ポスター

[2P]A. 分子·細胞生物学

2022年7月1日(金) 11:00 ~ 12:00 ポスター会場2 (宜野湾市民体育館)

[2P-022]電位依存性ナトリウムチャネル Nav1.2結合タンパク質の解析

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Voltage-gated sodium channels play essential roles in the generation and propagation of action potentials in neurons. These channels consist of a pore-forming alpha subunit and one or more auxiliary beta subunits. Nav1.2 is an alpha subunit widely expressed in the central nervous system (CNS). It has been known that Nav1.2 is preferentially expressed in unmyelinated axons of the CNS, such as parallel fibers of cerebellum, mossy fibers of hippocampus and striatonigral fibers. Nav1.2 is diffusely distributed along these axons. This singular Nav1.2 distribution in the unmyelinated axons gives rise to a speculation that Nav1.2 could have another role in the unmyelinated axons. To investigate about this, we examined Nav1.2 interacting proteins in the myelinated axons and unmyelinated axons. First, we generated polyclonal anti-Nav1.2 antibody, which can be used for immunoprecipitation (IP). Next, we fractionated striatonigral fiber enriched mouse brain homogenate to myelin basic protein (MBP) (+) and MBP (-) membrane fraction, containing myelinated and unmyelinated axon properties, respectively. To separate these two fractions, we performed sucrose gradient centrifugation (0.32M/0.85M) using membrane fraction of striatonigral fiber enriched mouse brain homogenate. To identify Nav1.2 interacting proteins, we performed IP of MBP (+) and MBP (-) membrane fractions using anti-Nav1.2 antibody and then IP-samples were analyzed by LC-MS/MS. Comparison of Nav1.2 interacting proteins between MBP (+) and MBP (-) membrane fractions showed many highly enriched proteins in each fraction.