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The unique CO_2-concentrating mechanism of cyanobacteria

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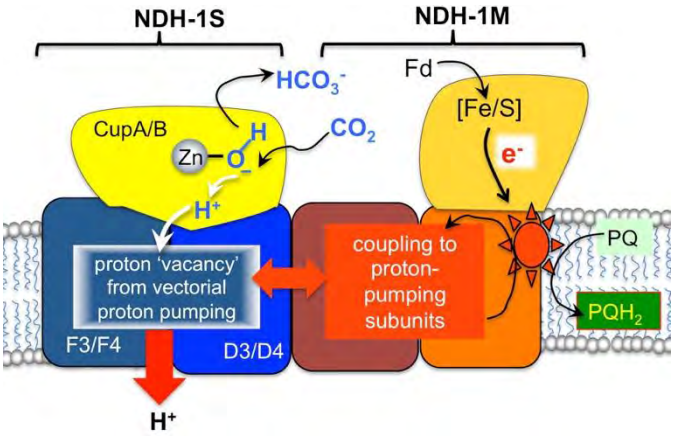
The cyanobacterial **CO_2-concentrating mechanism** (**CCM**) efficiently supplies CO_2 to the major CO_2 fixing enzyme, Rubisco. Essentially, it functions as a 'supercharger' for CO_2 by concentrating inorganic carbon as HCO_3^- within the cytoplasm, then converting to CO_2 within the Rubisco-containing carboxysome. This effectively saturates the active sites of Rubisco, minimizes wasteful photorespiration, and thus increases the maximal rate and efficiency of overall photosynthesis. As with other aquatic photosynthesizers, cyanobacterial growth and productivity is often limited by the availability of inorganic carbon (C_i), which may be acquired either as CO_2 or as HCO_3^-, depending upon specific mechanisms and environmental circumstances. Accordingly, cyanobacteria have evolved a surprisingly diverse and complex CCM. An overview will be given of the structure, activity, and regulation of the cyanobacterial CCM, focusing on the model organism, *Synechocystis* sp. PCC6803. This will include a discussion of the some of the biophysical techniques for the non-invasive tracking of the physiological state of cells experiencing fluctuations in the availability of inorganic. It is shown that careful application of chlorophyll fluorescence and other spectroscopic techniques reveals characteristic changes in the metabolic state of cells that have been challenged with inorganic carbon limitation. Our efforts have now turned to CO_2 uptake enzymology and progress towards understanding the specialized NDH-1 complexes that catalyze CO_2 uptake, will be discussed. These large, membrane-bound bioenergetic complexes share features of proton-pumping respiratory NDH-1 complexes, yet are unique to cyanobacteria. The cyanobacterial variants have additional protein subunits, some of which function to convert CO_2 to HCO_3^-. However, unlike typical carbonic anhydrases, these enzymes are coupled to an energy source that drives the reaction far-from-equilibrium and in favor of HCO_3^- formation. Our working hypothesis is that a metal-containing carbonic anhydrase is situated adjacent to the proton pumping domain(s) of the NDH-1 complex, which drives the $H_2O + CO_2 \rightleftharpoons HCO_3^- + H^+$ reaction to the right by removal of the proton from the active site of CO_2-hydration. Whatever, the actual mechanism, these enzymes could prove fundamentally interesting and potentially have far-reaching applied significances. Progress towards the development and application of molecular tools for site-directed mutagenesis to understand the specialized NDH-1 complexes that catalyze this critical CO_2 uptake mechanism will be presented.



Hypothesis for electron transfer coupled to proton pumping coupled to CO_2-hydration. Hypothetical metal center, possibly Zn^{2+} ion. CupA/B may provide all or some of the ligands (likely His and/or Cys) to the metal center. The metal facilitates the de-protonation of substrate H_2O forming a hydroxide capable of nucleophilic attack upon incoming CO_2 as in the case of carbonic anhydrases. Proton pumping activity removes H^+ from the active site to trap the deprotonated metal hydroxide formed upon deprotonation of the metal-bound H_2O.

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Dr. Robert L. Burnap received his bachelor's degree at the University of Michigan. He went on to the University of California at Los Angeles for a master's in biology and then to University of California at Santa Barbara, where he developed a doctoral thesis on the evolution of the oxygenic photosynthetic mechanism. His postdoctoral training was in the genetic manipulation and biophysical analysis of photosynthetic proteins at Purdue University under the direction of Professor Louis Sherman.

Research in the Burnap lab focuses on photosynthesis, the conversion of light (solar) energy to chemical energy that can be utilized to drive the metabolism of living cells. It involves the genetic engineering of the photosynthetic molecules in algae and the use of various biochemical and biophysical techniques to evaluate the function of photosynthetic mechanism. This is leading to a better understanding of photosynthesis and a better ability to harness its power. There are two major on-going projects: The first, supported by the National Science Foundation, investigates the metal-containing enzyme that utilizes light energy to split water – a process that yields energy-rich, bound hydrogen and releases oxygen. The second, funded by the Department of Energy, studies the metabolic signals that control the expression and activity of the carbon dioxide-concentrating mechanism, which supports the assimilation of atmospheric carbon dioxide in support of plant and algal growth.

Burnap teaches several courses at OSU, including cell & molecular biology, bioenergetics and bioinformatics.

Burnap has recently returned from an appointment as a rotating program director in the Division of Cellular and Molecular Biochemistry at the National Science Foundation.

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