University of Colorado Anschutz Medical Campus

Optogenetics and Neural Engineering

A proposed methods section that could help ensure that a researcher is thinking about their optogenetic stimulation parameters in adequate depth:

Optogenetic light excitation was achieved with a ***wavelength*** ***laser company (ex: SLOC)*** laser delivering collimated light through a custom made ***core diameter***, ***numerical aperture*** patch cord with a ***flat/etched*** face. Optic power at the tip of the fiber was measured pre-experimentation by collecting the entire beam within a ***light power meter (ex: Thorlabs S121C connected to a PM100D)*** with the laser continuously on and found to be ***number*** mW. Assuming no to little loss of optical power in air, and with an illumination area of ***area mm^2*** at the tissue site, the tissue was excited with an irradiance of ***number*** mW/mm^2. Stimulation was pulsed at * **parameters (ex: 1 ms 20 Hz pulses)*** using a TTL signal generated by ***generator number and manufacturer***. Rise and fall times of the pulsed laser resulted in an average irradiance of ***number*** mW/mm^2 across a single pulse, as measured with ***fast detector (ex: Thorlabs DET10A)*** connected to an ***oscilloscope (ex: Tektronix TDS 2024C)***. [1]

[1] **ON/OFF Considerations**: No system is instantaneous. The inevitable delay between a signal to change the status of a system (input) and the actual change (output) can be reasonably negated, but should still be acknowledged. Two examples:

1. An LED was pulsed on for 10 ms. It was noted that there is a ramp on time taking 1 ms, and a ramp down time of 0.5 ms. This can be estimated as a loss of 0.5 ms to the overall desired pulse. That represents (0.5/10) or 5%. It would therefore be reasonable to replace the last sentence with:

    a. Pulsing the LED resulted in ON/OFF rise/fall times, as measured with ***fast detector (ex: Thorlabs DET10A)*** connected to an ***oscilloscope (ex: Tektronix TDS 2024C),** of about 5% less than the power as measured continuous on and can therefore be negated.

2. A Laser was pulsed at 5 ms. It was noted that the ramp on time is near instantaneous, but *overshoots* the continuous on values by about 50% and then decays down to the continuous value over about 2.5 ms. When the TTL pulse ends, the laser ramped down in near instantaneous. Therefore, when this laser is pulsed, it delivers more optic power than when continuously on. Assuming linear decay, it can be estimated to be 25% more power over this pulse. You may account for this by either turning down the laser (suggested) or adjusting the TTL pulse duration. Use the last sentence of the paragraph above to describe this system.

Please acknowledge our facility in your publications. An appropriate wording would be:
"Engineering support was provided by the Optogenetics and Neural Engineering Core at the University of Colorado School of Medicine, funded in part by the *National Institute for Neurological Disorders and Stroke* of the National Institutes of Health under award number P30NS048154. The content is solely the responsibility of the authors and does not necessarily represent the official views of the National Institutes of Health."