**Investigating a stop-gain variant associate with cell differentiation and synaptic machinery in focal cortical dysplasia (FCD)**

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**Introduction:** FCD is a malformation of the cerebral cortex usually associated with cell abnormalities, giant/dysmorphic neurons and balloon cells and severe drug-resistant epilepsy. The mechanisms involved in the pathogenesis of type II FCD are not completely understood. We have previously identified a rare stop-gain variant, rs76555439, located in in C-terminal binding protein 2 (*CTBP2)* gene in a few patients with FCD. This variant is absent in our in-house control database of 29 samples as well as in two independents control databases: Exome Variant Server and Exome Aggregation Consortium (ExAC). Furthermore, *CTBP2* gene has been related to: i) transcriptional corepressors that associate with DNA-binding transcription factors and have been linked to the regulation of the transition of neural precursor cells to a differentiated state1and ii) specialized synapses known as synaptic ribbons2. In this context, the goal of this work is to investigate the frequency of this *CTBP2* variant in an additional cohort of FCD patients and healthy control individuals in order to assess a possible genetic association.

**Materials and Methods:** We are performing a validation study using Sanger sequencing in an additional cohort 17 type II FCD patients and 315 control individuals. All donors provided written informed consent prior to enter the study. Genomic DNA was isolated from peripheral leukocytes by the phenol extraction method. PCR primers targeting the variant region were designed using Primer3 on genomic DNA sequence. 50ng of genomic DNA for each sample was amplified and primer pair used for amplification was as follows: forward primer, 5′-GAAGACAGAAAAAGTTGCCATCCACACC-3′, and reverse primer, 5′-TCATCATCAGCAGACATAAGCAAGGCAGA-3´. PCR touchdown was carried out using a thermocycler for 35 cycles with annealing temperature of 58.7°C. To confirm the specificity of amplification, the PCR products were subjected to electrophoresis in 1% agarose gel. Sanger sequenced will be done by capillary electrophoresis on ABI 3500XL (Applied Biosystems).

**Results/Discussion and conclusion:** To date, we initiate the PCR standardization and our preliminary results indicatethat this*CTBP2* variant could have a relevant role in molecular mechanisms involved in this cortical malformation.

**References:** [1] Kim TW et al., Stem Cells 33: 2442-2455, 2015 [2] Rutherford MA, Synapse:69 (5):242-55, 2015