Proposition d'un sujet de stage au M2 ADAM (2018-2019)

Acceptez-vous que ce sujet soit proposé aux étudiants de l'itinéraire « Pro » ? OUI

Titre	**Role of PAC domain proteins in the green lineage: from *Marchantia polymorpha* to *Arabidopsis thaliana***
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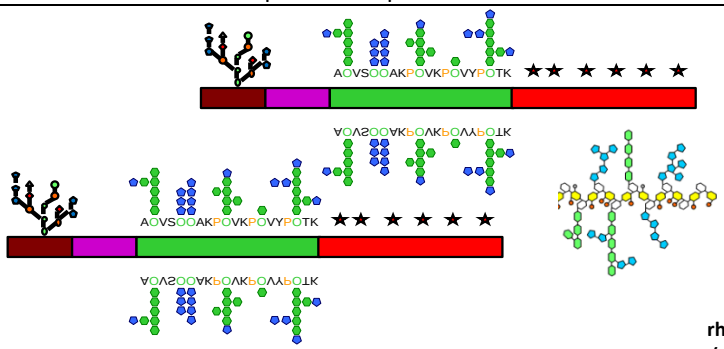
Résumé	Plant cell walls are the major component of plant biomass and play important roles during plant development and in response to environmental constraints. Plant cell walls are natural composite structures, mostly made of polysaccharides, proteins and lignin. Polysaccharides represent up to 95% of the primary cell wall mass, whereas cell wall proteins (CWPs) account for 5-10%. Cell wall proteomics has revealed the high diversity of protein content [Jamet *et al.* 2008, Proteomics 8: 893] and CWPs play many roles in plant cell wall plasticity [Frankova and Fry 2013, J Exp Bot 12: 3519]. However, not much is known about the cell wall supramolecular architecture in which CWPs are certainly major players via protein/protein or /polysaccharide interactions. The *Arabidopsis thaliana* AtAGP31 (ArabinoGalactan Protein 31) has been identified as a major protein in actively growing etiolated hypocotyls. From N- to C-terminus, it comprises several domains among which a *O*-glycosylated proline (Pro)/hydroxyproline (Hyp)-rich domain, and a PAC (Pro-rich protein, AGP, Cysteine containing) domain. The PAC-domain is found in proteins predicted to be secreted in all land plants including mosses and liverworts, *e.g.* *Physcomitrella patens* and *Marchantia polymorpha*. The PAC domain of AtAGP31 has been shown to interact *in vitro* with (i) cell wall polysaccharides among which galactans and (ii) *O*-glycans of the AtAGP31 Pro-Hyp-rich domain. We assume that AtAGP31 oligomers are formed *in muro* as well as non-covalent complexes between the PAC domain and cell wall polysaccharides [Hijazi *et al.* 2014, Ann Bot 114: 1087]. CWPs containing PAC domains could thus be part of non covalent networks allowing cell wall elongation [Hijazi *et al.* 2014, Front Plant Sci 5: 395]. This project will have four parts: (i) characterization of the binding properties of available recombinant PAC domains of *M. polymorpha* and *A. thaliana* to cell wall polysaccharides, based on preliminary results showing different specificities of interactions; (ii) screening and macrophenotyping of knock out mutants of *M. polymorpha* defective in either or both of the two PAC domain proteins obtained using the CRISPR/CAS9 technology; (iv) obtention of sur-expressors of both *M. polymorpha* PAC domain genes. The results obtained during this internship will pave the way for a PhD project. Indeed, the PAC domain protein family comprises only two members in *M. polymorpha*, compared to 14 members in *A. thaliana*. It will be easier to characterize a phenotype in *M. polymorpha* because of limited possible functional redundancy. In addition, complementation studies with *A. thaliana* proteins will be feasible. These mutants will provide valuable tools to understand the function of PAC domain proteins in plant cell walls.
Illustration	 **rhamnogalacturonan II (galactan side chains)** A model for protein/protein non-covalent networks in plant cell walls: the AtAGP31 protein as a central player. Its His-rich domain is represented as a pink rectangle, its AGP domain as a brown one, its Pro/Hyp-rich domain as a green one and its PAC domain as a red one. Stars stand for conserved Cys residues. The *O*-glycosylations on the AGP and Pro/Hyp-rich domains are shown.