ABSTRACT

UNDERSTANDING THE ORIGIN AND FUNCTION OF ORGANELLAR METABOLITE TRANSPORT PROTEINS IN PHOTOSYNTHETIC EUKARYOTES: *GALDIERIA SULPHURARIA* AND *ARABIDOPSIS THALIANA* AS MODEL SYSTEMS

By

Marc Linka

Membrane-bound compartments, especially the organelles mitochondria and plastids, are a hallmark of eukaryotic cells. Organellar metabolite transport proteins facilitate the exchange of metabolites across membranes in a cell and are crucial for connecting biochemical pathways that operate in separate compartments. Of particular interest to plant scientists is the plastid organelle of photosynthetic eukaryotes. Plastids synthesize and deliver major biological molecule classes such as carbohydrates, fatty acids, amino acids, and nucleic acids to the rest of the cell and thus the plastid has to be extensively connected to the cytosol. Plastids originated from an endosymbiotic cyanobacteria-like ancestor about 1.6 billion years ago and in this thesis the differences between plastid metabolite transporters of the eukaryotic model plant *Arabidopsis thaliana* and prokaryotic cyanobacteria were investigated. A phylogenomic analysis of 83 predicted plastid metabolite transporters from *Arabidopsis thaliana* has been conducted in collaboration with Dr. Debashish Bhattacharya. These studies allowed the conclusion that the majority of the transport proteins in extant plastids are absent from free-living cyanobacteria and originated from eukaryotic host genes. They represent true innovations associated with organelle evolution. Transporters became likely targeted to the endosymbiont via the endoplasmic reticulum of the host early in its evolution. Furthermore, the results suggest that export of photosynthates from the plastid in form of

sugar-phosphates has been a selective advantage to set-up a permanent endosymbiosis between the host and the endosymbiont. While these sugar-phosphate transport proteins are conserved in all photosynthetic eukaryotes, their biochemical properties co-evolved to meet the specific metabolic requirements in the distinct groups of the eukaryotic kingdom. As reported in Chapter 3, further studies have shown that in contrast to higher plants, the red alga *Galdieria sulphuraria* has a high affinity export system for triose-phosphates and lacks hexose-phosphates transport across the plastid envelope membrane. This reflects an adaptation for an efficient export of photosynthates from the organelle due to an absence of a plastidic starch pool in red algae.

Metabolite carriers facilitate also the transport of compounds in a single, highly compartmentalized cellular pathway. A prime example is the photorespiratory pathway. 2-phosphoglycolate is produced by the oxygenase reaction of the enzyme ribulose-1,5-bisphosphate Carboxylase/Oxygenase (RubisCO) and subsequently recycled to 3-phosphoglycerate in the compartments chloroplast, cytosol, peroxisome, and mitochondrion. A reverse genetic approach was used to identify the transport proteins involved in photorespiration and 32 candidates have been designated. Five of these were genetically analyzed to test their role in the recycling of 2-phosphoglycolate, leading to the discovery of a novel transporter required for a functional photorespiratory pathway. This protein is localized to the inner envelope membrane of mitochondria and the transporter most likely imports a cofactor from the cytosol, which is required for the mitochondrial glycine decarboxylase enzyme complex.