The brain consists of many types of neurons with distinct molecular organizations. A neuron’s functional roles in the information processing have been revealed by investigating the relationship between the artificial stimulation of the neuron and its outcome. Optical stimulation methods have received much attention recently with the technological development of modern optics. They have advantages over conventional electrical stimulation methods: precise spatiotemporal resolution and parallel stimulation-recording at multiple sites. These methods are also less invasive and more convenient than electrical stimulation methods. Another breakthrough combined optical stimulation with genetic engineering technologies, which is otherwise known as optogenetics. Light-sensing proteins of various living organisms are now available to be exogenously expressed in neurons and other target cells both in vivo and in vitro. Cellular functions such as the membrane potential, can thus be manipulated or probed by light. Optogenetics is borderless and interacting with a variety of fields. In 2010, optogenetics was chosen as the Method of the Year (MOTY) across all fields of science and engineering by the interdisciplinary research journal Nature Methods. The light, which has been employed only for the visualization of morphology and some functions of the cell, now becomes a tool to manipulate the cell or even the behavior. However, the genuine impact of optogenetics should be that it would integrate the biological systems, such as brain, and the optical systems, such as optoelectronics devices, using light as a mediator.
Is memory naturally disappeared with time? Here we found that melanin-concentrating hormone (MCH) neurons in the hypothalamus inhibiting or erasing memory during sleep. We generated transgenic mice in which MCH neurons express channelrhodopsin2 (ChR2) and activation of these neurons using optogenetics increased time in REM sleep. MCH neurons project throughout the brain and densely innervate hippocampus. This suggests that MCH neurons have a role in the regulation of both sleep/wakefulness and memory. To reveal physiological role, MCH neurons ablated mice were subjected to memory test. Interestingly, recognition, fear and spatial learning memory of these mice were significantly increased. Sleep deprivation failed to increase memory suggests the role of MCH neurons on memory during sleep. Conversely, activation of MCH neurons using DREADD or optogenetics inhibited formation of recognition and fear memory. To reveal this mechanism, retrograde tracer was injected into the hippocampus and revealed that MCH neurons were major neurons projecting from hypothalamus to hippocampus. Previous report showed that MCH neurons in the hypothalamus firing during non-REM sleep and REM sleep. Taken together, the activity of MCH neurons works to inhibit memory or erasing memory during sleep.

The mammalian brain consists of a complex ensemble of neurons and glial cells. Their production during development and remodeling is tightly controlled by various regulatory mechanisms in neural stem cells. Among such regulations, basic helix-loop-helix (bHLH) factors have key functions in the self-renewal, multipotency, and fate determination of neural stem cells. Here, we highlight the importance of the expression dynamics of bHLH factors in these processes. We propose the multipotent state correlates with oscillatory expression of several bHLH factors, whereas the differentiated state correlates with sustained expression of a single bHLH factor. We also developed a new optogenetic method that can manipulate gene expressions in neural stem cells by light. We used this technology to manipulate the growth and fate-determination of neural stem cells. I also introduce various applications of light-induced control of gene expressions in broad fields of biology.
Oculomotor control by pathway-selective optogenetic manipulation in primates

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Optogenetics enables temporally and spatially precise control of neuronal activity in vivo. One of the key advantages of optogenetics is that it can be used to control the activity of targeted neural pathways that connect specific brain regions. While such pathway-selective optogenetic control is a popular tool in rodents, attempts at modulating behavior using pathway-selective optogenetics have not yet been successful in primates. Here we develop a methodology for pathway-selective optogenetics in macaque monkeys, focusing on the pathway from the frontal eye field (FEF) to the superior colliculus (SC), part of the complex oculomotor network. We find that the optogenetic stimulation of FEF projections to the SC modulates SC neuron activity and is sufficient to evoke saccadic eye movements toward the response field corresponding to the stimulation site. Thus, our results demonstrate the feasibility of using pathway-selective optogenetics to elucidate neural network function in primates.

Optical control of the genome

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The genome consists of more than 20,000 genes and is essential for most of biological phenomena. To understand these biological phenomena, including diseases, and to utilize or modify them, approaches that enable the genome to be regulated at will are required. We developed a light-inducible, RNA-guided programmable system for endogenous gene activation based on the CRISPR-Cas9 system. We demonstrated that this optogenetic system allows rapid and reversible targeted gene activation by light. Using this system, we exemplified photoactivation of multiple user-defined genes in mammalian cells. The CRISPR-Cas9-based, photoactivatable transcription system (CPTS) offers a simple and versatile approach to precise gene activation. Additionally, we also developed another optogenetic system, named photoactivatable Cas9 (paCas9). We divided the Cas9 nuclease into two fragments and connected photo-inducible dimerization proteins to the fragments, leading to the development of paCas9 of which nuclease activity is switchable with light. Different from CPTS, paCas9 allows direct editing of DNA sequence of the genome by light stimulation. Genome editing technology and optogenetics technology have emerged as different technologies from each other so far. Our studies described above merge these emerging research fields together.