﻿\subsection{**Synaptojanins likely influence vesicle uncoating, but not scission dynamics.}**

In mammalian cells, disruption of Synaptojanin genes results in cellular accumulation of PIP2 at endocytic sites. Coated vesicles gather at the plasma membrane, suggesting a role for lipid hydrolysis in releasing coat proteins from nascent vesicles (ref?). As an alternate to forces from Dynamin constriction, Liu et al (refliu) have proposed that an interaction between PiP2-hyrdolyzing Synaptojanins and BAR proteins could drive membrane scission. In this model Rvs BAR domains would form a scaffold on the membrane tube, preventing hydrolysis of underlying PIP2. Synaptojanin would arrive at inavaginated membranes, and hydrolyse unprotected PIP2. This generates a lipid boundary between BAR-protected PIP2 at the tube and hydrolyzed PIP2 at the bud tip. A line tension thus formed at the interphase between the two lipid types would then generate enough force to pinch off a vesicle.

There are three Synaptojanin-like proteins in yeast- Inp51, Inp52 and Inp53. Inp51 exhibits a diffuse cytoplasmic signal, and Inp53 localizes to patches within the cytoplasm- cellular localization that is consistent with involvement in trans-Golgi signalling (refGolgi). Inp53 was not investigated further. Inp52 localizes to cortical actin patches that are endocytic sites. Two channel alignment shows that Inp52 patches arrive in the late scission stage, and localizes to the bud tip, consistent with a role in membrane scission.

Role of Inp51 and Inp52 are tested by following Sla1-GFP and Rvs167-GFP in cells with either Inp51, Inp52, or both deleted. Retraction events do not significantly increase compared to the WT in either inp51del or inp52del cells. Magnitude and speed of coat movement in inp51del is the same as the WT. In inp52del cells, coat movement also has the same magnitude and speed, but GFP signal is persistant after membrane scission. This delay in decrease of Sla1-GFP signal is consistent with delay in vesicle uncoating rather than membrane scissison. Similarly, Rvs167 disassembly has a delay, while the assembly is similar to WT, indicating a delay in removing endocytic proteins from the newly formed vesicle. Assembly of Rvs167 has a delay in inp51deleted cells, which could indicate a defect in recruiting proteins to endocytic sites, or in progression of endocytic invaginations. Since Sla1 movement is the same, we suggest a defect in the former rather than latter.