**Advanced proteomic study of the dentate gyrus in an epilepsy model presenting hippocampal sclerosis**

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**Introduction:** Mesial Temporal Lobe Epilepsy (MTLE) is the most common type of severe epilepsy in adults and it is characterized by histopathological abnormalities in the mesial temporal lobe structures, such as hipocampal sclerosis (HS). The dentate gyrus (DG) is a cortical region that is an integral portion of the larger functional brain system called hippocampal formation. Thus, there are numerous features of the DG that makes it unique in a neuroanatomical and functional way. Therefore we propose to study the DG in its both layers, molecular (ML) and granule (GL), obtained from a rat epilepsy model induced by the perforant pathway stimulation and displaying the classical features of HS.

**Materials and Methods:** Rats were induced as described by Norwood et al., 2010. Frozen sections were prepared and the DG was further divided into dorsal (dDG) and ventral portions (vDG) in the epilepsy model (n=3) as well as control animals (n=4). dDG (ML and GL) and vDG (ML and GL) were laser microdissected (Zeiss PALM). Total proteins were obtained using 8M urea extraction and analyzed by LC-MS/MS in an LQT-Orbitrap (Waters). Bioinformatics analysis was performed using MaxQuant and Perseus software and biological correlates were obtained using Metacore.

**Results:** We identified a total of 1115 proteins in the dDG-GL of which 69 were differentially expressed in the epilepsy model. We found 465 proteins in the dDG-ML of which 22 were differentially expressed. For the vDG, we identified 1083 protein in the GL of which 55 were differentially expressed and 583 in the ML of which 33 were differentially expressed. Interestingly, we found that enriched biological pathways were different between the GL and the ML in both regions (dorsal and ventral) of the DG. Moreover the dDG and vDG showed distinct proteomic profiles (figures 1 and 2).

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Figure 2. Venn diagram showing the number of proteins Differentially expressed in the epilepsy model in the vDG-GL and in the vDG-ML.

Figure 1. Venn diagram showing the number of proteins differentially expressed in the epilepsy model in the dDG-GL and in the dDG-ML.

**Discussion:** GL is composed mainly by cell bodies; whereas, the ML presents predominantly dendrites. Therefore, it is expected that these two regions have differences in protein composition. In addition, there is evidence that the two regions of the DG, dDG and vDG, are functionally and molecularly distinct. Thus, our study was designed to analysis these regions separately. Our results show that the mainly enriched pathway found in the dDG-GL are involved with molecules transportation and neuronal development, while in the dDG-ML are involved with immune response and energy metabolism. In the vDG-GL the represented pathways found are GABA neurotransmission and pathways involving changes in *Cl*- homeostasis in neurons, while the vDG-ML showed alterations in AMPA receptors pathways and in vascular development.

**Conclusion:** We haveshown that there are remarkable differences in protein expression among different layers and sub-regions of the DG of an animal model of MTLE with HS. The identified proteins indicate new molecular mechanism potentially involved in epileptogenesis. These mechanisms can now be further explored and may eventually be target in development of new treatments to cure epilepsy.

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