

Propostas de Planos de Tese de Mestrado

Instituto de Biologia Experimental Tecnológica (iBET)
Instituto de Tecnologia Química e Biológica (ITQB-UNL)

Genomics of Plant Stress Unit (GPlantS Unit)



Title: Exploring the role of the plant hormone modulator SLR1, in rice response to abiotic stress

Rice (*Oryza sativa*) is the second most important food crop worldwide with great economical and social relevance. Nevertheless, it has its productivity greatly hampered by environmental abiotic stresses, such as cold, drought and salt, decimating many harvests every year. DELLA proteins are coded by the so called “Green Revolution genes”, and they are gibberellin (a plant hormone) response modulators. In Arabidopsis, the five known DELLA proteins are involved in plant growth and development and also in stress responses¹. However, in rice, like most crops, there is only one gene coding for a DELLA (SLR1) protein². We hypothesize that SLR1 accumulates the roles of the five Arabidopsis DELLA proteins, and thus its regulation at protein level must be pivotal for its role in specific cellular processes, e.g. in the response to abiotic stress. We propose to use rice as a model crop to study stress tolerance mechanisms in which SLR1 is the hub, by studying its relation with several proposed interactors, already associated with hormone and stress signaling.

Detail of Internship and specific aims:

This internship proposal will be integrated in the European project DELLA-STRESS, Plant KBBE, and will provide the student with the opportunity to work within DELLA-STRESS consortium, in close collaboration with our partners in Spain (CSIC-Centro Nacional de Biotecnología) and Germany (Technische Universität München).

The main goal of this internship is to contribute to the characterization of SLR1 function in rice abiotic stress response. Towards this, the student will compare SLR1 protein levels in several rice commercial genotypes, with different degrees of abiotic stress tolerance. To understand the variations in SLR1 abundance, the student will also determine the transcript levels of SLR1 gene and of several other key genes of the gibberellin regulatory pathway, and help characterize SLR1 interactome. In parallel, he/she will help to characterize the performance under stress of transgenic Arabidopsis plants where SLR1 substitutes all Arabidopsis DELLA proteins.

Tasks:

1. Phenotypical analysis of several commercial and tolerance-contrasting rice varieties under stress conditions

2. Transcriptomic and proteomic analysis of selected rice varieties subjected to stress treatments

The working material of the tasks 1 and 2 will derive from stress assays performed in rice under control and stress conditions, either in a glasshouse or a special growing chamber for rice. Several varieties will be used in this study, either of commercial interest or known for their contrasting tolerance to the tested conditions.

3. SLR1 interactome screenings through yeast-two hybrid in cDNA stress libraries

As it is known that the function that DELLAs exert under stress is not only dependent on gibberellin activity but also on interactions with other proteins and transcription factors that will enhance the stress response, the selected student will perform a global search for SLR1 interactors through yeast two-hybrid screenings, taking advantage of stress libraries already available at the lab.

4. Heterologous expression of SLR1 in *Arabidopsis thaliana* mutants and tolerance assessment under stress conditions

It is our hypothesis that crops with a single DELLA must retain the differential functionality of all five DELLAs existing in Arabidopsis in a unique protein. To prove this, Agrobacterium-mediated transformation of already available tetra-DELLA Arabidopsis mutants will be performed to obtain transgenic plants overexpressing SLR1, as well as expressing the rice DELLA under the influence of an endogenous DELLA promoter. Several in vitro assays will be performed in these plants to assert their enhanced tolerance compared with the wild-type.

Techniques:

- Total RNA extraction
- Protein extraction
- DNA agarose and Protein SDS-PAGE gel analysis
- Reverse Transcription-PCR (RT-PCR)
- Real Time Quantitative PCR (qRT-PCR)
- Western Blotting
- Yeast-two hybrid screening system (Y2H)
- Agrobacterium-mediated plant transformation (floral dip method)
- *In vitro* stress assays (cold, salt, drought)

Place: Genomics of Plant Stress Unit, Instituto de Tecnologia Química e Biológica (ITQB)/Instituto de Biologia Experimental Tecnológica (iBET), Oeiras, Portugal, under direct supervision of Dr. Isabel Abreu and working together with PhD student Nuno Miguel Gonçalves.

Duration: 9 months to 1 year

Number of students: 1 highly motivated student that wishes to pursue a career in research. Proficiency in English is desirable.

Contacts and more information: Dr. Isabel Abreu (abreu@itqb.unl.pt)

Bibliography and suggested reading:

1. Colebrook, E. H., Thomas, S. G., Phillips, A. L. & Hedden, P. The role of

gibberellin signalling in plant responses to abiotic stress. *Journal of Experimental Biology* **217**, 67–75 (2013).

2. Achard, P. & Genschik, P. Releasing the brakes of plant growth: how GAs shutdown DELLA proteins. *J. Exp. Bot.* **60**, 1085–1092 (2009).