Application Note

Ion Channels:
ChR2

Tools:
SyncroPatch 384
Optogenetic Stimulation Tool

SyncroPatch 384 optogenetic stimulation and light-gated ion channels

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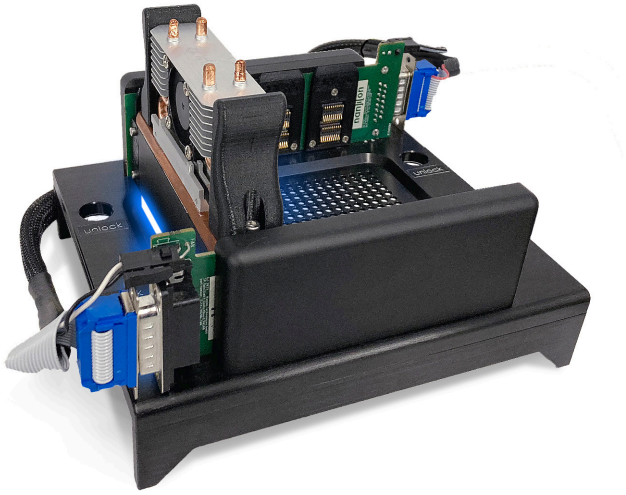
Summary

Channelrhodopsins (ChRs) are proteins that guide phototaxis in protists and exhibit light-gated channel conductance when their genes are heterologously expressed in cell lines such as HEK293 cells. ChRs are widely used optogenetic tools (e.g. Nagel *et al.* 2003), and are the prerequisite to a biological technique to control the activity of neurons or other cell types with light. For example, cation- and anion-selective ChRs enable stimulation and inhibition of neuronal action potential firing by depolarization and hyperpolarization of the membrane, respectively. More than 400 natural ChR variants have been identified so far, and sequencing projects add more each year (e.g. Govorunova *et al.*, 2022).

Light type	Dominant or Peak wavelength (nm)	
	Minimum	Maximum
UV	380	390
Visible	-	-
Royal Blue	440	460
Blue	460	480
Cyan	490	510
Green	520	540
Amber	588	592
Red-Orange	610	620
Red	620	645
Deep Red	650	670

The SyncroPatch 384 enables the control of membrane voltage by both voltage-clamp and optogenetics using the unique *Optogenetic Stimulation Tool*. The tool includes 6 LED-modules, each equipped with 96 LEDs.



The picture above shows one LED module mounted on the SyncroPatch 384 electrode board.

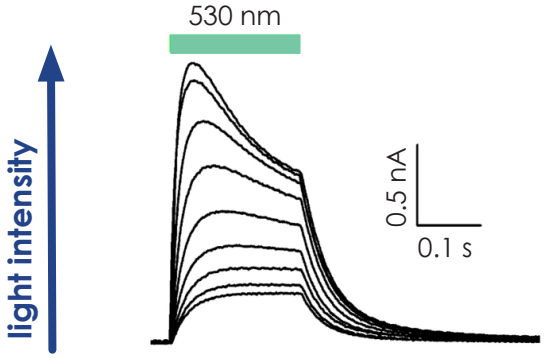


Figure 1: *A series of photocurrent traces generated by the ChR „HcKCR1" at 20 mV in response to 200 ms light pulses of incremental intensity (SyncroPatch 384 Optogenetic Stimulation Tool). The bar on top shows the duration of illumination. Modified from Govorunova et al., 2022.*

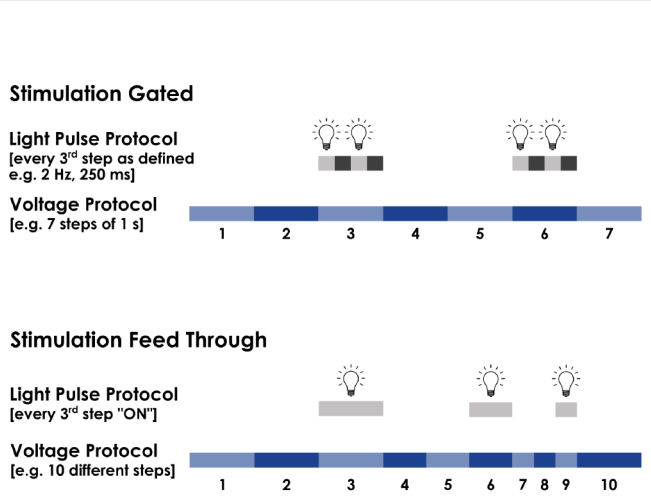


Figure 2: *Modes of operation. Stimulation Gated: Starts a previously defined light pulse protocol for the length of the 3rd, 6th, 9th... step of the voltage/current clamp protocol (except for the last step). Stimulation Feed Through: LEDs switch on at every 3rd, 6th, 9th... step (except for the last step). LEDs switch off at every other step and by the end of the protocol. Light intensity can also be defined.*

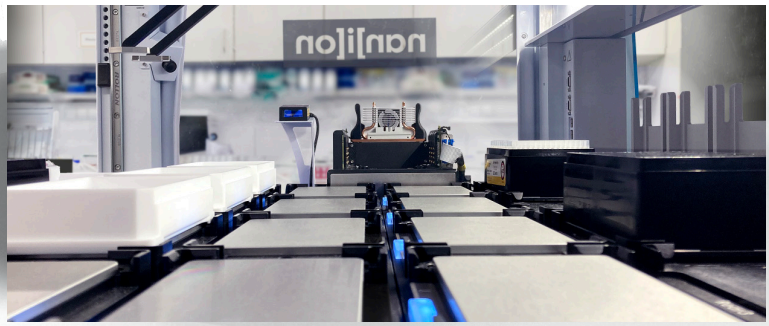
The LED control settings allow different modes of operation. Besides „On" (Switches the LEDs permanently on), „Off" (Switches the LEDs off) and „Stimulation" (starts a defined light pulse protocol) there are further more sophisticated modes available:

Stimulation Gated: Starts a previously defined light pulse protocol for the length of the 3rd, 6th, 9th... step of the voltage/current clamp protocol (except for the last step).

Stimulation Feed Through: The length of light pulses are defined by the voltage pulse protocol step. LEDs switch on at every 3rd, 6th, 9th... step (except for the last step). LEDs switch off at every other step and by the end of the protocol. Light intensity is defined by the command LED Current Settings.

References

1. Nagel, G., Szellas, T., Huhn, W., Kateriya, S., Adeishvili, N., Berthold, P., Ollig, D., Hegemann, P., Bamberg, E. (2003). Channelrhodopsin-2, a directly light-gated cation-selective membrane channel. PNAS. 100 (24): 13940 - 13945.
2. Govorunova, E.G., Sineshchekov, O.A., Brown, L.S., Spudich, J.L. 2022. Biophysical characterization of light-gated ion channels using planar automated patch clamp. Front. Mol. Neurosci., 15: :976910. https://doi.org/10.3389/fnmol.2022.976910



Key findings

1. The SyncroPatch 384 Optogenetic Stimulation Tool enables **optical modulation of cells and compounds using diverse wavelengths**, in up to 96 wells in parallel.
2. Optical Stimulation Tool enables **high temporal resolution**: fast and reversible On/Off switching of e.g. ChR2.
3. Multiple modes of operation ensure **sophisticated combination with electrophysiological stimuli** and the use of diverse experimental protocols.

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