OPTOGENETICS

Optogenetics [2] is the field of controlling precise events in the individual cells of tissues using a combination of optics and genetics. These changes in cellular events are done with enough temporal precision to synchronize with biological processes occurring in the body of the target organism and the methods to observe and read the effects of optogenetic control on the target organism are appropriately precise and fast as well. The timescale in which optogenetic control is done generally falls in the millisecond range.

History

The idea of optogenetics is not that old, it was first proposed by Francis Crick [3] in 1999. Richard Fork and Rafael Yuste did the first demonstration of optogenetic control (without the genetic component) by activating neurons in living tissue using lasers. This work was expanded upon by the induction of genetic components in 2002 when Boris Zemelman and Gero Miesenbock [5] used Drosophila rhodopsin photoreceptors to be activated by laser light to control the neural activity of mammalian neurons. In 2004 the two groups of Kramer and Isacoff [6,7] had started developing organic photoswitches which could interact with genetically introduced ion channels. By 2005 [4] Susanna Lima and Miesenbock had performed the first demonstration of control of the behaviour of an animal by such genetically targeted photosimulation by genetically circumscribing the neurons of the dopaminergic system of drosophilae and then using photosimulation to elicit noticeable behavioural changes in them. Also, in 2005 [2], the first published work related to a single component optogenetic system was published by Karl Deisseroth's laboratory the Bioengineering department of Stanford. This optogenetic system used channelrhodopsin (ChR), which is a light activated single component positive electrical charge channel obtained from unicellular algae, to control behaviour in cultured mammalian neurons. This channel allows millisecond precision control in mammals and only one gene expression is required for it to work. It works well not just with mammals but also with other model organisms like flies, worms, and zebrafish. Many other "microbial opsins" have been developed and these have a lot of experimental utility.

Control

In order to study the causal roles of action potential patterns of specific neurons in the target organism, and other such fast biological information processing, the experimenter needs millisecond scale precision in optogenetics. To obtain similarly fast readouts either electrical recordings can be taken, which are called optrodes, or various reporter proteins which are biosensors may be employed, like voltage sensitive fluorescent proteins. Before optogenetics were developed traditional genetic manipulations to study the causal roles of various genes took a long time, from hours to months. Like the microbial opsin mentioned before many such microbial opsins have been developed which are used to cause reversible excitation in the neural systems, a few such opsins are ChR1, ChR2, VChR1, SFO etc. Just like neural activity may be enhanced, it can also be suppressed temporarily and reversibly. This is done using Halorhodopsins (NpHR) , which are similar to microbial opsins as in, they are also light driven ion pumps. Aside from Halorhodopsins, Leptosphaeria maculans fungal opsins (Mac), and enhanced bacteriorhodopsin (eBR) have also been used. These light induced inhibitors have been shown to work even in freely moving mammals [8]. Nowadays, not only targeting of specific neurons, but actual precise control of well-defined biochemical events in mammals is possible even when the mammal is moving and behaviour is taking place. A family of single component optogenetic tools have been built which allows control of concentration of intracellular messengers between cells. Similarly, control of GTPases and adenylyl cyclases has been achieved. Using these we can have control which is specific to cell type and temporarily precise of multiple cellular functions in moving animals.

Delivery of Control

Optogenetics require that these light sensitive ion pumps be properly delivered to their target neurons in the brains of the living organism which is the target of the study in order to do the above processes. To do this various genetic targeting strategies have been developed like customized viruses, cell-specific promoters etc. A more versatile approach is using transgenes to genetically transmit into targeted cells and then use cre-dependent viruses to target only these cells. The customized viruses use cre recombinase enzymes. Optogenetics also require the electrical hardware to control these triggers once the triggers have been deployed to their target cells. This is achieved using fiberoptic coupled diode technology [9]. These optical fibres can be directly mounted on the skull of the target animal. They can also be embedded deeper inside the brain for better control. These optical fibre devices were first used in conjunction with optogenetics to address issues related to sleep wake transitions and narcolepsy.

Applications

Optogenetics has given highly precise insights into the working of neural circuitry. It has thus provided “in vivo” understanding of biological processes. For example pyramidal neurons of the prefontal cortex [10] have been studied both “in vivo” and “in vitro” by optogenetic methods. Similarly cholinergic interneurons [8] of the nucleus accumbens have been subjected to both optogenetic activation as well as optogenetic inhibition inside freely moving animals. Although these cholinergic cells are numerically only 1% of neurons they have strong controlling behaviour on neural circuit activity. Thus the information yielded by studying them is important. Optogenetic research has yielded information [1] related to Parkinson’s disease, autism, schizophrenia, anxiety, depression and other such psychiatric disorders. Optogenetic research in mice has provided some understanding of neurobiologic underpinnings of aggression and violence. Autism is particularly a difficult field to research and the changes in physiology have been difficult to test in a temporally precise and a causal manner which is now possible to test with optogenetics. Memory deficits in cognitive impairment, dementia, and post-traumatic stress disorder have been better understood by using optogenetic techniques in the hippocampus and the neocortex. “Fear memory” [1] formation and expression have been extensively studied in amygdala, hippocampus, and the neocortex of freely moving animals using optogenetic techniques. Optogenetic research is also useful to understand drug abuse as cholinergic cells mentioned above get activated by cocaine and thus optogenetic study is used to understand drug reward mechanisms. A side effect of optogenetic study has been preservation of ecology since a lot of microbial organisms used in such study are rare and live in specialized environments. Another side effect has been research in pure science. This is because a lot of these opsins were studied long before their practical use in optogenetics was considered. Thus to summarize optogenetics are a novel and a very powerful tool to understand causal behaviour of neural circuitry and other biological mechanisms in a temporally precise manner which has led to a lot of advancement of psychiatry and other avenues of investigation. Causal relationships between action potentials and real time behaviour of moving animals have been not just studied but controlled. Optogenetics should advance our knowledge of not just psychiatry but the causal properties of the neural system in a significant manner.

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