Master
Biology AgroSciences
Bordeaux B2AS

université de BORDEAUX

M2 INTERNSHIP SUBJECT

TITLE: Tracing the intracellular paths of targeted protein secretion

CONTEXT: Targeted protein secretion is a process crucial to eukaryotic cells to drive protein localization at the right place and at the right moment. Plants use this process to **adapt to environmental changes and coordinate their growth**. Identifying the intracellular paths that targeted secretion is using is valuable in many aspects, not only to extend our basic knowledge but also to understand how plants deal with changing environment to adapt their growth. To reach this goal, two biological processes are particularly important to consider: 1) the cell-to-cell communication mediated by the **plasmodesmata (PD) pores** that localize at plasma membrane and which interconnect virtually all cells within the plant body, establishing direct membrane and cytoplasmic continuity, a situation unique to plants; 2) the establishment of **polar domains at the plasma membrane** that instruct a directionality to plant hormone or ions fluxes and allow to orient plant growth directionality and adapt this directionality upon environmental changes. In both cases, subsets of proteins are targeted to either PD or polar domains of the plasma membrane through **unknown trafficking paths and cellular mechanisms**.

The candidate will work at the interface of two research groups: the group of Emmanuelle Bayer (PD) who lead a team of 10 people and the group of Yohann Boutté (polar domains) who lead a team of 4 people. The LBM is a very international laboratory of around 50 people amongst which half are students, post-doc and contract engineers.

OBJECTIVES: To follow the dynamic trafficking of markers of either PD or polar domains of the plasma membrane and identify critical steps of this process, the host groups are currently establishing, in plants, the Retention Using Selective Hook (RUSH) system. This system allows to trap newly synthesized proteins at the ER using specific molecular hooks and selectively release them upon rapamycin induction to monitor their dynamics of trafficking from the ER using live confocal imaging. The host groups have already a proof-of-concept of the method for soluble proteins and are now developing new hooks to trap membrane proteins. The candidate will be involved in this methodology development and will be supervised by Emmanuelle Bayer, Yohann Boutté and Matheus Montrazi (engineer in the group).

METHODS:

- Multisite Gateway molecular cloning
- Transient and stable transformation of Arabidopis plants
- Live cell confocal microscopy, dynamic tracking
- Confocal microscopy quantification, statistics

PREREQUISITES:

- Be highly motivated and interested by cell and molecular biology
- Be able to work within a group (attentiveness and adaptability) and have a certain degree of autonomy
- Be able to communicate in English

REFERENCES:

- Ito Y, Esnay N, Platre M, Wattelet-Boyer V, Noack L, Fougère L, Menzel W, Claverol S, Fouillen L, Moreau P, Jaillais Y, **Boutté Y***. (2021). Sphingolipids mediate polar sorting of PIN2 through phosphoinositide consumption at the *trans*-Golgi Network. ***Nature Communications***, 12(1): 4267.

- Wattelet-Boyer V, Brocard L, Jonsson K, Esnay N, Joubès J, Domergue F, Mongrand S, Raikhel N, Bhalerao RP, Moreau P, **Boutté Y***. (2016). Enrichment of hydroxylated C24- and C26-acyl-chain sphingolipids mediates PIN2 apical sorting at *trans*-Golgi network subdomains. ***Nature Communications***, 7:12788.

- Brault ML+, Petit JD+, Immel F, Nicolas WJ, Glavier M, Brocard L, Gaston A, Fouché M, Hawkins TJ, Crowet J, Grison SM, Germain V, Rocher M, Kraner M, Alva V, Claverol S, Paterlini A, Helariutta Y, Deleu M, Lins L, Tilsner J*, **Bayer EM*** (**2019**) Multiple C2 domains and transmembrane region proteins (MCTP) tether membranes at plasmodesmata. *EMBO Rep*. e47182, p1–26

- Nicolas W, Grison MS, Trépout S, Gaston A, Fouché M, Cordelières F, Oparka K, Tilsner J, Brocard L*, & **Bayer EM***. (**2017**) Architecture and permeability of post-cytokinesis plasmodesmata lacking cytoplasmic sleeve. Nature Plants. 3:17082

KEYWORDS: trafficking, targeted secretion, cell-to-cell communication, cell polarity, confocal microscopy

NAME OF THE SUPERVISOR:

BOUTTE Yohann

BAYER Emmanuelle

NAME and ADDRESS OF THE TEAM/LABORATORY:

Membrane Dynamics in Intra- and Inter-Cellular Trafficking. Laboratoire de Biogenèse Membranaire, UMR5200, Bâtiment A3 – Campus INRAe Bordeaux Aquitaine
71 Avenue Edouard Bourlaux – cs20032
33140 Villenave d'Ornon Cédex

E-MAIL: yohann.boutte@u-bordeaux.fr