

チラコイド膜脂質の生理機能解析

Introduction

Digalactosyldiacylglycerol (DGDG) is a major membrane lipid of oxygen evolving photosynthetic organisms such as plant, alga, cyanobacteria, comprising about 30% of polar lipids in the thylakoid membrane (Joyard et al. 1998). DGDG is important for the assembly and function of photosynthetic complexes (Dörmann and Benning 2002). DGDG synthase genes have been identified in plants (Dörmann et al 1999). However, no homolog of plant-type DGDG synthase genes was found in the genomes of cyanobacteria or red alga. Recently, we identified that *slr1508* of *Synechocystis* sp. PCC6803 (designated *dgdA*) encodes a non-plant-type DGDG synthase. Here we report the growth profiles and lipid and protein compositions of $\Delta dgdA$ mutants, especially under phosphate limitation.

Conclusion

- Cyanobacteria utilize non-plant type DGDG synthase for DGDG synthesis.
- The $\Delta dgdA$ mutant grew obviously slow under phosphate deprivation.

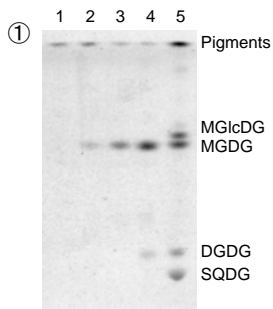


Figure 1. Accumulation of DGDG in *E. coli* by co-expressing cucumber MGDG synthase (csMGD1) and *slr1508*.

1. *E. coli* with empty vector for csMGD1
 2. *E. coli* with csMGD1
 3. *E. coli* with csMGD1 and pQE31
 4. *E. coli* with csMGD1 and *slr1508*
 5. *Synechocystis*
- E. coli* cells with solely expressed *slr1508* do not accumulate DGDG (data not shown).

Cyanobacterial DGDG synthase gene was identified as a non-plant type DGDG synthase. DGDG only accumulated by co-expressing cucumber MGDG synthase and *slr1508*(*dgdA*).

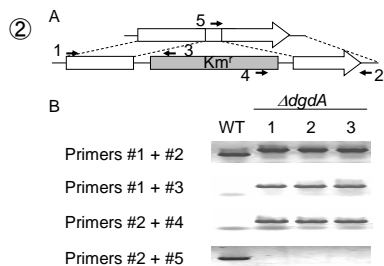


Figure 2. A. Schematic representation of *slr1508* in the wild type and in the mutant. Km: kanamycin resistance gene. B. Genotyping of the *slr1508* mutant. WT, the wild type; Mut, the *slr1508* mutant.

A null mutant of *dgdA* was obtained.

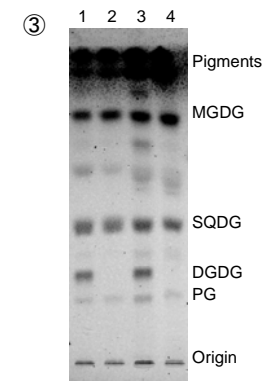
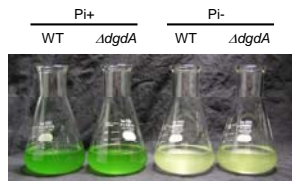


Figure 3. Lipid analysis of WT and the $\Delta dgdA$ mutant in optimal and phosphate deprived conditions.

1. WT in optimal medium
2. $\Delta dgdA$ in optimal medium
3. WT in phosphate deprived medium
4. $\Delta dgdA$ in phosphate deprived medium

No detectable DGDG was found in the mutant by lipid analysis.

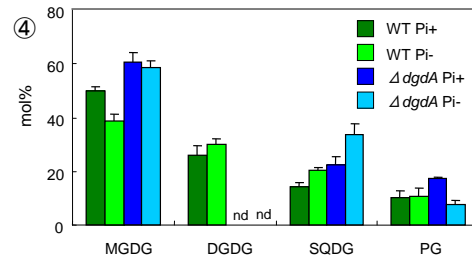


Figure 4. Lipid compositions of the WT and the $\Delta dgdA$ mutant in optimal and phosphate deprived condition. Each value is represented as mol%. SD is based on three independent experiments. Pi+, optimal condition; Pi-, phosphate deprived condition; nd, not detected (< 0.1 mol%).

In Pi- condition, content of SQDG and DGDG in WT increased with concomitant decrease of MGDG. In optimal growth condition, the *dgdA* mutant had no detectable amount of DGDG. Instead, content of the other lipids increased. In Pi- condition, content of SQDG increased to 33.6 % with decrease of PG.

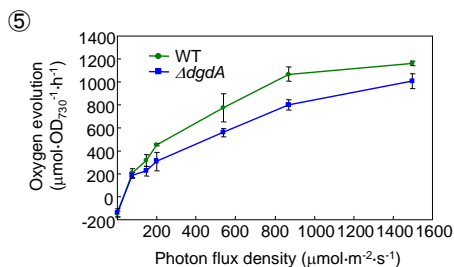


Figure 5. Oxygen evolution rates based on optical density of the culture. Filled green circle indicates the WT and filled blue square the $\Delta dgdA$ mutant. SD is based on three independent experiments.

Oxygen evolution rate of the mutant was slightly decreased compared to the wild type.

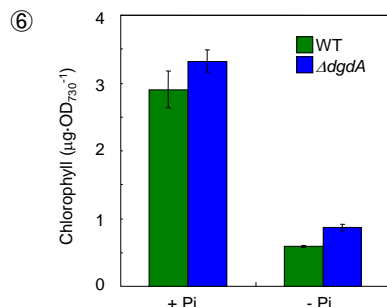


Figure 6. Chlorophyll contents of the wild type and the $\Delta dgdA$ mutant grown in optimal (Pi+) and phosphate deprived (Pi-) medium. The values are based on optical density of the culture. SD is based on three independent experiments.

The chlorophyll content was slightly higher in the mutant in both Pi+ and Pi- conditions.

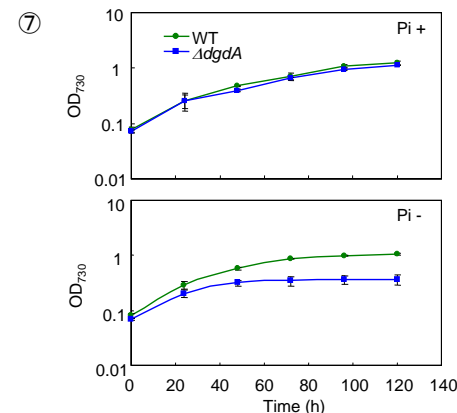


Figure 7. Growth curves in optimal (Pi+, upper panel) and phosphate deprived (Pi-, lower panel) medium. Filled green circle indicates the WT and filled blue square the $\Delta dgdA$ mutant. SD is based on three independent experiments.

We did not see difference between the wild type and $\Delta dgdA$ in Pi+ condition. In Pi- condition, the mutant grew slower than the wild type.