

WEBINAR Q&A

Regarding success rates of the experimental runs, did you look at the reproducibility for the different assays?

Ciria Hernandez: The success rate of the recordings, among the different types of cells, was between 80 and 85%, depending on the experimental conditions, the reproducibility was acceptable.

Regarding sweep intervals, which sweep interval did you use in your CaV1.2 voltage protocol?

Yuri Kuryshev: CaV1.2, usually we are using a 10 second interval but sometimes we are also using a 5 second interval. 10 seconds is our standard case and 5 second is used less.

Also, regarding your success rates for your experimental runs, and the reproducibility for your assays?

YK: Success rates are very dependent on the cell lines, and the channels with hERG and other potassium channels as well as sodium channels like 1.5, success rate is close to 90% but in best cases it's 95%. Normally, to consider an experiment successful we have a cut-off at 80%, so below 80% we consider the experiment to be a failure. For calcium channels, it is a little bit different. First of all, it's dependent on cell quality, some cases we have the success rate again as 80% but in many other cases, for instance with CiPA protocol to measure Calcium current, we need to increase the number of applications to at least 8 instead of 4. This with calcium channels, CiPA protocol then has a 25% success rate, it's not bad. On good days, the success rates are well above 50% for calcium channels. For sodium, 80% is minimal and 90% is standard.

Regarding your experiments that show direct coupling of the MC4R to Kir7.1, what are the physiological implications of your assay in terms of feeding behaviors and energy expenditure?

CH: We still don't fully understand the implications of the direct MC4R-Kir7.1 coupling in a live organism. What we do know is that both are present or are expressed where they need to be, which is the MC4R neurons of the paraventricular nucleus of the hypothalamus (PVN). I personally think that the direct coupling could represent an escape mechanism to control, to some extent, the function (excitability) of these neurons under certain circumstances where normal cAMP-induced responses are shunted. For example, in POMC deficiencies. The fact that these are two independent modular functions, is a testament of this, and one can envision, manipulating one pathway pharmacologically without affecting the other by biased drugs.