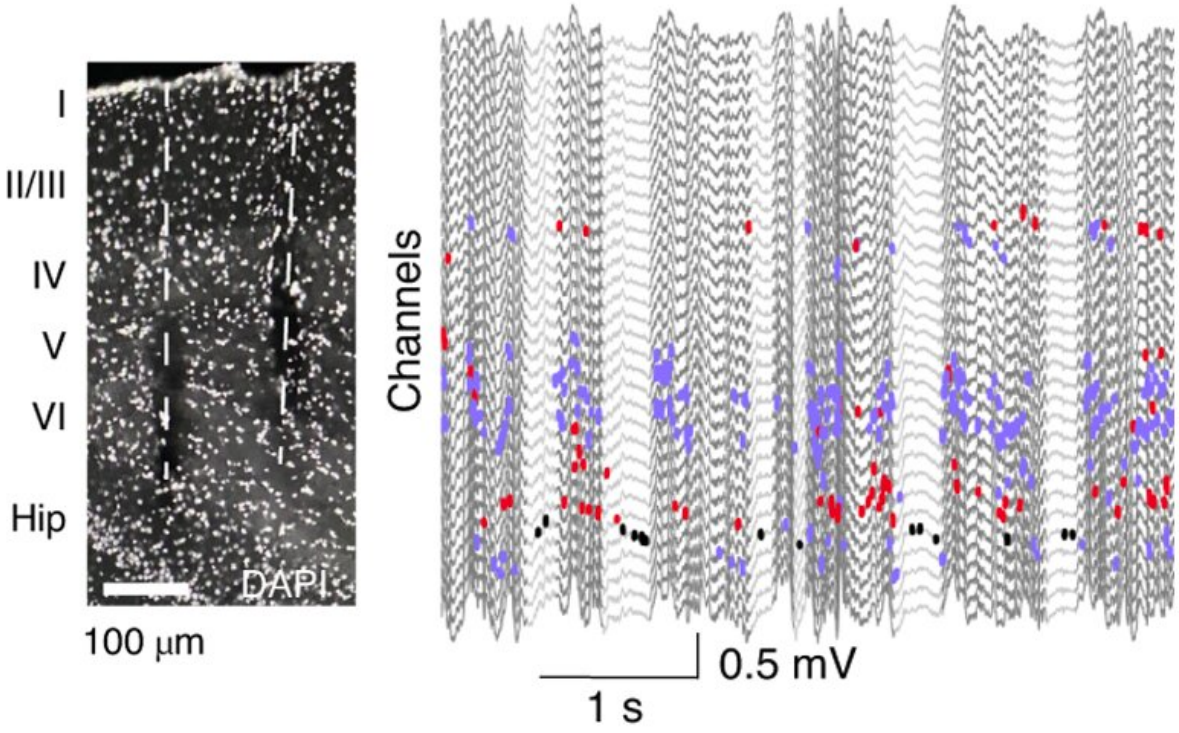
Researchers identify a class of neurons that are most active during non-REM sleep

March 16 2021, by Ingrid Fadelli



Sample recording showing the action potentials of a DOWN-state active cell (in black dots) spiking during the DOWN-state (light gray traces) of NREM sleep, flanked by action potential from pyramidal cells (red dots) and other inhibitory interneurons (blue dots). A brain histological section on the left shows the electrode position tract across all cortical layers. (histology may not be important). Credit: Valero et al

Typically, pyramidal cells and GABAergic interneurons in the brain are activated simultaneously. A team of neuroscientists at New York University, however, recently identified a unique class of neurons that do not fire at the same time as all principal neurons, cells and interneurons. Interestingly, the team found that these specific neurons are most active during the DOWN state of non-REM (NREM) sleep, when all other neuron types are silent.

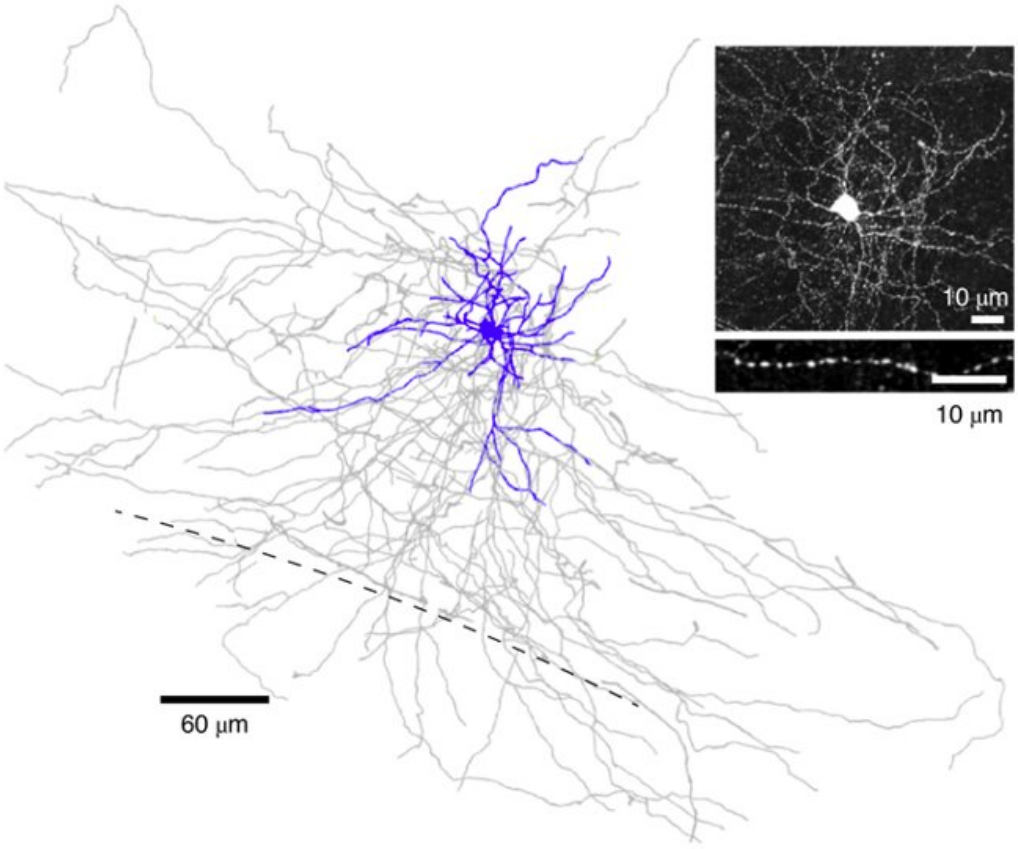
"As is often the case in science, our discovery was a true serendipity," György Buzsáki, one of the researchers who carried out the study, told MedicalXpress. "By collecting sleep recordings in deep layers of the cortex, we observed that spikes of some rare neurons occasionally occurred during the so-called 'DOWN state' epochs of sleep. No neuron was supposed to do such thing, as DOWN state is known (and identified by) by its complete neuronal silence (lack of spikes)."

The neocortex, a set of layers in a region of the brain called cerebral cortex, is rebooted thousands of times every night from the transient (50-300 ms long) DOWN state. In their study, Buzsáki and his colleagues identified a class of neurons that appear to be most active when all other neurons (i.e., excitatory pyramidal and inhibitory neurons) are silent, in the DOWN state, during NREM stages of sleep. In their follow up experiments, they showed that these neurons are neuroglia-form cells found in the deeper layers of the neocortex, which specifically express genes known as ID2 and Nkx2.1.

When they examined this class of neurons more in depth, Buzsáki and his colleagues observed that they had an entirely antagonistic relationship with all other known types of neurons in all wakefulness states (i.e., both when mammals are awake and asleep). This suggests that these neurons could have a unique function that sets them apart from all other cells in the brain.

"The reason why neither we nor others have seen these neurons earlier is that they are very rare and require methods to record from large ensembles of neurons in behaving animals," Buzsáki explained. "In several recordings of brain activity collected from animals, we found none, one and rarely two or more of these neurons among the hundreds we surveyed. Given the novelty and suspected importance of these neurons, Manuel Valero, a postdoctoral fellow in our group, decided to do a broad scale experiment by examining all known interneuron types whose molecular identity allowed their identification in a behaving/sleeping animal."

Initially, Valero examined a group of cells called nitric oxide-expressing neurons (nNOSs), as previous findings suggest that the volatile molecule nitric oxide plays a key role in enhancing slow brain waves during NREM sleep. Interestingly, they found that a very small fraction of nNOSs fired during DOWN states of NREM sleep.



3-dimensional reconstruction of soma, dendrites and axonal arborization of an intracellularly filled lD2/Nkx2.1 neuron. Credit: Valero et al.

"One day, as we were in our university's science building's elevator, we were talking to a colleague who mentioned that they were working on an unusual group of neurogliaform cells in deep cortical layers, which might be important because only this interneuron group increases disproportionally in the primate brain," Buzsáki said. "This colleague called them ID2/Nkx2.1, a small subgroup of nNOS family. By the end of the elevator ride, we agreed on a collaboration."

Manuel Valero then showed that when a light-sensitive channel was expressed by this unique class of interneurons, shining light on them by neuron-size microscopic LED lights revealed their identity.

The most important information that the investigators learned is that almost all ID2/Nkx2.1-expressing neurons were active specifically during the DOWN states of nonREM sleep. This finding could have important implications, as it suggests that these neurons have crucial physiological functions related to the activity of the mammalian brain when mammals or other humans are asleep.

"Discovering a new neuron type in the brain is a rare event," Buzsáki said. "Discovering a neuron whose activity is antagonistic in every possible way to all other known neurons is truly unexpected, almost like finding an English-speaking dog. All other tested interneurons inhibited our DOWN-state active neurons. They may be critical in adjusting the duration of the silent states and affecting the recruitment sequence of pyramidal cells when they reboot themselves from the silent state, as our study showed."

In addition to identifying this class of neurons that is particularly active in DOWN states of NREM sleep, Buzsáki and his colleagues showed that their artificial activation interferes with sleep-assisted enhancement of memory. While the evidence they collected might not be substantial enough to speculate on these neurons' entire functional repertoire, their unusual interaction with other brain cells hints at their possible involvement in a variety of different neural computations. New works examining these neurons could help to better understand their functions, perhaps unveiling their role in very specific physiological processes.

"In our next studies, we plan to focus on a number of research questions, such as will the activation/inactivation of the neurons we identified affect sleep structure? Do they have special receptor types which may be

selectively targeted by drugs? What do they do when they spike during waking? Are they present with the same ratio in each cortical region or are they enriched in some areas? And finally, why are these neurons more common in the human brain?"

More information: Sleep down state-active ID2/Nkx2.1 interneurons in the neocortex. *Nature Neuroscience*(2021). DOI: 10.1038/s41593-021-00797-6.