

**The 10th International Symposium on Inorganic Carbon Utilization by
Aquatic Photosynthetic Organisms**

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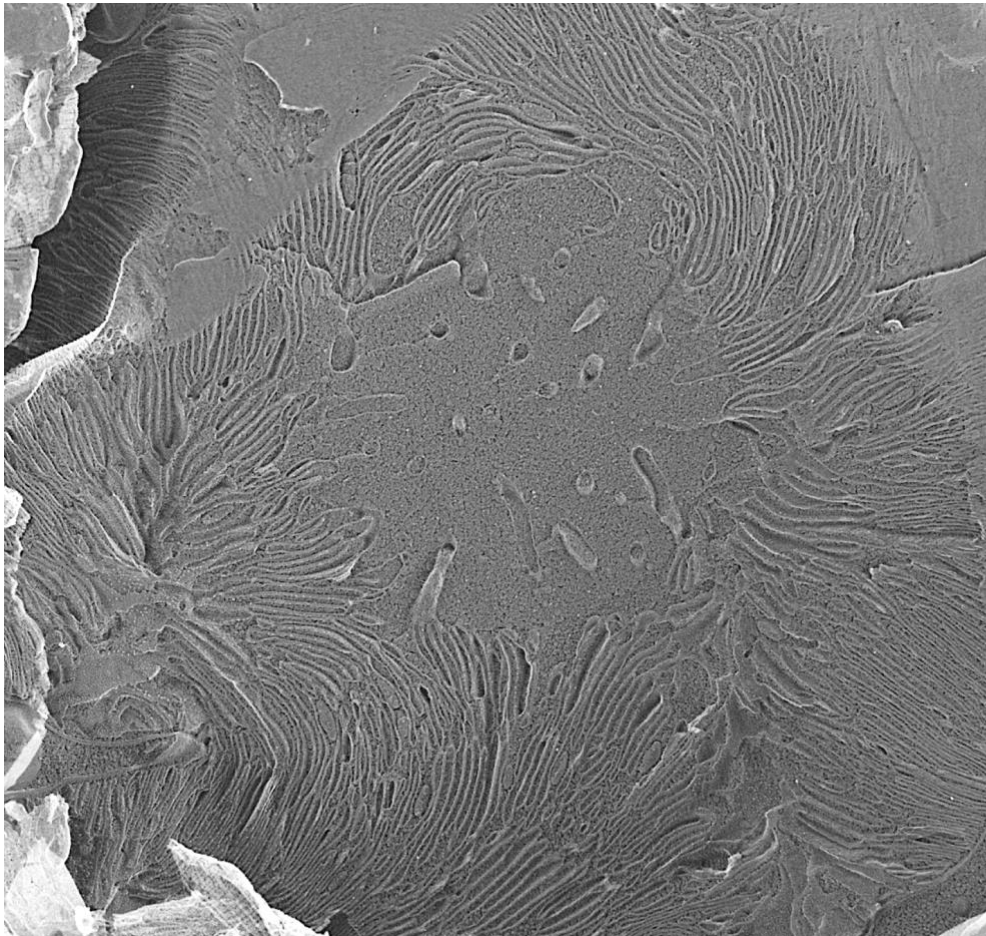


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4. Interplay among environmental factors, development of CO₂ concentrating mechanisms and phylogenetic constraints shaping Rubisco evolution across aquatic photosynthetic organisms

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Rubisco is the main CO₂-fixing enzyme in the biosphere and sustains the vast majority of food chains on Earth. Although Rubisco catalysis is currently an area of active study, there is still a strong debate about the main drivers shaping Rubisco evolution (phylogeny, catalytic trade-offs, environment), and its natural catalytic diversity, especially in aquatic organisms, remains poorly understood. Aquatic carbon fixation not only forms the basis of marine and freshwater food webs, representing half of the global primary productivity, but also exerts an important control on the global carbon cycle through regulation of the ocean's biological pump. Recently, a wide intraspecific laboratory-to-laboratory variability for Rubisco catalytic traits has been identified, which is in part due to the different methodologies used. Thus, an integrative and complete meta-analysis of study-to-study normalized Rubisco kinetic data becomes an essential requisite to succeed in providing new relevant information on Rubisco phylogenetic evolution and environmental adaptation in aquatic ecosystems. The aim of this study was to undertake an exhaustive compilation of all the data published so far on the *in vitro* Rubisco catalytic parameters of autotrophic organisms containing Rubisco, from bacteria to vascular plants, especially those inhabiting aquatic environments. Original data were corrected using novel formulation to eliminate methodological effects. Rubisco evolution was re-evaluated in the context of its environmental drivers (CO₂, O₂ and temperature) and in the context of the development and effectiveness of carbon concentrating mechanisms, which are present in most aquatic organisms as a consequence of low CO₂ diffusion in water and the well-known Rubisco kinetic limitations. The phylogenetic signal in Rubisco catalysis was also analyzed by evaluating the dependence of variation in kinetic traits on the phylogenetic tree constructed from the Rubisco large subunit sequence of the compiled species. Our results showed that phylogenetic constraints in Rubisco adaptation mostly determine the specificity factor values of current species. However, the adaptation of the Rubisco affinity for CO₂ and carboxylation efficiency has been more influenced by the environmental conditions than by phylogenetic constraints, either provoking an evolution of a more CO₂-specific and efficient Rubisco enzyme under limiting intracellular CO₂ conditions, or a relaxation of CO₂ specificity and efficiency when co-evolved with CCMs. In addition, the effectiveness of CCMs seems to determine the degree of relaxation of Rubisco CO₂ specificity and efficiency. Importantly, this analysis questions the canonical trade-off between Rubisco kinetic traits previously described for Spermatophyta, but also refutes the idea that phylogenetic constraints limit Rubisco adaptation to a great extent, and recognizes a larger plasticity of Rubisco kinetics than previously thought. This information will be especially useful for understanding the effects in the productivity and ecological success of the different aquatic organisms in future scenarios of Global Change.

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