Project 9.3 Experimental analysis of molecular determinants involved in epilepsy (NCN/OPUS)

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WWW: https://bit.ly/2SVfXgA

Background:

The effects of KCNB1 mutations that cause epileptic encephalopathy were analyzed mechanistically mainly using electrophysiology in heterologous cells in vitro. The developmental analysis was limited by the availability of single mutants in mice and zebrafish and did not explore the whole range of effects caused by KCNB1 mutations (Shen et al., 2016). The KCNB1 loss of function (LOF) or gain of function (GOF) cause specific morphological changes in the brain ventricles (Shen et al., 2016) and inner ear (Jedrychowska et al., 2020) in developing zebrafish embryos and larvae. The zebrafish transgenics express fluorescent markers in specific manner. The high-resolution microscopy of transgenic embryos and larvae in vivo provides information about developmental mechanisms as well as changes in activity of specific signaling pathways. These tools satisfy conditions necessary to study the developmental effect of different KCNB1 mutations in real time. This rationale was confirmed in preliminary experiments when analyzing the effect of overexpression of human mutated KCNB1 mRNA. Kcnb1 GOF causes cell delamination in brain ventricles and their expansion (hydrocephalus), and enlargement of the inner ear and otoliths. These features recapitulate the phenotype of the *kcnb1* GOF zebrafish mutant and constitute the rationale for the "brain and ear" *in vivo* test to be used when defining an effect of human mutations.

Aim:

Using the site-specific CRISPR-Cas9 mutagenesis in zebrafish, the representative Kcnb1 mutations that mimic the known human mutations of KCNB1 will be generated and analyzed by a combination of bioimaging, single-cell transcriptomics, electrophysiology and behavioral analysis. This will provide rationale for subfunctionalization of human KCNB1 mutations.

Requirements:

- Master's degree in biology, biochemistry or related field
- Eligibility for PhD studies in Poland
- Prior experience in molecular developmental biology and zebrafish studies is a bonus during selection of candidates, but necessary training will be provided
- Written and spoken fluency in English
- Willingness to learn and take new challenges, ability to work independently, analytical thinking
- Good interpersonal skills and a collaborative attitude

Number of positions available: 2

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