(P15) The mechanism which regulate isoform specific transport for synaptotagmin and the target molecules transported with synaptotagmin

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Inositol poly-phosphates (IPs) are considered to play crucial physiological functions in cells. However, the precise targets of IPs remained to be characterized. Therefore, to identify the molecular targets for IPs, our seniors had purified the IPs binding molecules and characterized synaptotagmin (Syt) I as the IP4, 5 and 6 binding molecules. Now, Syt family is composed of 15 members in mammal and plays important functions in several types of intracellular transport, endocytosis and exocytosis. However, the molecular mechanism which regulates the isoform specific targeting for synaptotagmin and the molecules which are transported with Syt-containing vesicles are almost unknown. Srg1 was first synaptotagmin isoform which was upregulated under the control of thyroid hormone (TH) signaling pathway specific in brain. To identify the regulatory molecules, I first performed yeast two-hybrid screening to isolate the interaction molecule for Srg1 and obtained HGF regulated tyrosine kinase substrate (Hrs) as one of the candidates. Hrs was expressed ubiquitously and was implicated in the decision for protein degradation or recycling on early endosome. Hrs can interact with Srg1 both *in vitro* and *in vivo*. This interaction was mediated via the unique linker region and C2A domain of Srg1, and the hinge and P/Q-rich regions of Hrs. The intracellular distribution of Srg1 in HeLa cells indicated that Srg1 might participate in the intracellular transport between TGN and endosome, in which Hrs resided on and functioned. Srg1 was most abundantly expressed in cerebellar granule cells and was considered to be significant in TH dependent cerebellum development. Therefore, finally the overexpression of Hrs deletion mutants with Srg1-GFP in mouse cerebellar granule cells decreased the number of Srg1-GFP labeled vesicles. On the other hand, the overexpression of Hrs deletion mutants with Syt I-GFP did not decrease that of Syt I-GFP vesicles. Thus, Hrs may be implicated in the regulation of stability of Srg1-labeled vesicles *in vivo*.