MedicalXpress

Light shone within brains of mice reveals secrets of sleep-wake cycle

October 19 2007

By flickering a special light inside the brains of sleeping mice to wake them up, Stanford researchers have shown that they can induce behavior in a living mammal by directly controlling specific neural cells. In so doing, they have answered fundamental questions about the process of waking up.

The research was published online this week in the journal *Nature*.

Researchers led by Karl Deisseroth, an assistant professor of bioengineering and of psychiatry and behavioral sciences, reported in April (*Nature*, April 5, 2007) they could genetically alter some kinds of brain cells to either increase or decrease their activity in response to pulses of different colors of light. Those experiments, done in brain tissue slices and with simple worms, were proofs of concept on a basic level.

"The new things in this paper are that first of all it's done in a freely moving, living mammal, delivering the light in deep brain tissue," Deisseroth said. "Secondly, we are asking and answering a real question relevant to neuropsychiatry."

Specifically, Deisseroth's group teamed up with that of neuroscientist and sleep researcher Luis de Lecea, an associate professor of psychiatry and behavioral sciences at Stanford. The team set out to determine whether the neurotransmitter hypocretin, long correlated with wakefulness and notably absent in narcolepsy patients, directly causes

waking and, if so, how long it takes for increased hypocretin activity to have that result.

"The mechanism by which these [hypocretin-producing] cells stabilize the sleep-wake cycle has been unknown," de Lecea said. "We knew that when they were missing we had this very unstable cycle, but we didn't know until now what were the actual effects of stimulating hypocretin neurons in the cycle."

The paper's lead authors are psychiatry postdoctoral researcher Antoine R. Adamantidis and bioengineering and chemistry doctoral student Feng Zhang. Bioengineering postdoctoral fellow Alexander Aravanis also is an author.

To rouse a mouse

In the study, the researchers used a virus to insert genes for producing light-sensitive proteins into hypocretin-producing cells in the lateral hypothalamus of mice, the area of the brain responsible for many primal behaviors. They left other mice unaltered as a control. The altered cells were engineered to increase their hypocretin delivery when blue light was shone. The researchers then implanted the fiber optics and exposed the lateral hypothalamus in each mouse to different frequencies of bursts of blue light.

What the researchers found was that mice whose cells had been altered woke up twice as fast when exposed to the light pulses as the mice that were unaltered, strongly suggesting a causal role for hypocretin. In addition they found that the increased hypocretin typically accomplished this result well within a minute after starting, indicating the timeframe on which it operates.

Until now, researchers' main tools to study waking were drugs or

electrodes, neither of which offered the combination of speed and resolution of the new technique. Since mice typically only sleep for several minutes at a time and drugs generally require more time than that to take effect, manipulation of sleep circuits with drugs is usually inconclusive. The light pulse, or "optogenetic" technique, was uniquely valuable in answering questions about hypocretin and waking because it induces the desired effect almost immediately, de Lecea said.

In addition, the technique targets only the cells of interest, while implanted electrodes would gather information from an indistinguishable jumble of different nearby neurons. In general, within the hypothalamus, different cell types are too intermingled for researchers to easily tease out specific cell activity.

The optogenetic technique allowed for the most natural way available to simulate the brain's own release of hypocretin, Adamantidis said.

"We can really mimic the physiological conditions of hypocretin activation, which was impossible before," he said.

The team plans to continue collaborating with and helping other groups to apply optogenetic methods to psychiatric and neurological questions, Deisseroth said.

"This study shows that these fast optical tools are living up to their initial promise of helping us understand the workings of neural circuits that control behavior," he said, "and now there are many exciting experiments to be done."

Source: Stanford University