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Light induced ATP synthesis for synthetic biology applications

Christin Kleineberg Max Planck Institute for Dynamics of Complex Technical Systems, Germany

A unique feature of living cells is an ability to extract energy from their environment and to use this energy to carry out activities such as growth, movement or reproduction. In general, energy of nutrients (in cellular respiration) or light (in photosynthesis) is transformed via electron transfer chains (respiratory or photosynthetic) into a proton gradient across the cell membrane which is finally utilized for adenosine triphosphate (ATP) production. On the other hand, bacteriorhodopsin (bR) is widely known as proton pump machinery that transports the protons to the other side of the membrane due to light stimulation. Racker and Stoeckenius have studied a model system by combining bR and isolated ATP synthase in phospholipid vesicles for light induced ATP production. In order to design and construct a synthetic cell in the synthetic biology context, this kind of energy conversion "apparatus" should be well defined and optimized. Therefore, our idea is to build up a light-driven ATP generating "bioreactor" using the bottom up synthetic biology approach. In this context, functional parts (ATP-generating functional part and light-driven proton gradient functional part) are created, investigated, optimized separately and combined afterwards.

kleineberg@mpi-magdeburg.mpg.de

Comprehensive examination of intracellular metabolite of *Pseudomonas Taiwanensis* VLB120 using liquid chromatographic techniques coupled with mass spectrometer detection

Gossa Garedew Wordofa Technical University of Denmark, Denmark

A bsolute quantification of free intracellular metabolites is a meaningful tool in both pathway discovery and metabolic engineering efforts. Today, improved metabolomics tools make it possible to detect and quantify a wide range of metabolites providing useful information for pathway discovery and to improve the understanding of cellular systems and metabolism. However, most of these tools were designed for specific classes of metabolites and microorganisms; but in many studies sampling protocols are simply taken from the literature without validating them for the given conditions and the investigated organism, which can lead to highly biased results. Thus, sampling, quenching and extraction protocols need to be validated for each microorganism and metabolite of interest. Currently, we are investigating the metabolic state of *Pseudomonas taiwanensis* VLB120 to provide a useful reference dataset of absolute intracellular metabolite concentrations. We are also testing the suitability of different metabolomics tools with the objective of identifying a better sampling and quenching/extraction techniques for *P. taiwanensis* VLB120 metabolome study.

gowor@biosustain.dtu.dk