Université de technologie de Compiègne – Thesis proposal

Part 1: Scientific sheet	
Thesis proposal title	**Study of the interaction of salicylic acid on enzymes of plant metabolism**
PhD grant	Contrat doctoral sur allocation Ministère
Research laboratory	UMR CNRS 7025 Génie Enzymatique et Cellulaire Thème Métabolisme Végétal et Bioressources http://www.umr7025-gec.fr/
Thesis supervisor(s)	Eric RUELLAND
Scientific domain(s)	Main: Biology Secondary
Research work	Faced to attacks by pathoghens, plants will synthesize a small bioactive molecule (a phytohormone) named **salicylic acid (SA)**. SA will then be transduced into plant cells to activate further signaling pathways that are necessary for the plant response. Several aspects of SA-induced regulation are known. **SA induces a redox change** in the cells that conditions the monomerization of NPR1 protein. The monomers of NPR1 then shuttle into the nucleus and interact with transcription factors, allowing the induction of SA-response genes (Vlot, et al., 2009). However, many questions regarding SA signalling pathway remain unanswered. Dozens of unrelated plant proteins capable of binding SA were recently identified by several biochemical high through-put SA-binding screenings (Manohar, et al., 2015). This data challenges the common paradigm where a hormone is recognized by a single protein or a single protein family. The main goal of this project is to understand the mechanisms of SA binding to proteins. This will be achieved by **combining molecular dynamics calculation with methods of protein/ligand biochemical assays**. The molecular dynamic will help to model and **simulate such interaction over time**. Thus, the amino acid residues involved in the protein/SA interaction will be identified. **These findings will be validated experimentally by the production of recombinant proteins**. Several complementary **SA/protein binding assays** will be carried out *in vitro* using the purified proteins in their native form but also with mutated proteins where amino acid residues are substituted based on the data collected from *in silico* modelling. For the project, we have chosen to work on a limited number of candidate SA-binding proteins. The priority was given to proteins whose activity is essential for plant physiology or those with potential functions in the SA signalling cascade. These are i/ Glutathione-S-transferase (AtGSTF8) ii/ Thioredoxins m1 iii/ a betaCarbonylAnhydrase (AtCA5) iv/ Glutathione peroxidase1 (AtGPX2). We successfully used this approach with GAPDHA1 (Pokotylo et al., 2020). The PhD student will apply the techniques and methodology validated on GAPDH on the other proteins while implementing new ones (especially for biochemical assays of protein/SA interaction, such as isothermal titration calorimetry). The PhD student will mostly be in charge of the biochemistry aspects of the project. This project is part of a bigger project aiming at deciphering the crucial role of SA in the balance between growth and immunity in plants.
Key words	Salicylic acid, protein ligand interaction, protein production, molecular dynamics
Requirements	Plant physiology, recombinant protein purification
Starting time	October 2022
Location	UTC Centre de recherche Compiègne, France

Part 2: Job description	
Duration	36 months
Additional missions available	Participation in UTC lessons may be considered.
Research laboratory	The scientific approaches developed at the GEC laboratory respond to three main concepts enabling scientific and technological discoveries related to biology: bioresources, biomimicry and bioinspiration.
Material resources	The laboratory has all the necessary equipment for cloning, for the production and purification of recombinant proteins in Escherichia coli (incubators, protein electrophoresis, ChemiDoc BioRad). The laboratory has machines SPR (Surface Plamsonic Resonance) ICT (isothermal titration calorimetry).
Human resources	The GEC currently has 57 members, 34 of whom are permanent (Research/Research/Technical Staff) and 23 PhD/post-doctoral students. The laboratory is bi-localized in Compiègne (UTC) and Amiens (UPJV).
Financial resources	The project has the financial support of the Hauts-de-France region through obtaining STaRS grant funding.
Working conditions	The thesis student will be required to work on the bench. He/she will comply with the safety rules that will be explained to him when he arrives at the laboratory. His supervisor will be Eric Ruelland. A thesis monitoring committee will be set up. Meetings on request and at regular intervals to take stock of the progress of the work will be organised. The student will also present his project at various internal events (doctoral days, GEC seminars...) or external, depending on the opportunities. Applications for mobility to work with Prague employees will be submitted.
Research project	Project SAABA (L'acide salicylique au coeur de la balance entre protection et croissance chez les plantes). Project ASPIC. Dispositif Stars Hauts de France
National collaborations	I. Kleiner (LiSA UPEC, Créteil) ; T. Bouceba (Plateforme interaction bimoléculaire, SU, Paris) ; E. Issakidis (IPS2 Saclay).
International collaborations	L. Burketova (UEB, Prague), V. Kravets (NASU, Ukraine).
International cosupervision (cotutelle)	
Contact	E-mail eric.ruelland@utc.fr

Please contact first the thesis supervisor before applying online
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