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SLEEP AND LEARNING

Sleeping through brain excitation and inhibition

Sleep is controlled by a cocktail of neurotransmitters, but it is difficult to measure these in the brain. A new study by Tamaki et al. reveals how the balance between excitation and inhibition oscillates as the brain moves through sleep stages and how this impacts upon memory consolidation and stabilization.

Sofia Pereira and Penelope A. Lewis

When you fall asleep, does your brain really just switch off? Of course not; you may feel rested after a night of sleep, but your neural circuitry has been actively working its way through a complex architecture of sleep stages. Sleep provides a series of carefully interleaved, yet very different, states of cognitive and physiological processing. These states are defined not only by stereotyped patterns of neural activity, but also by very specific changes in pharmacological milieu. Unfortunately, it is notoriously difficult to measure neurotransmitter concentrations within the healthy human brain, although this can be done through dialysis in non-human animals[1].

In this issue of *Nature Neuroscience*, Tamaki and colleagues overcome this challenge by using magnetic resonance spectroscopy (MRS) in conjunction with polysomnography for sleep staging to determine concentrations of GABA and Glx (a combined measure of glutamate and glutamine) in healthy sleeping humans[2]. Using this novel combination of methods, they calculate the excitatory–inhibitory ratio (termed 'E/I balance') in visual cortex during wakefulness, rapid eye movement (REM) sleep and non-REM (NREM) sleep. The E/I balance is the ratio between Glx and GABA and has been proposed as a reliable index of plasticity[3]. Tamaki et al. show an increase in E/I balance during NREM, driven by a decrease in GABA, and a decrease in E/I balance during REM, driven by a decrease in Glx.

In addition, Tamaki et al. used the texture discrimination task (TDT) to probe visual learning before and after sleep. The TDT involves identification of a central letter (the test stimulus) while simultaneously detecting the orientation of a target array in the visual periphery (three lines in either horizontal or vertical orientation). A visually confusing mask stimulus is presented at a varying delay after the target array, and participants gradually learn to identify the target even when this delay is very short.

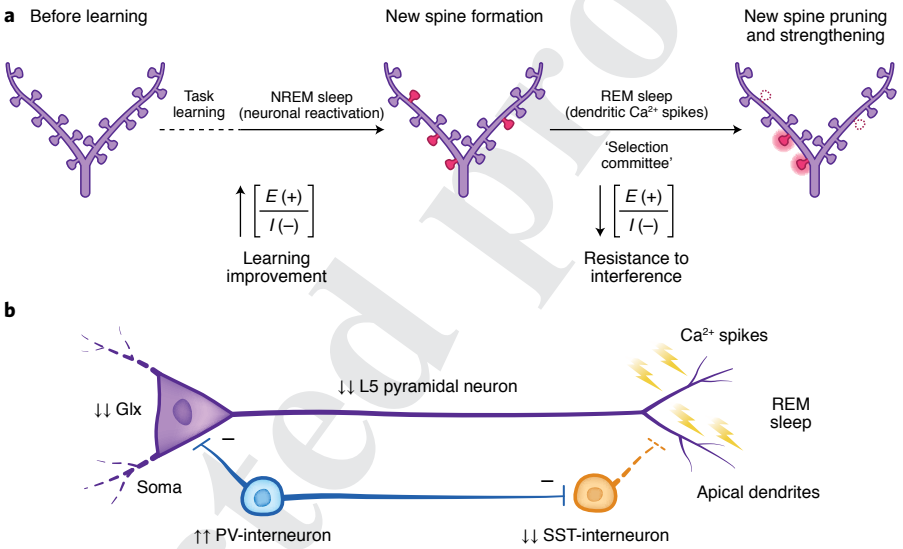

Fig. 1 | [Insert figure title.] **a**, Learning leads to new spine formation (red spines) during NREM sleep, accompanied by an increase in the E/I balance, which manifests as performance improvement at a behavioral level. Next, REM sleep acts to strengthen some of the new spines and prune others, while the overall E/I balance is decreased and performance is stabilized and protected from interference. **b**, During REM sleep, glutamatergic L5 pyramidal neurons in the motor cortex undergo strong suppression of firing (dashed purple lines and downward arrows) due to perisomatic inhibition by parvalbumin (PV) interneurons (blue, upward arrows), which also inhibit somatostatin (SST)-expressing interneurons (dashed orange lines and downward arrows). The apical dendrites, now released from SST-mediated inhibition, are subject to calcium spikes, which drive spine pruning and strengthening. The soma, however, is prevented from firing and releasing glutamate (Glx, downward arrows), which we speculate could result in an overall decrease in the E/I balance, as reported in the visual cortex by Tamaki et al.[2]. Figure adapted from ref. [7].

Tamaki et al. found a relationship between visual learning as gauged by the TDT and shifts in E/I balance in the sleeping visual cortex. Thus, while the E/I increase in NREM occurred whether or not participants had learned the TDT before sleep, the E/I decrease in REM occurred only after learning the task. Additionally, the NREM increase predicted task improvement across sleep, while the REM decrease predicted the extent to which pre-sleep learning is protected against interference from post-sleep learning of a similar task.

The authors interpret these findings as evidence for complementary roles of NREM and REM sleep, with the former providing a highly plastic milieu conducive to general performance enhancement and the latter offering stabilization of these offline gains by rendering them resistant to retrograde interference. The idea of complementary roles for NREM and REM sleep is in good keeping with the literature, in which many authors have speculated about the interdependence of sleep stages[4]. However, the idea that plasticity occurs only during NREM, while REM is dedicated to stabilization, is decidedly out of keeping with this literature. Importantly, however, this apparent clash is due to a difference

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in terminology, since Tamaki et al. are using systems-level definitions and thus define 'plasticity' as a state in which visual learning is labile and 'stabilization' as a state of learning that is resilient to retrograde interference. At the physiological level, a growing body of evidence has implicated REM sleep in various forms of synaptic and intracellular plasticity, from the expression of immediate early genes and the translation of plasticity-related products[5] to spine remodeling, including both strengthening and pruning[6]. In a very elegant study, Li and colleagues showed that motor learning in mice is followed by a surge of new spines in the apical dendrites of layer 5 (L5) pyramidal neurons in the motor cortex during NREM sleep. These newly formed spines then undergo what could be thought of as strict scrutiny by a selection committee during REM, which maintains and strengthens some spines while pruning others[6] (Fig. 1a). Although the exact guidelines used in this selection process remain unclear, they appear to be linked to local dendritic calcium spikes in L5 pyramidal neurons[6]. These calcium spikes produce a highly plastic state in the apical tuft which stabilizes new spines and eliminates others. However, due to interneuron-mediated inhibition, action potentials are strongly suppressed, despite the dendritic calcium spikes[7] (Fig. 1b). We speculate that this increased inhibitory activity during REM sleep could also result in an overall decrease in the E/I balance in the motor cortex, as shown by Tamaki and colleagues in visual areas.

Importantly the E/I decrease in REM did not predict consolidation of a learned visual task in Tamaki and colleagues' data. Instead, it predicted protection of that material against potential interference induced by learning a very similar but critically distinct visual task after sleep. The authors therefore suggest that REM sleep is critical for stabilization of the pre-sleep learning. This is entirely consistent with the pattern of selective spine preservation which Li and colleagues observed in REM[6] (Fig. 1a). One might speculate that the strengthening of selected spines represents a physical manifestation of stabilization, while the pruning of other spines, which makes space for new spines to grow at the next learning opportunity, allows for subsequent learning without overlap or interference.

Taken together, these findings support a model in which the increase in the E/I balance during NREM sleep, possibly related to neuronal reactivation[8], leads to a highly labile state characterized by new spine formation on a cellular level[6] and by offline performance gains on a behavioral level[2]. The decrease in the E/I balance that seems to characterize REM sleep might tilt the scale toward suppression of neuronal firing[7], thus enabling resources to be reallocated to a cascade of intrasynaptic plasticity-related events, which culminate in selective spine strengthening and pruning[6] and stabilization of behavioral performance gains[2].

In reality, of course, no matter how much we might speculate about the relationship between E/I balance and plasticity, MRS measures neurotransmitter concentrations, and Tamaki and colleagues' findings are most parsimoniously interpreted in this light. Although MRS suffers from various imprecisions, such as its low spatial and temporal resolution, detection of both intra-and extrasynaptic GABA and glutamate (which blurs any link between neurotransmitter concentration and cell firing) and intrinsic biochemical challenges (since glutamate is GABA's precursor)[9], these findings appear to be in line with the results of at least some studies using pharmacological manipulations in sleeping humans[10]. For instance, administration of tiagabine, a GABA reuptake inhibitor known to increase slow wave sleep (SWS) duration, significantly impaired procedural memory performance compared to the placebo, while declarative memory was unaffected[10]. In light of the new MRS findings from Tamaki et al.[2], one might speculate that the extra GABA tipped the E/I balance in the wrong direction: toward a decrease (more inhibition) instead of an increase (more excitation), potentially preventing the usual NREM-related plasticity from occurring. REM sleep duration was also significantly reduced in this study, which might have diminished the extent to which the new motor skill could be stabilized.

As a side note, it is well known that GABA initializes NREM sleep[11], so the new findings that GABA decreases during NREM and that such decreases even explain performance gains feel paradoxical. Tamaki et al. acknowledge this and offer a potential solution. They suggest that the GABA that initiates this sleep stage may also inhibit GABAergic interneurons, leading to a net decrease in this neurotransmitter. This requires further investigation.

Tantalizingly, Tamaki et al.[2] find that REM theta has a negative relationship with REM E/I balance, as well as a positive relationship with the protection of pre-sleep learning against interference. The authors are careful not to over-interpret this finding, given that electroencephalography data collected in the MRI scanner are understandably noisy. However, REM theta is known to be important for memory, since its disruption can lead to deficits in object–place memory and fear conditioning[12]. REM theta disruption may even prevent downscaling, eventually leading to oversized place fields and spatial memory deficits[13]. Although the above studies both focus on hippocampal theta, it is still tempting to ponder whether their findings might extrapolate to cortical theta and, if so, whether a causal relationship might exist between reductions in cortical E/I balance in REM, the corresponding cortical theta increases and the synaptic pruning process that seems to support stabilization.

REM sleep's function has remained elusive since its discovery in 1952 by Eugene Aserinsky, with proposals ranging from emotional processing to problem-solving and memory corticalization[14,15]. Tamaki and colleagues' new approach enables us to take a fresh look at this old problem by taking into consideration the balance between excitatory and inhibitory neurotransmission and its effects on consolidation.

At a more general level, the pioneering combination of MRS and polysomnography paves the way for more studies of how neurotransmitters fluctuate across sleep within specific structures of the human brain. For instance, it would be valuable to determine whether the E/I balance varies across sleep in a similar manner in other cortical areas and how such measures relate to memory replay as well as to consolidation of declarative or motor tasks. Furthermore, given our above speculation about the importance of theta for pruning and memory processes, it would be interesting to search for relationships between E/I balance, theta and consolidation in a variety of cortical regions.

Far from switching off for the night, lying down to sleep is now known to be the cue for many brain processes to begin gearing up. This valuable work from Tamaki et al. introduces an exciting new method through which we can begin to better understand these processes. ❐

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References

1. Vanini, G., Lydic, R. & Baghdoyan, H.A. *Sleep (Basel)* **35**, 1325–1334 (2012).
2. Tamaki, M. et al. *Nat. Neurosci.* https://doi.org/10.1038/s41593-020-0666-y (2020).
3. Bang, J. W. et al. *Nat. Hum. Behav.* **2**, 507–513 (2018).
4. Giuditta, A. et al. *Behav. Brain Res.* **69**, 157–166 (1995).
5. Seibt, J. & Frank, M. G. *Front. Syst. Neurosci.* **13**, 2 (2019).

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6. Li, W., Ma, L., Yang, G. & Gan, W.-B. *Nat. Neurosci.* **20**, 427–437 (2017).
7. Sun, L., Zhou, H., Cichon, J. & Yang, G. *Phil. Trans. R. Soc. Lond. B* https://doi.org/10.1098/rstb.2019.0234 (2020).
8. Yang, G. et al. *Science* **344**, 1173–1178 (2014).
9. Shibata, K. et al. *Nat. Neurosci.* **20**, 470–475 (2017).
10. Feld, G.B. et al. *Sleep (Basel)* **36**, 1317–1326 (2013).
11. Chowdhury, S. et al. *eLife* **8**, e44928 (2019).
12. Boyce, R., Glasgow, S. D., Williams, S. & Adamantidis, A. *Science* **352**, 812–816 (2016).
13. Swift, K. M. et al. *Curr. Biol.* **28**, e4 (2018).
14. Landmann, N. et al. *Neurobiol. Learn. Mem.* **122**, 28–40 (2015).
15. Almeida-Filho, D. G., Queiroz, C. M. & Ribeiro, S. *Cell. Mol. Life Sci.* **75**, 3715–3740 (2018).

Competing interests

The authors declare no competing interests.

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