

Title of the PhD project:

SEISPE - The Seipin protein of *Phaeodactylum*: structure and function specificities

PhD supervisor:

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Host laboratory:

Laboratoire de Physiologie Cellulaire & Végétale

Biogenèse, dynamique et homéostasie des lipides membranaires (LIPID) team

Project summary: Microalgae are a diverse group of photosynthetic microorganisms. Many species of microalgae produce oil in response to stress, making them particularly interesting for the production of third-generation biofuels. However, the mechanisms of formation of lipid droplets (LDs), the oil-containing organelles, remain poorly understood. The Seipin protein is known to be involved in the biogenesis processes of LDs in many organisms. We identified PtSeipin, a homolog of this protein in the diatom *Phaeodactylum tricornutum* and investigated its function using genetics tools (KO and overexpression). We confirmed its essential function in the regulation of LD formation but also revealed functional specificities that distinguish PtSeipin from its homologs in other organisms. Seipin proteins have two transmembrane domains and are anchored in the endoplasmic reticulum (ER). The central domain, located in the ER lumen, allows oligomerisation of the protein and its structure (studied by cryo-EM in yeast, *Drosophila* and humans, modelled by alphaFold in many other species) shows remarkable conservation, yet the diatoms Seipin exhibit some particular features. The N- and C-terminal regions, on the other hand, are highly variable and show no conservation (sequence or structure). These regions are involved in the interactions with the many partners of Seipin, which are essential for its function. The objectives of this thesis are therefore to study the structure of the PtSeipin protein and its various functional regions and to identify its protein partners. This study will allow us to understand the functional specificities of PtSeipin compared to its counterparts.

Preferred skills: We are looking for a motivated and curious student, interested in basic research with application potential and willing to learn a variety of techniques with the help of experts. Background/experience in at least one of the following is desired: molecular biology, biochemistry, structural biology. An experience in cell culture and cell biology would be appreciated.

Student role: The project is composed of two parts in which the PhD will participate. The first part focuses on understanding the roles of different parts of the PtSeipin protein and involve structural biology and complementation of KO mutants with mutated or non-mutated proteins. The PhD will participate in the preparation of samples for Cryo-EM and Cryo-EM experiments, in collaboration with Felix Weis at IBS. The PhD will benefit from the team's expertise to generate the microalgae transformed lines (vector preparation, microalgae transformation) and perform phenotypic studies (growth, microscopy observations, lipidomics analysis). The second part will focus on protein-protein interactions (yeast 2-hybrid, BiFC, co-IPs) and enzymatic activities evaluation.

Keywords: microalgae, lipid droplets, protein structure, protein-protein interactions

Relevant publications of the team:

Lupette J, Tardif M, Brugière S, Couté Y, Salvaing J, Maréchal E **2022**. Quantitative proteomic analyses reveal the impact of nitrogen starvation on the proteome of the model diatom *Phaeodactylum tricornutum*. *Proteomics*.

Le Moigne, Guéguen and Salvaing **2022**. Lipid Droplets : more than a fat storage. *Advances in Botanical research*

Guéguen N, Le Moigne D, Amato A, Salvaing J and Maréchal E **2021**. Lipid Droplets in Unicellular Photosynthetic Stramenopiles. *Frontiers in Plant Science*

Jaussaud A., Lupette J., Salvaing J., Jouhet J., Bastien O., Gromova M. and Maréchal E. **2020** Stepwise Biogenesis of Lipid Droplets in Nitrogen Starved *Phaeodactylum tricornutum* Cells. *Frontiers in Plant Science*

Lupette J., Jaussaud A., Seddiki K., Morabito C., Brugière S., Schaller H., Kuntz M., Putaux JL, Jouneau PH., Rébeillé F., Falconet D., Couté Y., Jouhet J., Tardif M., Salvaing J. and Maréchal E. **2019** The Architecture of Lipid Droplets in the Diatom *Phaeodactylum tricornutum*. *Algal Research*