

Abre-se 1 lugar para estágio de Mestrado em Biologia Celular e Biotecnologia (ano lectivo 2008/2009), para o Grupo de Engenharia de Plantas, no Instituto de Tecnologia Química e Biológica, em Oeiras. Pretende-se candidato com forte interesse em Investigação na área de Biologia Celular em Plantas. Dá-se preferência a alunos com média igual ou superior a 16 valores, com interesse em seguir para doutoramento .

Oferece-se um de dois possíveis Projectos de Mestrado (ver em baixo), com a duração máxima de um ano.

Os portenciais candidatos deverão enviar carta de apresentação/motivação, acompanhada de *Cv*, do qual deverão constar grau académico, classificação final e ano de conclusão, classificação em disciplinas pertinentes à área de candidatura, bem como quaisquer outros elementos que o candidato julgue valorizar a sua candidatura. Deverão ainda acompanhar a candidatura 2 cartas de recomendação.

Enviar candidatura para:

Isabel Abreu, Ph.D.

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Projecto de Mestrado em Biologia Celular e Biotecnologia (ano lectivo 2008/2009)

Tema do Projecto - SUMOproteomics – Identification of pos-translational modified proteins by SUMO, in response to drought-stress, in Arabidopsis thaliana model plant

Orientador - Dr. Isabel Abreu,

Investigadora no grupo EGP – ITQB, Oeiras

Plano detalhado - SUMO modification of proteins (sumoylation) regulates transcription factor activity, by altering protein subcellular localization and protein-protein interactions. In plants, little is known about the function of sumoylation and so far no in vivo SUMO targets have been identified. By concentrating our efforts in the study of the sumoylation pathway, using a genetic approach, we showed that sumoylation regulates several aspects of plant development and drought response. Although valuable information has been collected with this approach, a complete understanding of how SUMO controls these processes greatly depends on the identification and functional characterization of its targets. Furthermore, SUMO can reveal itself as a powerful tool in the identification of novel transcriptional regulators when used as a bait to find proteins that become modified as a response to specific treatments or conditions. The big challenge now lies in finding the right strategies to proceed to the identification of sumoylated proteins within a context

that will allow their function to be addressed. We propose to start by using drought stress as the treatment that induces sumoylation and expect to be able not only to study the regulation of known proteins involved in the response to drought but also to identify and characterize novel transcriptional factors involved in this process. Our results show that plants missing a key (although not unique) ligase in the sumoylation pathway (AtSIZ-1) are more susceptible to dehydration, leading us to believe that this process mediates drought response. As mentioned above, one of the difficulties that arise when trying to understand how sumoylation regulates cellular functions in plant cells has been that so far no in vivo targets for SUMO have been identified in plants. The difficulty to identify in vivo SUMO targets in plants probably arises from the small percentage of modified targets that are present at a given time. To overcome this, we will use transgenic Arabidopsis plants carrying a tagged SUMO protein and identify nuclear proteins of interest by 2D SDS-PAGE followed by Mass Spectroscopy Analysis. The goal is to identify new targets for SUMO in response to drought stress and to use this knowledge to design strategies to increase drought tolerance in plants.

Objectivos

Identify targets for SUMO, in response to drought stress;
Design new strategies to increase drought tolerance in plants.

Trabalho experimental

In vitro growth of transgenic Arabidopsis plants overexpressing a SUMO tagged protein;
Induction of drought stress in the plants;
Subcellular fractionation to enrich sample in SUMO modified proteins;
Specific protein purification for SUMO targets;
Analysis of SUMO targets by 1D and 2D SDS-PAGE;
Protein identification by Mass Spectroscopy;
Use of Bioinformatics tools for protein identification.

Duração e carga horária - 6 meses a 1 ano, 35 horas semanais (para 6 meses)

Local de realização - Lab. de Engenharia Genética de Plantas, ITQB, Oeiras (abreu@itqb.unl.pt)

Número de estagiários – 1

Projecto de Mestrado em Biologia Celular e Biotecnologia (ano lectivo 2008/2009)

Tema do Projecto – Assessment of heavy metal distribution in PIB-ATPases Arabidopsis thaliana mutants

Orientador - Dr. Isabel Abreu,
Investigadora no grupo EGP – ITQB, Oeiras

Plano detalhado – Transition metals are essential minerals for normal plant growth and development, but they can be toxic if a certain threshold is exceeded. In order to grow healthy, a

plant must be able to efficiently extract these metals from soil and to correctly distribute them within its cells and organelles. To do this, cells have several membrane transport systems, such as heavy metal ATPases (P1B) (HMA), Natural resistance associated macrophage protein (Nramps), used in metal uptake and distribution within cells and organelles, and metal-chelators and chaperones, involved in regulating metal availability within cells. Considerable genetic and biochemical efforts are being made towards understanding plant metal homeostasis. These have proven invaluable in elucidating the physiological function of several of the proteins involved in these processes, but little effort was done to look directly at metal distribution within plant cells and how it is affected when some of these proteins are absent. This work aims to address how metal distribution is affected in *Arabidopsis thaliana* mutants in which P1B-ATPase genes are disrupted. We propose to look at the overall metal distribution in young seedlings and within root and leaf cells. We will grow and analyze T-DNA insertion P1B-ATPase *Arabidopsis* mutants, using among others, Synchrotron based X-Ray Fluorescence methods. The gathered information will complement the available molecular and phenotypical data on these metal transporters and unequivocally contribute to a better understanding of metal uptake and detoxification in a wide range of organisms. Ultimately, this information will be used in the design of plants capable of metal hyper-accumulation with applications in phytoremediation of heavy metal contaminated soils.

Objetivos

Map heavy metal distribution in *Arabidopsis* Plants;

Analyze differences in heavy metal distribution between control and P1B-ATPases *Arabidopsis* mutant plants;

Design new strategies to increase heavy metal tolerance in plants.

Trabalho experimental

Growth of SALK line T-DNA mutants for *Arabidopsis* P1B-ATPases mutants for 2/3 generations to obtain homozygous T-DNA insertion mutants;

Phenotypic characterization of homozygous mutants;

Design and implementation of heavy metal stress assays;

Use of Synchrotron based X-Ray Fluorescence methods to assess heavy metal distribution in different plant organs in the plants under study;

Use of Bioinformatics tools for result analysis.

Duração e carga horária - 6 meses a 1 ano, 35 horas semanais (6 meses)

Local de realização - Lab. de Engenharia Genética de Plantas, ITQB, Oeiras (abreu@itqb.unl.pt)

Número de estagiários – 1