pre-KSS1 vs. post-KSS2 TMR testing session) and between-subject factor Group (No-reactivation vs. Reactivation vs. Interference), suggesting similar sleepiness levels before (KSS1 mean score: 3.9 ± 1.7) and immediately after (KSS2 mean score: 3.6 ± 1.3) the nap. Similarly, no significant effects (all $p > .13$) were disclosed in the repeated measure ANOVA conducted on mean reaction time (RT) and reciprocal RT (mean 1/RT; Basner, Mcguire, Goel, Rao, and Dinges, 2015) in the PVT before and after the nap (within-subject factor Moment; between-subject factor Group). Mean RTs were 342 ± 55 ms before the nap and 343 ± 49 ms after the nap. Reciprocal RTs were 3.165 ± 0.309 ms^{-1} before the nap and 3.115 ± 0.382 ms^{-1} after the nap.

TMR-related effects on memory

Wake conditions. In the Wake conditions, participants correctly recalled 81 ± 11% and 70 ± 14% of word pairs at pre- and post-TMR recalls, respectively. The ratio of

Table 1. Stages duration in minutes (mean ± standard deviation)

Duration (min)	No-reactivation	Reactivation	Interference	F	p	No-reactivation vs. Reactivation	No-reactivation vs. Interference	Reactivation vs. Interference
Wake	57.7 ± 27.7	20.9 ± 13.2	25.2 ± 12.4	16.7	<.001	$t = -5.1$, $p < .001$	$t = -4.1$, $p < .001$	$t = 0.5$, $p > .1$
N1	10.0 ± 9.7	18.4 ± 11.6	11.7 ± 6.3	3.6	<.05	$t = 2.7$, $p < .05$	$t = 0.5$, $p > .1$	$t = -1.7$, $p > .1$
N2	20.9 ± 21.6	33.5 ± 17.6	37.1 ± 13.6	3.8	<.05	$t = 2.0$, $p > .1$	$t = 2.4$, $p = .054$	$t = 0.5$, $p > .1$
N3	1.5 ± 3.4	19.7 ± 13.5	15.3 ± 12.6	21.9	<.001	$t = 6.1$, $p < .001$	$t = 4.3$, $p < .001$	$t = -1.2$, $p > .1$
REM	2.9 ± 7.1	9.7 ± 9.1	1.4 ± 2.7	5.5	<.01	$t = 2.9$, $p < .05$	$t = -0.6$, $p > .1$	$t = -2.9$, $p < .05$
TST	35.3 ± 32.2	81.3 ± 33.5	65.5 ± 14.4	12.3	<.001	$t = 4.7$, $p < .001$	$t = 2.8$, $p < .05$	$t = -1.3$, $p > .1$
RD	93.0 ± 28.1	102.2 ± 27.2	90.6 ± 6.1	0.8	>.4	x	x	x
#Stimulation	4.7 ± 8.3	220.5 ± 117.8	235.8 ± 132.5	47.5	<.001	$t = 7.9$, $p < .001$	$t = 7.8$, $p < .001$	$t = 0.5$, $p > .1$

Note. TST, total sleep time; RD, recording duration; #Stimulation, number of words presented during sleep. The last three columns show Tukey post-hoc tests values.

successful/unsuccessful encoded word pairs presented was 81.7 ± 12.0% (range: 50%–100%). Analyses were performed on relative retention scores (see *Fig. 3B*). The repeated measure ANOVA conducted on relative retention scores with between-subject factor Group (Reactivation vs. Interference) and within-subject factor TMR (Cued vs. Uncued word pairs) failed to disclose a main effect of TMR [$F(1, 26) = 0.09$, $p = .8$], but the Group [$F(1, 26) = 13.3$, $p = .001$] and the TMR by Group interaction [$F(1, 26) = 19.3$, $p < .001$] effects were significant. Post-hoc analyses evidenced lower retention scores in the Interference group as compared with the Reactivation group for cued pairs [$t(26) = 4.9$, $p < .001$], whereas no differences were observed for uncued word pairs [$t(26) = 1.1$, $p = .3$]. Also, performance for cued word pairs as compared with uncued word pairs improved in the Reactivation group [$t(13) = 5.3$, $p < .001$] but declined in the Interference [$t(13) = -2.3$, $p = .04$] group.

Nap conditions. In the Nap conditions, participants correctly recalled 89 ± 11% and 77 ± 14% of word pairs at pre- and post-TMR sessions, respectively. The ratio of successful/unsuccessful encoded word pairs presented was 86.6 ± 11.4% (range: 60%–100%). The repeated measure ANOVA conducted on relative retention scores (see *Fig. 3A*) with between-subject factor Group (No-reactivation vs. Reactivation vs. Interference) and within-subject factor TMR (Cued vs. Uncued word pairs) failed to disclose any significant effect of TMR [$F(1, 51) = 1.1$, $p = .3$], Group [$F(2, 51) = 0.8$, $p = .5$], or TMR by Group interaction [$F(2, 51) = 0.4$, $p = .7$]. Because it was reported that TMR memory benefits are dependent on prior learning (Creery et al., 2015; Cairney, Lindsey, Sobczak, Paller, and Gaskell, 2016), we also performed a repeated measure ANCOVA between cued and uncued word pairs with performance at the pre-TMR test as a covariate. Again, after correcting for cross-participants variability, there was no impact of TMR on memory performance ($p > .1$). Altogether, these results suggest that cueing during NREM sleep had no beneficial (nor detrimental) consequences on memory consolidation.

ISI-related TMR effects. In the Nap conditions, post-hoc analyses were then performed on three sets of word pairs according to the duration of the ISI: short (1,000–1,290 ms), medium (1,290–1,373 ms), and long (1,373–1,500 ms). This analysis was based on the assumption that reactivation effects during sleep may be contingent upon ISI duration (i.e., the time between the offset of the first and the onset of the second word of the pair). Indeed, a prior study showed that the effect of TMR was abolished at a short ISI (200 ms), whereas it was preserved at a long ISI (1,500 ms; Schreiner et al., 2015). Repeated measures ANOVA were conducted on retention scores separately within each set. In the short and medium ISI sets, there was no effect of the TMR procedure and no TMR by Group interaction ($p > .3$). In the long ISI set, however, the TMR effect was significant [$F(1, 23) = 4.7$, $p = .04$], with higher retention scores for cued than for uncued word pairs. In addition, the TMR by Group interaction effect was non-significant [$F(1, 23) = 0.14$, $p = .7$], suggesting a similar increase in retention scores for cued word pairs in the Reactivation and Interference groups.

Modulations of TMR-related EEG power during sleep

Sleep EEG time–frequency power for word pairs is shown in *Fig. 4*. As a reminder, analyses focused on two sets of periods of interest: during the presentation of the words (vs. baseline) and during the 1-s silent period following the presentation of the words (vs. baseline) in two frequency bands of interest, i.e., the theta and sigma bands.

The repeated measure ANOVA conducted on theta spectral power during the presentation of the words with within-subject factors Window (baseline vs. first word vs. second word) and Location (frontal vs. central) and

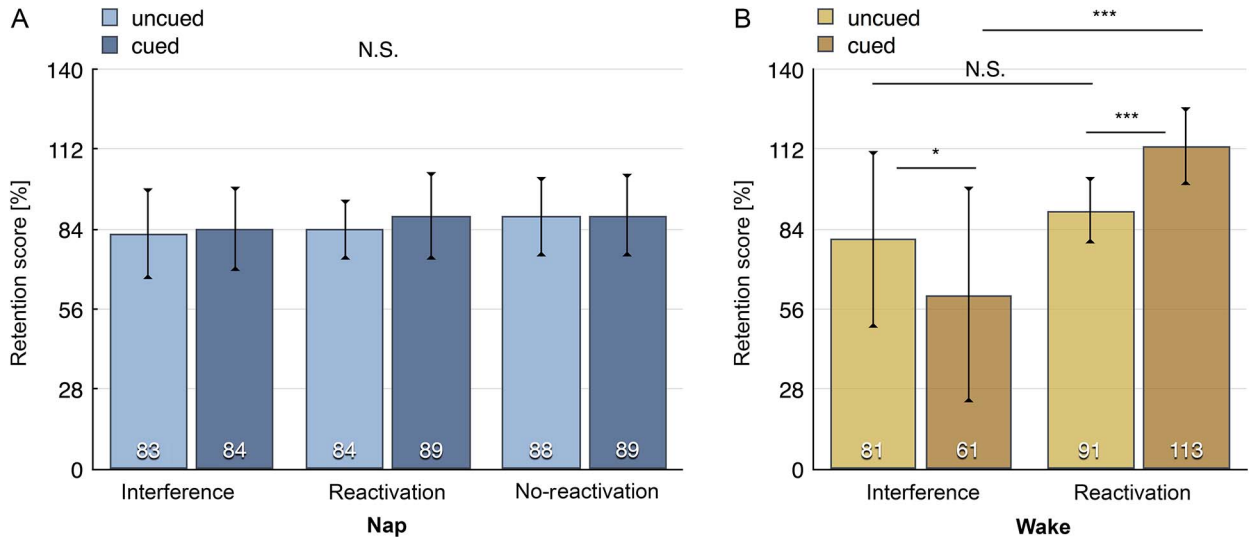

Fig. 3. Relative retention scores for cued and uncued word pairs after TMR. (A) In the Nap conditions, performance was similar in the Reactivation, Interference, and No-reactivation groups. (B) In the Wake conditions, cueing during passive wakefulness improved the performance when word pairs were identical to learning (Reactivation group), and reduced the performance when word pairs were interfering (Interference group). Bar plots are mean retention scores and error bars are standard deviations.
***$p < .001$, *$p < .05$, N.S. non-significant

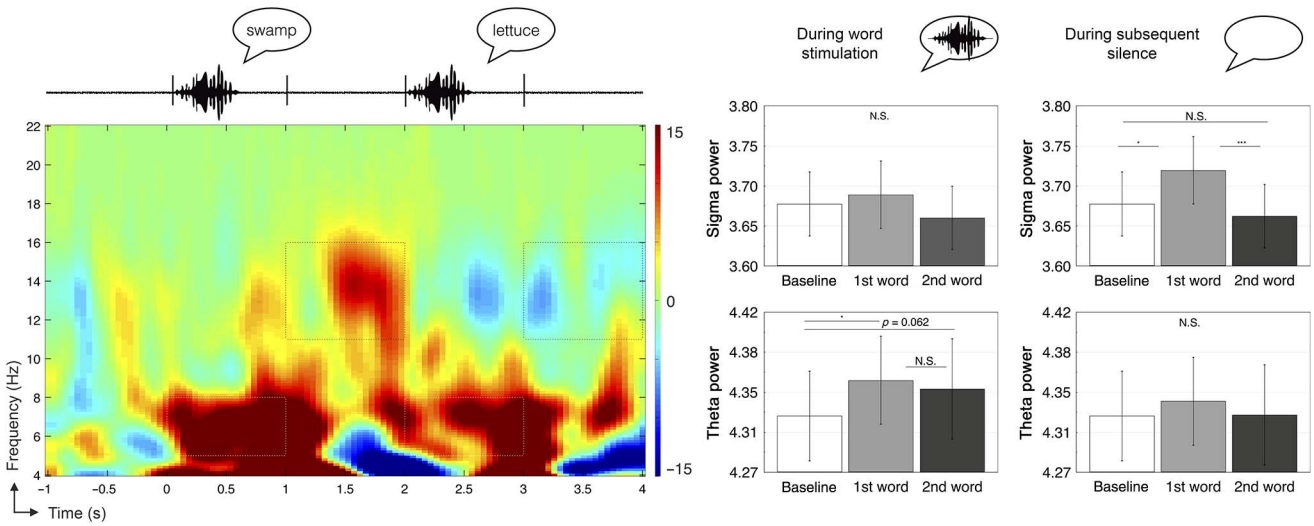

Fig. 4. Time-course of TMR-related EEG power on central electrodes during sleep. (Left) Time–frequency graph of EEG power during the auditory presentation of the two words of a pair during NREM sleep. (Right) Averaged power for periods of interest (baseline corrected with respect to the 1-s time window preceding the onset of the first word) and statistics reported. There is increased theta power at $t = 0$ with the presentation of the first word of the pair, followed by increased power in the sigma band. The onset of the second word of the pair (at $t = 1.9$ s) was also associated with increased theta power, but sigma power decreased to baseline level. Inspection of time–frequency plots suggests a decreased sigma power triggered by the onset of the second word of the pair. ***$p < .001$, *$p < .05$, N.S. non-significant

between-subject factor Group (Reactivation vs. Interference) disclosed a main Window effect [$F(2, 46) = 4.9$, $p < .05$)]. As compared with baseline, power increased during the presentation of the first word (post-hoc $t = -3.0$, $p < .05$) and exhibited a trend to increase during the presentation of the second word ($t = -2.3$, $p = .06$), at the same level than the first word (no significant differences between the first and second words: $t = 0.67$, $p = .8$). All other effects were non-significant ($p > .1$). Analyses conducted on sigma power reported no significant effects ($p > .1$).

The repeated measure ANOVA conducted on theta spectral power during the 1-s time windows following the offset of the first and second words reported no significant effects ($p > .1$). However, the same ANOVA conducted on sigma power disclosed a main Window effect [$F(2, 46) = 6.1$, $p < .01$]. Sigma power increased after the offset of the first word of the pair as compared with baseline (post-hoc $t = -2.5$, $p < .05$), then decreased during the presentation of the second word of the pair ($t = 3.4$, $p < .01$) at the same level than baseline (no significant differences between post second word and baseline: $t = 0.9$, $p = .7$).

In addition, an expected main Location effect was present in both ANOVA for both frequency bands, with higher sigma and theta power in central than frontal electrodes ($p < .05$). However, all Location by Window interaction effects were non-significant ($p > .1$).

To further investigate the TMR-related modulation effects in theta and sigma bands, we computed correlation analyses between power frequency changes in sigma and theta bands. That is, we correlated sigma power change after the first word [(1-s following the presentation of the first word of the pair − baseline)/baseline × 100] with theta power change during the first word [(presentation of the first word of the pair − baseline)/baseline × 100]. Results disclosed a monotonic relationship (Spearman correlation) between theta power changes in frontal electrodes and sigma power changes both in frontal ($R = .47$, $p < .05$) and central ($R = .44$, $p < .05$) electrodes. Theta power in central electrodes, however, did not correlate with sigma power although the relationship pointed in the same direction ($R = .19$, $p = .4$ and $R = .3$, $p = .2$ for, respectively, frontal and central electrodes).

Relations between memory performance and electrophysiological markers

In the Nap conditions, correlation analyses were computed between the memory retention scores and slow wave sleep (SWS) duration, frontal delta power, and central sigma power. Delta and sigma power were calculated over all stage N2 windows. Delta power was calculated over all stage N3 windows. SWS duration and frontal delta power were not correlated with memory retention both for cued and uncued word pairs ($p > .4$). However, sigma power on central electrodes was correlated with memory retention for uncued pairs ($R = .41$, $p = .04$) but not for cued pairs ($R = .05$, $p = .81$), although the effect did not resist correction for multiple comparisons ($p > .025$ after correction for two frequency bands). However, correlations with memory retention scores were not significantly different between cued and uncued scores (Williams $T = 1.67$, $p = .11$). Correlation plots are available in the supplementary material.

DISCUSSION

This study aimed at investigating the benefits of a TMR procedure on memory consolidation using either verbatim or interfering presentation of learned word pairs, during

subsequent wake or sleep offline periods. During the post-learning consolidation interval, half of the word pairs (cued pairs) was presented aurally with the correct (A–B) or incorrect (A–C) feedback, while the other half (uncued) was not presented. After the nap/wake period, participants were asked to recall all of the learned word pairs in a cued recall test. Results show that TMR during the offline wake period selectively enhanced (Reactivation group) or decreased (Interference group) memory recall for cued word pairs as compared with uncued word pairs. However, the same TMR procedure did not show any beneficial or detrimental effect on memory consolidation when applied during NREM sleep. Indeed, memory retention similarly decreased for cued and uncued word pairs in both conditions, to the same extent in participants who did and who did not receive stimulations during sleep. Spectral power analyses conducted on the NREM sleep EEG revealed increased sigma power after the presentation of the first word, coming back to baseline after the presentation of the second word of the pair. Theta power systematically increased during the presentation of the two words of the pair, and the change in theta power was related to the change in sigma power.

Lack of TMR-related effects during sleep?

In prior reports, the presentation during sleep of learning-related cues, or of just the first word of a word pair, selectively enhanced memory retention for cued material (e.g., Rudoy et al., 2009; Oudiette, Antony, Creery, and Paller, 2013; Schreiner and Rasch, 2015a). Here, we found that verbatim presentation of word pairs during post-learning sleep does not result in the previously reported TMR-related memory gains. Similarly, the presentation of interfering word pairs had no impact (positive or negative) on memory retention. Why is there a lack of TMR-related effects in this study? Changes in functional connectivity (van Dongen et al., 2012) or plasticity-related oscillatory responses to cues (Cox, Hofman, de Boer, and Talamini, 2014) without changes in memory consolidation have already been reported in the literature. Previous studies reported that TMR is only effective if learning is not too strong (Creery et al., 2015; Cairney et al., 2016). Likewise, sleep-related consolidation was found greater for weaker associations: in a word-pair paradigm, the benefits of sleep were evidenced when the learning criterion was set at 60% but not at 90% (Drosopoulos, Schulze, Fischer, and Born, 2007). In this study, we fixed the learning criterion to 75% based on another study conducted in our laboratory (Gilson, Farthouat, Simor, Schmitz, Peigneux, n.d.), where the cueing of unrelated sounds associated with a word-pair learning paradigm successfully improved sleep-related consolidation. Here, participants in the Nap conditions had, nonetheless, a high performance score around 90%. The lack of TMR benefits may be explained by the fact that associations were already very strong. Notwithstanding, this hypothesis seems unlikely because the absence of TMR benefits persisted after controlling for the pre-TMR recall score as a covariate.

In contrast, our results are in agreement with findings (independently published during the course of our experiment) that auditory feedbacks during sleep block the memory benefits. Indeed, Schreiner et al. (2015) showed that presenting a cue during sleep followed by the correct response (i.e., the feedback) 200 ms later actually blocked the selective memory benefits of the TMR procedure, which was not the case when the feedback was delayed by 1,500 ms or absent. Furthermore, this blockade effect was not specific to the auditory material. Presenting an unrelated pure tone 200 ms after the cue also blocked the cueing benefits and annihilated EEG-related changes. Based on these findings, the authors suggested that TMR-induced memory reactivation takes place during a critical plasticity window of at least 1 s after the cue (i.e., at least one full cycle of a sleep slow oscillation), a process actually disrupted if a novel stimulation occurs during this time period. In our experiment, the ISI varied between 1,000 and 1,500 ms. Based on Schreiner et al.'s proposal, we divided our word pairs into three sets: short, medium, and long ISI. Our results show a significant beneficial impact of TMR only on the long ISI items set (between 1,373 and 1,500 ms). However, it should be kept in mind that this is an *a posteriori* analysis, and that our paradigm was not designed to do such analysis. In particular, the sample was underpowered with only 13–14 items per set. Therefore, these results should be interpreted with caution and need further replication. Nonetheless, the results reported in this study successfully replicate and complement the finding that a long ISI is required to allow enhancing-related memory consolidation processes, and suggest that the lower boundary for ISI duration to obtain such an effect is longer than initially expected, around 1,300 ms.

A role of post-cueing time for memory consolidation

In the Nap conditions, we found that presentation of the first word of the pair (i.e., the cue) elicits increased power in the spindle-related frequency sigma band (11–6 Hz), and that this effect is abolished after the presentation of the second word (i.e., the feedback). Sigma activity in NREM sleep is a hallmark of sleep spindles, known to promote plasticity mechanisms (Rosanova, 2005). Several studies argue that spindles are related to sleep-dependent memory consolidation processes and are a neural correlate of memory consolidation (e.g., Fogel and Smith, 2011; Cox, Hofman, and Talamini, 2012; Tamminen, Ralph, and Lewis, 2013). For example, spindles density was associated with memory retention using a word-pair learning task (Schabus et al., 2004; Gais, Mölle, Helms, and Born, 2002). According to the active system consolidation theory (Born and Wilhelm, 2012), spindles play a crucial and active role in the reactivation of memories, under the top-down control of slow oscillations, for information transfer (initially stored in the hippocampus) and reallocation into neocortical stores. In line with this theory, several studies found positive correlations between spindles density or sigma power and the memory advantage (Antony, Gobel, O'Hare, Reber, and Paller, 2012; Cousins, El-Deredy, Parkes, Hennies, and Lewis 2014; Creery et al., 2015; Fuentemilla et al., 2013; Holz et al., 2012; Schreiner et al., 2015). Spindles modulations were spatially localized to learning-related areas (Cox et al., 2014), even without impact on memory retention on subsequent recall. Consequently, one possible interpretation is that increased sigma power/spindle activity following the presentation of the cue (i.e., the first word of the pair) reflects

the initiation of the brain oscillations associated with the memory reactivation transfer mechanism, a process then disrupted by the – too early – presentation of another stimulus (here, the second word). At variance with the Schreiner et al.'s (2015) study, the high level of memory performance reached by our participants at pre- and post-TMR recall sessions did not allow contrasting spectral analyses between remembered and forgotten word pairs. Therefore, we cannot ensure that our results are memory specific.

Why sigma activity is disrupted remains an open issue. Schreiner et al. (2015) and Schreiner and Rasch (2016) hypothesized that presentation of a new auditory material disrupts the neural synchronization state subtending successful associations in memory. Here, we bring new evidence that disruption of sigma synchronization is time-locked with stimulus delivery, which supports this hypothesis. A similarly disruptive effect was also documented in a rodent's study (Bendor and Wilson, 2012). After rats were trained to learn the associations between tones and food locations (either to the left or the right direction), the authors observed that re-presenting the sounds during sleep induced a biased reactivation of cue-related place cells. Interestingly for our purpose, reactivation persisted for a few seconds (up to 10 s) and stopped at the presentation of the next cue. Here, like in a prior report (Schreiner et al., 2015), sigma activity was disrupted both when an associated (learned) or a novel (interfering) word was presented. Why were there no differences between the associated and the interfering words? One possible explanation is that presentation of the second word shortly after the cue coincides with spindle activity initiated by cue presentation (a suggestion also supported by the event-related potentials analyses available in the supplementary material), eventually preventing an efficient processing of the second word. Accordingly, sleep spindles are proposed to disrupt auditory perception (Cote, Epps, and Campbell, 2000) by blocking transmission at the level of the thalamus. However, this view was recently challenged by an animal study disclosing preserved auditory responses in the primary auditory cortex during spindles in rodents (Sela, Vyazovskiy, Cirelli, Tononi, and Nir, 2016). Interestingly, the auditory stimulation induced an earlier termination of some spindles, in line with our findings. In the Sela et al.'s (2016) study, recordings were limited to primary auditory cortex. It remains possible that primary processing is intact but that higher cortical regions are affected, which would explain the non-specificity of the target. The mechanisms underlying spindles blockade (and possibly also slow and theta oscillations) still remain to be clarified.

Alternatively, there is the possibility that sigma activity related to the presentation of the first word is actually related to pure auditory processing. Accordingly, sensory stimulations during N2 sleep were shown to trigger spindles outside of the context of memory tasks (Sato, Fukuoka, Minamitani, and Honda, 2007; Andrillon, Poulsen, Hansen, Leger, and Kouider, 2016). However, we believe this interpretation unlikely since presentation of the target (i.e., the second word) did not result in increased sigma power, although we cannot completely exclude that an on-going spindle prevented the treatment of the second stimulus. It remains possible that scalp EEG reflects a mix between auditory responses and memory processes. Notwithstanding, future TMR studies should control that auditory stimulation replay (e.g., of novel word pairs) does not trigger the same typical increase in sigma power.

Presentation of the words was also systematically accompanied with an immediate increase in theta power. Modulation of theta activity was reported previously during sleep and also during wake (for a review, see Schreiner and Rasch, 2016). More precisely, increased theta power preceded increased sigma power, and the percentage of increased theta power correlated with the percentage of increased sigma power. These data support the updated theoretical model for memory reactivation during sleep (Schreiner and Rasch, 2016) proposing that theta activity, together with gamma hippocampal activity, represents the reinstatement of memory presentations, which are then stabilized when immediately followed by spindles. Note, however, that increased theta power does not guarantee in itself memory boosting, as it has to be closely followed by increased sigma.

Altogether, our results further support the hypothesis that non-specific auditory stimulation during the post-cue presentation window is detrimental to the neurophysiological processes subtending memory reactivation elicited by the first cue, and reinforce the idea that a sufficient time interval between the first and second elements of an established association is a crucial factor modulating the effectiveness of the TMR procedure on memory reactivation and the associated neurophysiological processes.

TMR during wake modulated memory consolidation

Although verbatim TMR during sleep did not benefit memory for the cued word pairs, presenting verbatim the word pairs during quiet post-learning wakefulness actually enhanced the memory for the cued items. Conversely, presenting interfering word pairs decreased the performance for the cued items. In previous studies, TMR during wakefulness has led to inconsistent results: either destabilization of memories (Diekelmann et al., 2011), improvement of performance (Oudiette et al., 2013), or no effect (Rasch et al., 2007; Rudoy et al., 2009; Antony et al., 2012; Cousins et al., 2014; Bendor and Wilson, 2012; Schreiner and Rasch, 2015b). However, in those studies, memory-related cues alone were usually presented, whereas we represented the learned material in itself, verbatim, in this study. Therefore, in the Reactivation group, cues may have also acted as reinforcement trials. These results are in agreement with previous work suggesting that when a memory is reactivated (the reactivation being triggered by the presentation of the first word of the cue), it is (re)transformed from an inactive to an active and labile state (Nadel, Hupbach, Gomez, and Newman-Smith, 2012). This memory is then either updated (with reinforcement trials) and re-stabilized (as in our Reactivation group) or put in a labile state and susceptible to interference (as in our Interference group). It should be noted that in our Interference group, the participants were not asked to remember the interference word pairs presented during the TMR procedure, although we cannot rule out that some participants did so. Also, considering that the duration of the retention interval was shorter in the Wake than in the

Nap conditions (i.e., 20 vs. 120 min), we chose not to directly compare memory retention scores between these conditions. Indeed, it cannot be precluded that retention scores might drop to lower levels in the Wake than in the Nap conditions in a situation in which retention times are equated. Notwithstanding this limitation, the analysis (reported in the supplementary material) shows that between-group effects are present for cued items only, both in the Reactivation and Interference groups.

TMR does not alter memory consolidation for uncued word pairs

We found that sigma power in NREM sleep tended to positively correlate with memory retention for uncued word pairs, but not for cued word pairs. As stated above, there is compelling evidence that sigma power participates in memory consolidation processes (Schabus et al., 2004; Gais et al., 2002). This result suggests that even if verbatim TMR of word pairs was inefficient to improve memory for the cued word pairs, it had no detrimental effect on memory for the uncued word pairs. Also, the observation of persistent associations between sigma power and memory retention for uncued but not for cued word pairs in this study provide indirect evidence for a specific detrimental effect of the TMR procedure. Recent research has found this type of dissociation between correlations of sleep parameters (in that case SWS duration) and memory decays of cued and uncued items (Cairney et al., 2016). Also, SWS duration correlated with performance improvement in the natural 90-min sleep condition, but not in the 40-min cued sleep condition (Diekelmannet al., 2011). Cairney et al. (2016) hold the view that TMR takes on the naturally sleep-dependent memory consolidation process (linked to SWS and spindles). Still, this proposal is speculative but may represent an interesting future area of research.

CONCLUSIONS

In this study, we demonstrate that TMR using verbatim or interfering verbal associations exerts an impact on the consolidation of target associations in memory during offline post-learning wakefulness but not during NREM sleep. We propose that the TMR-related neurophysiological processes subtending memory reactivation and further integration in memory stores are annihilated/disrupted when the cue is followed shortly after by a second auditory stimulation. Our data bring new evidence in support of the hypothesis that successful memory reactivation during sleep requires a sensitive silent plasticity period following TMR. Further studies are needed to evaluate the importance of the temporal dynamics of cueing processes during sleep.

Authors' contribution: JF and MG designed the experiment, acquired the data, ran the analyses, and wrote the manuscript. PP designed the experiment, contributed to the analyses, and wrote the manuscript.

Conflict of interest: The authors declare no conflict of interest.

Ethics: This study was conducted in accordance with the ethical principles in the Declaration of Helsinki and was approved by the local research ethics committee (Université libre de Bruxelles, 017/2012). Before participation, all subjects gave written informed consent.

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Electronic Supplementary Material (ESM)

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ESM1

Table S1. Words used to constitute learning and interfering word pairs (see the main text). Stimuli are unrelated French words, neutral, two-syllabic. Word pairs are, for each participant, randomly tagged at the beginning of the session as “to be reactivated” or “no reactivation”

Fig. S1. Correlation between mean sigma power over N2 sleep periods on central electrodes (C3 and C4) and memory retention for uncued (left, $p < .05$) and cued (right, $p > .8$) word pairs. p-Value threshold after correction for multiple comparisons is $p = .025$

Fig. S2. Grand average of the evoked-related potentials (band-pass filter: 0.9–30 Hz) of the representative electrode FP1 for the first (in green) and second (in blue) words of the pair. Onset of auditory stimuli was set at $t = 0$. Shaded error bars represent the standard error. The black bold line at the bottom indicates the time window for which the difference between both ERPs was significant

Fig. S3. Reactivation group

Fig. S4. Interference group