MOLECULAR PLANT PHYSIOLOGY

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Photosynthesis and respiration in photosynthetic organisms are the fundamental processes that support all life on earth. The capture of CO_2 from the atmosphere and its transformation into biomass is of vital importance to the health and productivity of natural environments and to the economic viability of agricultural industries. Our research uses a functional genomics approach to understand the fundamental genetic and biochemical factors that determine how photosynthetic organisms perform in their environments.

We investigate how CO_2 is captured from both atmospheric and aquatic environments by plants, algae and cyanobacteria and then used for growth. We also study how photosynthetic and respiratory mechanisms are involved in the adaptation of these organisms to variable environments and how these mechanisms limit the efficiency of growth.

A long term aim is to identify genetic information that might be used to modify agricultural plant species in order to either increase yields, or to tolerate extreme environmental conditions such as high temperatures, higher levels of light and CO_2 and lack of water.

Leaves of the cactus *Kalanchoe daigremontiana.* This plant performs a special CAM photosynthesis which allows it to conserve water during growth in dry environments.

HIGHLIGHTS

- We applied gene tagging and proteomic techniques and to study protein:protein interactions present in unique protein micro-compartments known as carboxysomes (polyhedral bodies) from a freshwater cyanobacterium. Carboxysomes play an essential role in efficient CO_2 fixation in cyanobacteria. The main carboxysome shell protein, CcmM, was found to interact with the CO_2-fixing enzyme, Rubisco. This work has importance in understanding the composition of these proteinaceous structures and ultimately how they are assembled and function.

- We used a mutant of *Amaranthus edulis* (C_4 dicot) that lacks phosphoenolpyruvate carboxylase (PEPC) to study its role in stomatal movement and showed that PEPC activity is required for normal stomatal opening in response to both light and low CO_2. Our studies imply that carbon metabolism associated with malate production via PEPC plays an important role in the stomatal regulation of plant water use, most likely by serving as one of the anions that contribute to the maintenance of the proton and charge balance which occurs during stomatal movement.

- We have identified new Rubisco enzyme variants which improve the growth fitness of a newly engineered *E coli* strain. This was achieved by coupling directed evolution approaches with a screen using *Escherichia* cells that have been engineered to be dependent on Rubisco for survival. Kinetic characterisation of these evolved Rubisco variants has uncovered novel molecular interactions that improve our understanding of structure-function relationships in Rubisco.

- We have developed new mass spectrometric techniques which allow us to examine the isotopic discrimination of both *in vivo* and *in vitro* reactions with respect to the use of O_2 and CO_2 species as substrates. This involves the on-line and real-time analysis of gases from the liquid or gas phases of the reaction without separate isolation and introduction into the mass spectrometer. This new approach has been applied to studies of the Rubisco carboxylase reaction, the respiratory oxidase reactions and whole leaf gas exchange during photosynthesis.