Nitrate sensing and uptake in diatoms: from molecular evolution to functional characterization

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Project Summary

Every five breaths we take, one comes from diatoms. Diatoms are the major and most diverse group of phytoplankton. Their adaptive capacity suggests that they have developed sophisticated mechanisms to perceive and respond to environmental variations. Marine diatoms are often exposed to unpredictable environmental conditions changes. In particular, nitrogen (N) concentration in the ocean shows significant temporal and spatial variability, which affects growth and the distribution of phytoplankton. The molecular mechanisms that allow diatoms to efficiently cope with N availability remain largely unknown. Recent genomics and metagenomics studies uncovered an unexpected complexity and revealed that diatoms host a larger suite of N transporters than one would expect for a unicellular organism. Indeed, data have shown the presence of multiple NH_4^+ (AMT) and NO_3^- (NPF/NRT2) transporter genes differentially regulated (Rogato et al., 2015; Busseni et al., 2019, Molecular Biology and Evolution, in revision). The project aims at investigating the molecular mechanisms of N sensing and uptake in marine diatoms, focusing on the identification and functional characterization of specific transporters. The low and high affinity nitrate family members (NPF/NRT2) are expected to be of particular relevance because NO_3^- represents the major N source in the ocean. For this reason, taking advantage of recent advances in diatom genomics and combining genome- enabled tools with classical ecophysiological and biophysical approaches, the project aims to provide a comprehensive and unprecedented look into the role of the NO_3^- (NPF/NRT2) transporter genes in diatoms. Gain and loss of function mutants in the model species *Phaeodactylum tricornutum* will be generated and characterized. RNA sequencing will be performed to compare the transcriptome profiles of wild type and mutant strains. Lines containing NPF/NRT2 transporters fused to fluorescent proteins will be obtained in order to define their subcellular localization. Overall, these data will provide a springboard to shed light on the evolutionary advantages of these N transporter genes and on the N metabolism in diatoms. The integrated work will allow to define the function of diatom NO_3^- transporter genes and to assess their role under stress conditions. This analysis will provide useful markers for mining metagenomics and metatranscriptomic data. Moreover, the results will have an impact on creating novel genetic resources and possible biotechnological applications. Knowledge on N regulation will also potentially accelerate the discovery of new strategies to improve biomass production, a major challenge to address future demands of food and energy.