Neurobiology of Learning and Memory 109 (2014) 122–130

Contents lists available at ScienceDirect

Neurobiology of Learning and Memory

journal homepage: www.elsevier.com/locate/ynlme

Sleep deprivation during a specific 3-hour time window post-training impairs hippocampal synaptic plasticity and memory

Toni-Moi Prince [a], Mathieu Wimmer [b,1], Jennifer Choi [c,1], Robbert Havekes [c], Sara Aton [d], Ted Abel [c,*]

[a] Neuroscience Graduate Group, University of Pennsylvania, Philadelphia, PA 19104, USA
[b] Department of Psychiatry, University of Pennsylvania, Philadelphia, PA 19104, USA
[c] Department of Biology, University of Pennsylvania, Philadelphia, PA 19104, USA
[d] Department of Molecular, Cellular & Developmental Biology, University of Michigan, Ann Arbor, MI 48109, USA

ARTICLE INFO

Article history:
Received 5 August 2013
Revised 22 October 2013
Accepted 21 November 2013
Available online 28 December 2013

Keywords:
Sleep loss
Long-term potentiation
Hippocampus
Spatial memory

ABSTRACT

Sleep deprivation disrupts hippocampal function and plasticity. In particular, long-term memory consolidation is impaired by sleep deprivation, suggesting that a specific critical period exists following learning during which sleep is necessary. To elucidate the impact of sleep deprivation on long-term memory consolidation and synaptic plasticity, long-term memory was assessed when mice were sleep deprived following training in the hippocampus-dependent object place recognition task. We found that 3 h of sleep deprivation significantly impaired memory when deprivation began 1 h after training. In contrast, 3 h of deprivation beginning immediately post-training did not impair spatial memory. Furthermore, a 3-h sleep deprivation beginning 1 h after training impaired hippocampal long-term potentiation (LTP), whereas sleep deprivation immediately after training did not affect LTP. Together, our findings define a specific 3-h critical period, extending from 1 to 4 h after training, during which sleep deprivation impairs hippocampal function.

1. Introduction

Despite the fact that sleep exposes an organism to increased risk of attack or predation due to time spent unaware of its surroundings, sleep is an evolutionarily conserved phenomenon that is critical for survival. Inadequate sleep contributes to both physical and mental exhaustion and deterioration. Modern society suffers from unprecedented rates of sleep loss. According to the Center for Disease Control, 7–19% of adults in the US report inadequate sleep, and an estimated 70 million Americans suffer from chronic sleep disorders. Although the vital function of sleep has yet to be determined, lack of sleep is detrimental to cognitive function.

One of the most notable negative consequences of sleep loss is impaired memory (Harrison & Horne, 2000). Memory is composed of at least three stages; acquisition, consolidation, and retrieval (Abel & Lattal, 2001). The effects of sleep deprivation have been examined on both the acquisition and consolidation of memory. Early studies explored the effects of sleep deprivation on memory acquisition (Stern, 1971), and it has since been repeatedly demonstrated that chronic sleep deprivation impairs acquisition (learning) (Abel, Havekes, Saletin, & Walker, 2013; Chee & Choo, 2004; Durmer & Dinges, 2005; Hagewoud et al., 2010; Havekes, Vecsey, & Abel, 2012; Prince & Abel, 2013; Van Der Werf et al., 2009; Youngblood, Zhou, Smagin, Ryan, & Harris, 1997). More recently, however, multiple laboratories have explored the effects of acute sleep deprivation and sleep fragmentation during consolidation, showing that consolidation benefits from sleep and is hindered by sleep loss (Florian, Vecsey, Halassa, Haydon, & Abel, 2011; Graves, Heller, Pack, & Abel, 2003; Hagewoud et al., 2010; Hagewoud et al., 2010; Inostroza, Binder, & Born, 2013; Rolls et al., 2011; Vecsey et al., 2009).

Consolidation in hippocampus-dependent memory tasks is particularly sensitive to sleep loss. Sleep deprivation-induced deficits have been described for associative memory tasks such as contextual fear conditioning and for spatial memory tasks such as the Morris water maze task and the object-place recognition (OPR) task, which is used in the present study (Binder et al., 2012; Florian et al., 2011; Graves et al., 2003; Smith & Rose, 1996; Smith & Rose, 1997). OPR, in particular, is an ideal paradigm for examining the effects of sleep deprivation on hippocampal function because it is comparable to tasks that test declarative memory in humans, it is dependent on the hippocampus, and it is not aversive (Bussey, Duck, Muir, & Aggleton, 2000; Mumby, Gaskin, Glenn, Schramek, & Lehmann, 2002; Oliveira, Hawk, Abel, & Havekes, 2010; Shrager, Bayley, Bontempi, Hopkins, & Squire, 2007; Winters, Forwood, Cowell, Saksida, & Bussey, 2004; Winters, Saksida, & Bussey, 2008).

Several studies have assessed whether sleep deprivation during specific time windows after training affects long-term memory

* Corresponding author. Address: Department of Biology, University of Pennsylvania, Philadelphia, PA 19104-6018, USA. Fax: +1 215 573 1297.
E-mail address: abele@sas.upenn.edu (T. Abel).
[1] These authors made equivalent contributions to the paper.

1074-7427/$ - see front matter © 2013 Elsevier Inc. All rights reserved.
http://dx.doi.org/10.1016/j.nlm.2013.11.021

(Graves et al., 2003; Palchykova, Winsky-Sommerer, Meerlo, Dürr, & Tobler, 2006; Smith & Rose, 1997). In these studies, a sleep deprivation sensitive window, within the first 4–6 h of consolidation, has been demonstrated in the consolidation of contextual memory, object recognition memory, and spatial memory. Memory is resistant to the effects of sleep deprivation if animals are sleep deprived after this window (Graves et al., 2003; Palchykova et al., 2006; Smith & Rose, 1997). These findings suggest that there is a critical period during which memory is vulnerable to the effects of sleep deprivation. However, little work has been conducted to examine the precise timing of this sensitive time window. The aim of this study was to define the temporal parameters for the impact of sleep deprivation on memory consolidation.

Aside from the effects of sleep deprivation on behavioral measures of memory, sleep deprivation also disrupts synaptic plasticity, a neural correlate of memory. Campbell and colleagues demonstrated that 12 h of sleep deprivation impairs hippocampal long-term plasticity (LTP), a form of synaptic plasticity (Campbell, Guinan, & Horowitz, 2002). LTP deficits have been observed *in vitro* after 4–5 h of sleep deprivation as well (Kopp, Longordo, Nicholson, & Lüthi, 2006; Vecsey et al., 2009). The impact of short periods of sleep deprivation is specific to late-phase LTP (L-LTP), which requires protein synthesis and the cyclic adenosine monophosphate (cAMP) signaling pathway (Vecsey et al., 2009). Critically, however, the effects of sleep deprivation on hippocampal LTP during a period of active memory consolidation have not previously been examined. By assessing hippocampal LTP following training, in the sensitive window for sleep deprivation, we aimed to more accurately determine the effects of sleep and sleep loss on hippocampal plasticity associated with memory consolidation.

Previously, we demonstrated that as little as 6 h of sleep deprivation immediately after task training disrupts long-term spatial memory in OPR (Florian et al., 2011). Here we aim to better define the critical period during which sleep is essential for hippocampal memory consolidation. By sleep depriving mice during two different time windows, we demonstrate that as few as 3 h of sleep deprivation during consolidation can affect both long-term memory and LTP.

2. Methods

2.1. Subjects

One hundred C57BL/6J adult male mice (2–4 months of age) were pair housed and kept on a 12 h/12 h light/dark schedule with lights on at 7:00 AM (ZT 0). Food and water were available *ad libitum* throughout the experiments. All experiments were approved by the Institution of Animal Care and Use Committee of the University of Pennsylvania and were carried out in accordance with all National Institutes of Health guidelines.

2.2. Sleep deprivation

To assess the effects of sleep deprivation (SD) on memory, mice ($n = 58$) were sleep-deprived using the gentle handling technique involving manual cage tapping, cage shaking, nestlet disturbance, and gentle animal prodding (Ledoux, Sastre, Buda, Luppi, & Jouvet, 1996; Vecsey et al., 2013). Prior work using electroencephalographic recordings has shown that this procedure effectively retains animals in a state of wakefulness for several hours (Meerlo, De Bruin, Strijkstra, & Daan, 2001). The frequency of these manipulations was monitored throughout the sleep deprivation period (Fig. 5A and B). Separate groups of mice were sleep deprived in one of the two 3-h periods for behavior and electrophysiology experiments (ZT 1–4 and ZT 2–5) after behavioral training as described in Fig. 1A and B. Non-sleep deprivation (NSD) time-matched control groups were used for comparison with the two SD experimental groups.

2.3. Object-place recognition (OPR)

For this task, we used a previously established design that has been shown to be hippocampus dependent (Fig. 1A; Havekes, Canton, et al., 2012; Havekes, Vecsey, et al., 2012; Oliveira et al., 2010). Mice ($n = 80$) were handled for 2 min each day, for 6 consecutive days leading up to experimentation. The task was conducted in a grey rectangular box (40 cm × 30 cm × 30 cm) built of polyvinyl chloride plastic. At the beginning of the light phase (ZT 0), mice were placed in the empty box for 6 min for habituation. Mice were then removed and placed back in the home cage. After 3 min, mice were placed in the box with 3 different objects (a 100 ml glass bottle, a white cylinder, and a metallic rectangular tower) for 3 consecutive 6-min training sessions. Each training session was separated by a 3-min interval during which the animals were returned to the holding cages. At completion of the training sessions, NSD mice were left undisturbed in their home cages and SD mice were deprived of sleep by gentle handling. Twenty-four hours following the training session, mice were re-introduced to the spatial context in a single test session. In this session, one of the objects was repositioned (the displaced object: DO), thereby changing the spatial configuration of the objects in the box. Mice were allowed to explore objects for 6 min. Exploration was recorded during training

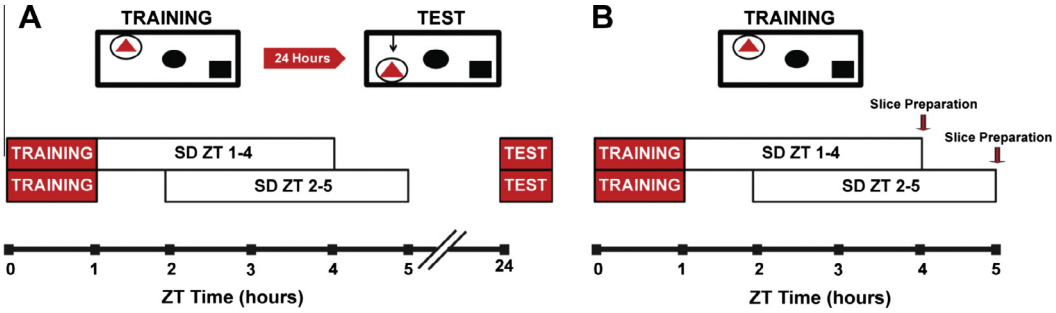

Fig. 1. *Schematic depicting the behavioral and LTP experimental design.* (A) Behavioral experimental design: The top diagram depicts the OPR task used to examine hippocampus-dependent memory. The 3 training sessions include repeated exposure to 3 distinct objects. Training sessions began at lights on ZT 0. The test session occurred 24 h following training. The bottom diagram depicts the post-training sleep deprivation time periods for each behavioral experiment. After the last training session, mice were subjected to either an immediate or delayed 3-h sleep deprivation period to assess the specific time window for sleep deprivation to impair memory. (B) LTP experimental design: The top diagram demonstrates that mice were subjected to the same training as those that were in the behavioral experiment assessing memory. However, there was no later test period for these animals. The bottom diagram depicts the sleep deprivation periods after training as well as when hippocampal slices were collected for field recordings. Recordings obtained from NSD control groups were later pooled, and LTP from the sleep deprivation groups were compared to this pooled group. Prior to pooling the data from the NSD control groups, we determined that the NSD control groups were not significantly different from each other.