**Evaluation of the multiprotein complex formation between Na, K-ATPase and Glutamate Transporters (EAAT1 and EAAT2) through molecular docking**

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The formation of macromolecular complexes in the plasma membrane of cells encompasses a wide range of physiologically relevant functions across various cell tissues. These complexes are primarily stabilized by interactions between the protein subunits that constitute them, the presence of peptides, and certain lipids. These lipids either interact directly with the complex or create an appropriate membrane microenvironment. The experimental hypothesis proposes the formation of a complex between Na, K-ATPase (NKA) and excitatory amino acid transporters 1 and 2 (EAAT1/EAAT2), stabilized by the presence of FXYD peptides, in neuronal and glial cells. This complex would be involved in the reuptake of glutamate from the synaptic cleft, where deficiencies in this process lead to glutamate excitotoxicity, resulting in neuronal death. Currently, there are no bioinformatics studies that challenge the experimental data on the formation of this complex, which are capable of providing a significant amount of structural data. Among the tools of structural bioinformatics, macromolecular docking, with steps for optimizing the generated complexes in solvent or explicit membrane, combined with binding energy calculation algorithms, provides data describing the affinity of protein-protein interaction through ΔG values and dissociation constant. Using this methodology, it was possible to assemble the system with the α1 and α2 subunits of NKA and the EAAT1/EAAT2 transporters, verify the conformation adopted between the proteins, identify the amino acids at the interaction interface, determine the protein-protein interaction affinity values, and understand how the FXYD peptides influence these affinity values. The most stable complex occurred between the α1 subunit and the EAAT1 transporter in the presence of FXYD2 peptide. The findings indicate an increase in stability in the presence of FXYD2 and FXYD7, suggesting a possible role of these peptides in enhancing glutamate reuptake in the synaptic cleft by increasing the association of NKA and EAAT1/EAAT2 proteins.

Keywords: Macromolecular Docking; Bioinformatics; Glutamate Transporter; Na, K-ATPase; FXYD