

DISSERTATION SUMMARY

The role of phosphatidylglycerol in the photosynthetic electron transport processes

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Thylakoid membranes in chloroplasts and cyanobacterial cells are the sites of the primary processes of oxygenic photosynthesis. The lipid composition of thylakoid membranes is highly conserved and consists of mostly glycolipids and about 10% phosphatidylglycerol (PG) as the only phospholipid. PG is considered to play an important role in the ordered assembly and structural maintenance of the photosynthetic apparatus.

In order to investigate the function of PG in the photosynthetic electron transport processes we studied a *pgsA*⁻ mutant which was constructed from a cyanobacterial strain, *Synechocystis* PCC 6803. This mutant is defective in PG phosphate synthase gene, and it is incapable of synthesizing PG and requires PG supplementation for growth. The deprivation of PG from the growth medium decreased the PG content in the mutant cells and it blocked cell growth and led to 50% decrease of oxygen evolving activity on the chlorophyll basis (Hagio et al. 2000).

Artificial quinones which are known to function as efficient electron acceptors for PSII completely inhibited the oxygen evolution of the *pgsA* mutant cells after 5 days of PG deprivation.

Direct evidence for suppression of the electron transfer between Q_A and Q_B was obtained by the thermoluminescence and flash-induced fluorescence measurements of *pgsA* mutant cells (Gombos et al. 2002). The observed effects were reversed if PG was re-added to the cultural medium. These results suggest that PG acts and binds close to the Q_B-binding site of PSII.

Two PG molecules were found at the periphery and one more at the central core of PSI when the PSI reaction center structure resolved at 2,5 Å revealed by Jordan et al. (2001). It has been suggested that the depletion of PG from PSI complexes can cause changes in the function of PSI.

We cultured *pgsA* mutant cells in the PG depleted medium for weeks in order to follow depletion and degradation of PG from PSI complexes. Sixty percent decrease was observed in the activity of PSI by measuring flash-induced absorbance changes at 705 nm after 3 weeks of PG deprivation. At the same time chlorophyll content of the cells

decreased to 20% of the initial concentration. PSI activity and the normal chlorophyll content were fully recovered if the cells were grown in the PG-containing medium for a few days.

The observed changes were confirmed by 2-D spectrum/decay-imaging spectroscopy.

The measurement showed significant differences between *pgsA* mutant cells cultured in the presence or absence of PG. The most striking effect was the very low fluorescence at 730 nm in the cells grown in the absence of PG. A broad peak at 730 nm represents long-wavelength absorbing chlorophyll a band typical in PSI reaction center. It can be concluded that PG depleted *pgsA*⁻ cells cannot form proper PSI reaction center complex at all.

These results were also supported by Deriphath PAGE, a special native gel electrophoretic method (Peter and Thornber 1991). We showed that the amount of the aggregated form of PSI, which is the most efficient form in cyanobacterial energy transfer process, decreased after 3 weeks of PG deprivation. The loss of PSI trimers can be reversed by adding PG again.

We separated PSI monomers from PSI trimers on a preparative HPLC anion-exchange chromatography column. The separated forms were identified by measuring fluorescence emission and excitation spectra of fractions at 77 K, and the proteins were characterized by SDS PAGE.

On the basis of our experiments we concluded that PG is essential for promoting the formation of protein oligomers.

References

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