Prof. Josef Komenda's group
Laboratory of Photosynthesis

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Title of the project: Dissecting PSI Assembly: Characterization of Deletion Mutants and Affinity Purification of Tagged Assembly Proteins in *Synechocystis* sp. PCC 6803

For how many student/s: 1-2

Description of the project:

Photosynthesis powers life on Earth by harnessing light energy to convert inorganic carbon dioxide into organic compounds, with oxygenic photosynthesis in cyanobacteria, algae, and plants producing oxygen as a byproduct through the photosynthetic electron transport chain. This chain relies on photosystems I and II (PSI and PSII), the cytochrome b6f complex, and electron carriers. PSI plays a pivotal role by oxidizing plastocyanin or cytochrome c_6 to energize electrons for reducing ferredoxin. Over the past two decades, high-resolution structural models of PSI from various organisms have revealed its architecture, including the core PsaA/PsaB heterodimer and peripheral subunits like PsaF, PsaI, PsaJ, PsaK, PsaL, and PsaM, which coordinate pigments and support oligomerization. Yet, while PSI's mature structure is well-mapped, its biogenesis, the regulated assembly of subunits, pigments, and cofactors within the thylakoid membrane, remains poorly understood.

In this project, student(s) will investigate PSI assembly in the model cyanobacterium *Synechocystis sp.* PCC 6803 by mastering essential molecular biology and biochemistry techniques. Hands-on tasks include:

- Culturing *Synechocystis* on small and large scales to optimize growth conditions.
- Performing genetic manipulations: designing primers, conducting PCR amplification, running agarose gels, and preparing overexpression plasmids to tag PSI assembly proteins.
- Cell lysis, harvesting membranes, and using affinity purification to isolate tagged PSI proteins and subcomplexes.
- Analyzing samples via polyacrylamide gel electrophoresis (PAGE) on isolated membranes and purified proteins to visualize native complexes and assess their composition and complexity.

Through these steps, student will gain practical skills in microbial cultivation, genetic engineering, protein purification, and gel-based complex analysis, contributing insights into PSI biogenesis while deepening their understanding of photosynthetic machinery.

Requirements:

A curious and imaginative mindset is highly valued. The student is expected to be proficient in both spoken and written English. Language requirements also include a basic understanding of written scientific texts. Previous laboratory experience is an advantage but not required. However, as a significant portion of the project will take place in a wet lab, the student should be comfortable handling glassware and performing tasks that require fine motor skills.