2022 International CRAG "Severo Ochoa" PhD Program

The International "Severo Ochoa" PhD Program of the Centre for Research in Agricultural Genomics (CRAG) is advertising four PhD positions in its 2022 call. This is a four-year program beginning in mid 2023. Doctoral students enrolled in this program will obtain their PhD Degree from either the Autonomous University of Barcelona (UAB) or the University of Barcelona (UB). More information about the doctoral program at CRAG can be found here: http://www.cragenomica.es/crag-phd-program

If interested in applying to the International CRAG "Severo Ochoa" PhD Program, **please carefully read the Application requirements and procedure** and check out all **available projects.**

Application deadline is **Wednesday October 5, 2022.** Shortlisted applicants will be interviewed during October 2022. Successful applicants will start their PhD projects in mid 2023.

This four-year PhD grant is funded by the "National Programme for the Promotion of Talent and its Employability 2022" from the Spanish Ministry of Science and Innovation.

Application requirements and procedure

Eligibility

1. The program is aimed at national and international students who have completed one of the following options by October 2022:

- studies that lead to an official Spanish (or from another country of the European Higher Education Area) university degree in Biology, Biochemistry, Biotechnology, or related areas and that have 300 credits (ECTS), of which at least 60 must correspond to master level.
- a degree in a non-Spanish university not adapted to the European Higher Education Area that gives access to doctoral studies in Biology, Biochemistry, Biotechnology or related areas.

2. Candidates are selected exclusively on merit, on the basis of their curriculum. Academic grades and the curriculum of applicants are evaluated, as well as reference letters and a motivation letter. No selection criteria for positive or negative discrimination are applied.

3. Applicants should have obtained a Bachelor degree after January 2019.

4. Candidates cannot be in possession of a PhD Degree.

5. Candidates cannot have been hired as predoctoral students for more than 12 months before the start of the CRAG "Severo Ochoa" PhD Program

6. Candidates cannot have started a pre-doctoral fellowship funded by the Spanish "Plan Estatal de Investigación, Desarrollo e Innovación Tecnológica" or any previous "Plan Nacional".

How to apply
Applicants should complete and submit the on-line application through **CRAGjobs**.

Applicants will be asked to upload the following documents:

- Curriculum vitae
- A motivation letter, including a brief summary of work experience and a statement of research interests and career goals (2 pages maximum).
- A scanned copy of the student's certified Academic Record, including a detailed record of study / transcript (a list of attended courses and corresponding grades): these documents must show the grades attained in exam periods.
- Copy of passport (international applicants only).
- Any additional files considered relevant to the application, but please only provide documents that are important to support it. Do not overload the application with certificates and documents of lesser significance.

In addition, applicants must ensure the submission of two reference letters from university lecturers or scientists with whom the applicant has studied or worked. Letters should be sent directly by the referees to **PhDprogram@cragenomica.es**, and should also be received by the application deadline, **October 5, 2022.** Only letters with official letterhead and signature will be accepted. Candidates are responsible for ensuring that referees submit these letters, and should consider that referees may need some time to prepare and send their letters within the deadline. Applications without reference letters will not be considered.

Please **download** referee request.

The doctoral program is in English. Therefore, a good knowledge of English is absolutely required. We encourage candidates to support the application with scores of internationally valid language exams like TOEFL or other tests. However, they are not mandatory: a verifiable education in English, or a reasonably long stay in an English speaking country are also convincing.

In the motivation letter, **applicants should indicate up to two research projects in which they would like to work, in order of preference** (see **Available Projects**, below). Moreover, if candidates have a particular interest in any one of these projects, they should also indicate it. More information on the research activities of each group can be found at CRAG website.

Applicants must submit information in English (CV, and motivation letter including summary of work experience). If the certified academic records are not in English, Catalan or Spanish, applicants must also attach a translation in one of these languages.

Applicants must upload all the required documents as **PDF files of less than 10MB**.

Please note that we can only consider applications that are complete.

Selection procedure

Applications will be reviewed through a selection process involving CRAG group leaders, including the Principal Investigators that will host the fellows. Students are preselected according to their written application, grades, and reference letters.

Short-listed candidates will be interviewed during October 2022. Candidates who are accepted for the program will be notified by email shortly after the interview period. These PhD positions are funded by the Spanish Ministry of Science and Innovation (MICIU) and CRAG. CRAG will assist the selected candidates to submit the required documents at the Spanish MICIU website in end October/November 2022. Applicants who have not been successful but have received a positive evaluation will be put on a waiting list to cover possible candidate withdrawals and future positions.

Available Projects

CEX2019-000902-S-22-1 – Characterization of the implication of subcellular distribution of RNA silencing machinery during viral infection (CEDIVI)

Principal Investigators: **Nicolas Bologna and Juan Jose Lopez-Moya**

The Potyviridae is the largest family of viruses with RNA genomes that infect plants, collectively causing rather significant losses in many crops. Despite having been studied extensively in many aspects, the mechanism beyond how the virus infects and the plant defends from these infections, in particular the subcellular localization distribution of gene products, remains largely elusive. For example, although cytoplasmic viral replication is presumed to take place in association with ER membranous structures, potyviral proteins like Nia and NIb are known to accumulate in the nucleus, and other protein like P1, a critical modulator of pathogenicity, has been reported to traffic to the nucleolus. Other than for virus movement, little is known about the subcellular localization of many viral components. In plants, small(s)RNA dependent RNA silencing is essential to regulate important developmental processes and adaptive responses to bacteria and virus. Although each sRNA pathway possesses specific characteristics in terms of biogenesis and accessory proteins, the basic mechanism of RNA silencing falls into four consensus biochemical steps: (i) induction by dsRNA, (ii) dsRNA processing by DICERLIKE (DCL) enzymes, (iii) sRNA 3'-O-methylation by HEN1 and (iv) sRNA incorporation into ARGONAUTES (AGO) proteins to generate the RNA-induced silencing complexes (RISCs)4. Research performed in the RNA silencing field over the past 15 years has mainly focused on the identification of the main players and their respective functions during viral infections. However, very little is known about the influence of subcellular localization of the RNA silencing machinery during virus infection.

The laboratory of Dr. Juan Jose Lopez-Moya has experience working with potyvirids, in particular for the characterization of RNA silencing suppressors (RSSs) within members of the family, and other topics, such as insect mediated transmission, genetic resistance, and biotechnological applications. Recently, mixed infections of unrelated viruses have been incorporated to the laboratory portfolio.

Complementarily, the laboratory of Dr. Nicolas Bologna has an extensively experience working in plant RNA silencing pathway in Arabidopsis thaliana, in particular studying the subcellular localization of DCL and AGO proteins during the biogenesis and mode of action of the miRNA pathway.

Combining different approaches and pioneer tools developed in both host laboratories, including ultrapure subcellular fractionation procedures, AGOs/DCLs fluorescence reporters, catalytic deficient DCL2, DCL3, DCL4, high-throughput sequencing, and experimental characterization of RSSs, we will decipher the importance of subcellular distribution of RNA silencing machinery and viral components during infection to tackle new spots for designing new strategies of resistance to virus infection.

Selected publications:

1. 1.Inoue-Nagata A. K . et al. (2022) The Journal of General Virology, 103(5)
2.Martínez, F . et al., (2014) Journal of virology, 88(18), 10725-37.
3.Pasin F. et al., (2022) FEMS microbiology reviews.DOI/10.1093/femsre/fuac011
4.Bologna NG, Voinnet O. (2014) Annu Rev Plant Biol. 65:473-503.
5.Giner A., et al. (2010) PLoS pathogens, 6(7), e1000996

CEX2019-000902-S-22-2 – Role of OsRAV genes and their GRN in adaptation of rice to abiotic stress

Principal Investigator: **Soraya Pelaz**

Transcription factors in the RAV family play important roles in plant physiological processes in model plants, such as flowering time, light response, leaf senescence, floral development, hormone signaling, and abiotic stress responses. In Arabidopsis thaliana, six members of the RAV family of transcription factors have been identified: RAV1, RAV1-like, RAV2 (TEMPRANILLO 2; TEM2), RAV2-like (TEM1), RAV3, and RAV3-like. TEMs are largely redundant genes that repress flowering by directly controlling FT transcription and gibberellic acid (GA) biosynthesis in response to photoperiod, low ambient temperature, age of the plant or hormone content. Abiotic stress conditions cause adverse effects on the overall growth and yield potential of diverse crop plants affecting flowering time. Many plant species are induced to flower following drought stress, a process known as drought escape that maximizes the chances to set seeds “escaping” from a potentially lethal drought condition. It has been shown that in this process there is a complete requirement of GIGANTEA (GI) under long days (LD). It has been proposed that GI may facilitate FT activation through interaction with TEM and SHORT VEGETATIVE PHASE (SVP) repressors, thus preventing their action/binding to the FT promoter. Opposite to drought, flowering is delayed upon salt increase. Salinity-induced flowering depends on GA-signalling and GI degradation which results in low FT expression. We have uncovered that tem1 tem2 mutant plants better cope with mild and high salt stress compared to wild types. Under salt stress conditions, the double tem1 tem2 mutant accelerated floral transition and germination because of up-regulation of floral integrators and GA biosynthetic genes. Furthermore, senescence is delayed in tem1 tem2 mutants, likely due to the observed down-regulation of genes involved in the biosynthesis of the senescence promoting phyto-hormone JA. Moreover, physiological analyses indicated that tem mutants tolerated better oxidative stress at maturity as they increased the production of important secondary metabolites with anti-oxidant properties involved in ROS scavenging. Taken all together, these findings indicated a key role for TEM in adaptive growth as negative regulators of plant development in response to salt stress.

In addition, TEM involvement in the responses to drought was tested. We found that the precocious flowering time of the mutants is not affected when water is reduced, indicating that tem mutants show a constituve drought escape and they flower at the same time regardless the amount of water. Interestingly, these mutants also showed a reduced water loss due to reduced transpiration and perhaps the increased trichome number, and are more resistant to dessication than wild-type plants. Thus, tem mtants became almost insensitive to drought (unpublished). TEM genes seem central integrators of the plant responses to environmental stimuli that affect plant development.

Unlike Arabidopsis, rice flowering is more rapidly induced under SD (Oryza sativa) than under LD. We have found than one of the four OsRAV genes acts as a repressor of flowering. Thus, downregulation of the renamed OsTEM1, results in early flowering. In collaboration with Michael Purugganan (NYU), it was found that OsTEM1 was down-regulated during the dry season in rainfed field conditions, further supporting its role in drought responses in rice. These studies suggest novel and crucial roles for TEM/RAVs in adaptive growth. Better performance under salt and drought stress has a great impact

on plant productivity, allows adaptation to environmental adversity and enhances resilience to global climate change.

Selected publications:

1. Matías-Hernández et al., J Ex Bot. 169, 1214-24 (2014).
2. Castillejo, Pelaz, Curr. Biol. 18, 1338–1343 (2008).
3. Osnato et al., Nat. Commun. 3 (2012), doi:10.1038/ncomms1810.
4. Marín-González et al., Plant Physiol. 169, 1214–1224 (2015).
5. Aguilar-Jaramillo et al., Plant J. 100, 522–535 (2019).

CEX2019-000902-S-22-3 – Molecular dissection of the brain of two domestic species from an evolutionary perspective

Principal Investigators: **Marcel Amills and Sebastian Ramos Onsins**

Brain is by far the most complex organ of mammals, showing a striking regional differentiation to fulfill multiple functions related with body homeostasis, perception, locomotion, communication, memory, and behavior. The molecular dissection of the brain would be a fundamental step to understand the genetic basis of many poorly explored behavioral traits (e.g. maternal care, aggression, stress) with huge implications in animal breeding and farming. A comprehensive overview of genes expressed in the mammalian brain categorized on a regional basis has not been done yet in any domestic species, with the only exception of pigs . Elucidating the biology of the brain at a fine transcriptomic scale would be also essential to understand the dramatic behavioral transformations (decreased brain size, tameness, reduced fear and aggression etc) associated with animal domestication. Transcriptomic studies have highlighted differences in the expression of brain genes between dogs and wolves, pigs and wild boars, and domesticated and wild rabbits, but results were not very conclusive . Performance of selection scans comparing domestic species and their wild counterparts have revealed selective sweeps related with traits selected during domestication, such as pigmentation, reproduction and nutritional adaptation , but selection for mutations with causal effects on behavioral traits have been much more difficult to pinpoint. One obvious pitfall of the aforementioned studies is that in all of them selection scans and transcriptome analyses are completely decoupled. This is an important issue because the functional consequences of mutations are not the same in all tissues. Here, we plan to establish an atlas of gene expression of the goat brain and compare it with data already available in pigs, humans and mouse. Moreover, for every brain region under study, we will identify sets of expressed protein-encoding and long non-coding RNA (lncRNA) genes and variation coming exclusively from such genes will be scanned to identify selective footprints. In this way, we will generate independent maps of selective sweeps for every brain tissue in domestic and wild goats and pigs, thus determining whether any brain region is specially enriched in selective sweeps and if different domestication contexts (pigs vs goats) share common selective sweeps in the tissue space. Since we will just use, in each selection scan, sets of SNPs mapping to expressed mRNAs or lncRNAs, the correction for multiple testing will be much less stringent than when using a whole-genome set of SNPs and, in consequence, we will be able to detect genes under selection with increased sensitivity and accuracy. The student is expected to make a stay at the Roslin Institute under the supervision of Dr Lingzhao Fang in the context of the Goat GTEx project which analyses gene regulation in goat tissues.

Selected publications:

1. Sjöstedt et al. 2020. Science 367:eaay5947
2. Albert et al. (2012). PloS Genet. 8:e1002962, 2.
3. Wang et al. 2014. Annual Rev Anim Biosci. 2: 65-84.
4. Guan et al. 2020. J Anim Sci. Biotechnol. 11: 35.

CEX2019-000902-S-22-4 The role of GELP proteins in plant cell wall remodelling and adaptation to stress

Principal Investigator: **Robertas Ursache**

Abiotic stress conditions, such as nutrient imbalance, temperature stress, drought, osmotic stress and salinity are unfavorable for plant growth. Most plants stop growing upon exposure to severe drought, osmotic stress or salt stress. These changes are tightly correlated with alterations in cell walls which are remodeled not only during normal growth, but also in response to stress. Despite a number of studies providing anatomical and genetic evidence for cell wall remodelling in a wide range of plant species, our knowledge on mechanisms and proteins which are essential for this process, is very limited. In this project, using selected members of GELP (GDSL esterases/lipases) family of proteins, I propose to investigate their role in cell wall remodeling in several Arabidopsis thaliana tissues, including xylem vessels, during normal growth and in response to environmental stress, such as salt stress and drought. Arabidopsis thaliana will be used as a model organism as it is very accessible and has been a favourite object of developmental studies. By comparing available tissue-specific RNA-seqbased datasets, I isolated a group of GELP proteins, as potential candidates to be involved in cellulose-based xylem cell wall modifications based on their specific expression pattern corelating with initial cellulose-based xylem cell wall thickening. Why GELPs? GELPs belong to a family of poorly described proteins containing a conserved GDSL (Glycine-AspartateSerine-Leucine) motif at their N-terminus. Plant GELPs have flexible enzymatic sites, allowing a broad substrate binding and acting as multifunctional enzymes. They have been shown to be important for a number of physiological and biochemical processes essential for plant growth and development, organ formation and lipid metabolism . Recently, I demonstrated an example of the essential function of particular GELPs in transesterification of suberin monomers in Arabidopsis root to build a functional suberin barrier for selective uptake of water and nutrients. While some of the members of the same family are required for suberin polymerization, few other members have been shown to act as esterases and capable of degrading suberin . The purpose of this project is not only to discover the function of selected GELPs in cell wall remodeling, but also to establish new tools for manipulating the cell wall properties and cross-linkages between different cell wall polymers.

Selected publications:

1. Jang, G. & Choi, Y. D. Drought stress promotes xylem differentiation by modulating the interaction between cytokininand jasmonic acid. Plant Signal Behav 13, e1451707 (2018).
2. Sharma, N. K., Gupta, S. K., Dwivedi, V. & Chattopadhyay, D. Lignin deposition in chickpea root xylem under drought. Plant Signaling & Behavior 15, 1754621 (2020).
3. Ramachandran, P., Augstein, F., Nguyen, V. & Carlsbecker, A. Coping With Water Limitation: Hormones That Modify Plant Root Xylem Development. Front. Plant Sci. 11, 570 (2020).
4. Su, H.-G. et al. Genome-Wide Identification, Evolution, and Expression of GDSL-Type Esterase/Lipase Gene Family in Soybean. Front. Plant Sci. 11, 726 (2020).
5. Gao, C. Genome engineering for crop improvement and future agriculture. Cell 184, 1621–1635 (2021).