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An Essential Domain in C-terminus Responsible for Golgi Localization and Recycling of Mouse Itm2C

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Itm2c is a member of the Itm2 gene family that is expressed in both embryonic and adult brains. We previously found that C-terminus truncated Itm2C showed a disrupted Golgi localization and a mislocalization to the plasma membrane. The present study was aimed to find a minimal or essential domain affecting the intracellular distribution of Itm2c in neurons. To construct mutant vectors carrying reporter gene, various deletion mutant of C-terminus-truncated Itm2C sequences were inserted into pcDNA3.1B or pEGFP-N1 vector, and their intracellular distributions were analyzed from the transient transfection into CHO and mouse N2a neuroblastoma cells. The minimally deleted mutant Itm2C (△266-269) showed neither Golgi localization nor exocytosis and instead distributed throughout the cytoplasm of the transfected cells. The amino acid (aa) sequence prediction showed that the region containing 266 to 269 aa is a putative prenyl group binding site. These results suggest that C-terminus region of Itm2C is an essential for cellular trafficking either as just a membrane component of recycling vesicles or associated components of other unknown molecules that are endocytosed. Presently, we are trying to elucidate the Itm2C domains specific for the respective trafficking and to clarify the putative prenylation sites using site-directed mutants.

Keywords · Itm2c, Exocytosis, Recycle, Deletion mutants