

Exit of different cargoes from the Golgi and their post-Golgi trafficking

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Proteins and lipids are synthesized in the ER, delivered at the Golgi complex, then exit the Golgi complex and finally, delivered toward their final destination. Until now the information about the Golgi-to-plasmalemma transport (GPT) and the formation of Golgi-to-PM carriers (GPC) and their fate is controversial. For a long time, the vesicular model played a role of paradigm for all steps of intracellular transport. However, several years ago it was demonstrated that the transport from the Golgi complex to the plasma membrane (PM) is carried out by irregular GPCs [1,2]. Pathways for different cargoes are not well established. It is not clear whether there is a necessity for the fusion of the post-GC carriers with endosomes. Here, we examined mechanisms of GPT, namely, the three-dimensional structure of post-Golgi carriers, patterns of their exit from the Golgi complex, their transformation during their delivery to the baso-lateral PM and their dependency on the amount of cargo transported and fusion with endosomes.

All reagents, the cells and the cargo synchronization protocols were described in [3]. We studied conventional cargoes: PCI, PCI-GFP (PFP), tsVSVG, tsVSVG-GFP (VFP); ASGPR, albumin, and GFP-albumin (AFP) using STEM and TEM tomography, correlative light electron microscopy (CLEM), high pressure freezing (HPF), immuno-EM, focused ion beam (FIB-SEM). The GPC exit from GC depends on the amount of cargo moving through it. The temperature-sensitive glycoprotein G of vesicular stomatitis virus (VSVG) and procollagen-I (PCI) can also exit from the last two medial Golgi cisternae when a large amount of them is transported, whereas when a small amount of PCI is transported, it passes through the last medial cisterna and the trans-most cisterna (TMC) and then exits the TGN zone. Albumin exits the GC through TMC and TGN, where accumulative large vacuoles are formed. Albumin is enriched in the vacuole(s). Most of these vacuoles contain low concentration of VLDL. Smaller but distinct vacuoles are enriched in VLDL. This vacuole could be connected with the PM through thin tubule. Fusion of GPCs with endosomes and then their subsequent fission is necessary to remove resident Golgi proteins from GPCs and replace SNAREs in GPCs. Using advanced methods of the high-resolution 3D imaging (STEM and TEM tomography, FIB-SEM) after a cryo-immobilization procedure (HPF) has been shown that near the GC and during their passage to the PM, GPCs are always connected with at least one GC/TGN/endosome structure. The exchange of SNARE proteins ensures the subsequent fusion of GPCs with the PM. Thus, the kiss-and-run model is the most powerful model for the explanation of GPT.

Keywords:

Golgi, post-Golgi, transport, 3DEM, tomography

Reference:

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