Abstract ID : **58**

Use of an archaeal protein to study the structural implications of a mutation responsible for episodic ataxia 6

Content

Glutamate transporters (SLC1 family) have been extensively characterized in the past two decades for their crucial role in ensuring a proper signal transmission in multicellular eukaryotes. A large number of both biochemical and structural studies has validated the co-dependence of the transport of one molecule of amino acid and three Na+ ions by each of the three protomers that characterize their structure, as well as the presence of a chloride channel which is conserved in all domains of life that works in a thermodynamically uncoupled manner from the other substrates. The imbalance in the homeostasis finely regulated by these transporters is associated with several neurological diseases, among which Episodic Ataxia 6 (EA6) is provoked by the single point mutation from proline to arginine located in the transmembrane 5. Such mutation is responsible for a decrease in amino acid and Na+ transport, concurrent to an increase in chloride current. Here we present the characterization of the substrate transport dependence of the anion conductance on the archaeal homologue GltTk, using the Solid-supported membrane (SSM) – based electrophysiology. The comparison of the anionic currents between the wild type and its mutants suggests that the amplitude of negative current is strictly dependent upon the transport rate of the amino acid and the Na+ ions, apart from the P208R mutant in which the anionic current is consistently higher. Moreover, the structure of the P208R mutant in nanodiscs, obtained with Cryo-EM, shows that the substitution of the proline with an arginine creates a salt bridge with a phospholipid, probably responsible for the widening of the chloride channel. These results shed lights on the complicated framework of the dual role of Glutamate transporters and show the involvement of the phospholipids in the protein dynamics.

Primary author(s) : Mrs. COLUCCI, Emanuela (University of Groningen)

Co-author(s) : Prof. GUSKOV, Albert (University of Groningen); Prof. SLOTBOOM, Dirk-Jan (University of Groningen)

Presenter(s) : Mrs. COLUCCI, Emanuela (University of Groningen)

Contribution Type : Poster

Submitted by **COLUCCI, Emanuela** on **Tuesday 05 April 2022**