

UNIVERSITY OF GOTHENBURG

Vesicle Transport with Emphasis on Chloroplasts

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Akademisk avhandling för filosofie doktorsexamen i Naturvetenskap med inriktning biologi, som med tillstånd från Naturvetenskapliga fakulteten kommer att offentligt försvaras torsdag den 7 mars 2013 kl. 10:00 i Hörsalen, Institutionen för biologi och miljövetenskap, Carl Skottbergs Gata 22B.

Fakultetsopponent: Prof Patrick Moreau, Laboratoire de Biogenese Membranaire, University of Bordeaux, Frankrike

ISBN: 978-91-85529-52-0

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Abstract: The plants on which we depend for food and oxygen need photosynthesis to prepare their own food. Photosynthesis takes place in the chloroplast. Inside chloroplasts is a specialized membrane called the thylakoids in which the photosynthesis activity takes place. The thylakoid membrane does not produce its own lipids, so instead they are transported from the envelope membrane to the thylakoid. Similarly, most of the proteins needed for maintenance of the photosynthetic apparatus and thylakoids are imported into the chloroplast across the envelope membrane and transported to the thylakoid. The lipids produced in the envelope membrane were suggested to be transported in three ways; through diffusion, through contact between thylakoids and the envelope membrane are well separated from each other by an aqueous solution, the stroma, which makes it hard for the lipids to move between the two compartments. Biochemical and ultrastructure data show vesicle transport inside chloroplasts. One of the vesicle functions in chloroplasts is to transport lipids from the envelope to the thylakoid to maintain its membrane structure.

Proteins transported from the envelope to the thylakoids take four routes (Sec, Tat, SRP and Spontaneous pathways). Only a few proteins have been shown or hypothesized to follow these pathways. For many proteins it is unclear how they are transported to the thylakoids. It has been shown that vesicle transport in the chloroplast is similar to the cytosolic secretory system, which transports both lipids and proteins between different compartments in the cytosol. This hypothesis became more likely when putative protein components of the COPII transport pathway i.e. Sec23/Sec24, Sec13/Sec31 and Sar1 (which operate between the endoplasmic reticulum and the Golgi apparatus) were suggested to exist in chloroplasts.

This thesis reports that indeed vesicle transport inside the chloroplast is similar to that of the cytosolic secretory system. The Sar1 homologue CPSAR1 (CP = chloroplast localized) has been characterized and shown to be important for embryo development and thylakoid biogenesis. Other studies have already shown that proteins such as VIPP1, THF1, ADL and FZL in the chloroplast do have an impact on vesicle transport and are also involved in thylakoid maintenance and biogenesis. This gives an indication that CPSAR1 could be involved in vesicular transport as well as collaborating with these proteins. Indeed it has been shown that CPSAR1 may interact. CPSAR1 could be involved in several functions. Previous data shows its involvement in ribosome biogenesis, which is also indicated by genes co-expressed with CPSAR1 (on the publically available ATTED-II database) that have roles in protein synthesis.

If there is a functional vesicle transport system in chloroplasts we expect there to be more components that are similar to vesicle transport in the cytosol. A bioinformatics approach predicted components like tethering factors, SNAREs, Rab GTPase, etc., to be present in chloroplasts. It was also proposed that the transport of cargo proteins in vesicles from the envelope to thylakoids would occur in a similar way to the secretory system in cytosol.

One of the Rab GTPases, CPRabA5e has been found in the chloroplast and is localized in the stroma and thylakoids. It has been suggested that it binds to the thylakoid in its active form and has a role in vesicle tethering and fusion similarly to its homologue in yeast. Ultrastructure analysis of CPRabA5e mutant chloroplasts shows accumulation of vesicles at low temperature compared to wild type indicating a role in vesicle transport. Furthermore, CPRabA5e has been shown to have a role in seed germination, oxidative stress and maintaining the size of plastoglobuli.

There has been clear evidence of vesicle transport inside chloroplasts and this transport is related to the secretory system in the cytosol. Two proteins in the chloroplast similar to proteins found in the secretory system are CPSAR1 and CPRabA5e, whose roles have been further characterized in chloroplast vesicle transport. At the same time other predicted components need confirmation of their localization. Finally, the cargo protein transport using vesicles need experimental verification to fill the model of vesicle transport inside chloroplasts.

Keywords: cargo, chloroplast, CPRabA5e, CPSAR1, lipid, protein, transport, vesicle

Gothenburg, March 2013

ISBN: 978-91-85529-52-0