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TÍTULO PROYECTO : SYSTEMS BIOLOGY TO DISSECT EARLY STEPS OF THE NITRATE SIGNALING PATHWAY IN ARABIDOPSIS THALIANA.		
DISCIPLINA PRINCIPAL : BIOLOGIA MOLECULAR		
GRUPO DE ESTUDIO : BIOLOGIA 3		
INVESTIGADOR(A) RESPONSABLE : RODRIGO ANTONIO GUTIERREZ ILABACA		
DIRECCIÓN :		
COMUNA :		
CIUDAD : Santiago		
REGIÓN : METROPOLITANA		

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INFORME FINAL
PROYECTO FONDECYT REGULAR

MODIFICACIONES ACADÉMICAS

El informe no presenta modificaciones académicas.

PROJECT RESULTS:

Describe the results of your research in reference to its original and/or modified Project objectives.

The maximum extension of this section is 5 pages (Arial or Verdana font, size 10).

The long-term goal of our research is to understand how plants sense and respond to changes in nitrogen (N) availability. Nitrate is the most important N source for plants, particularly in agricultural soils. Nitrate availability is one of the most important factors that limit plant growth and agricultural productivity worldwide. Besides its nutritional role in plants, nitrate and other N forms can also act as signals to regulate the expression of hundreds of genes causing modulation of growth and developmental processes. Although genes and processes affected by changes in external or internal nitrate have been identified by us and other researchers around the world, the molecular mechanisms involved in nitrate sensing and signaling networks are still poorly understood.

In this proposal, we hypothesized nitrate signaling in *Arabidopsis* involves a receptor (e.g. NRT1.1/NPF6.3) that upon sensing activates a pathway that results in increased cytosolic free Ca^{2+} . Transient increase in cytosolic free Ca^{2+} activates kinases/phosphatases that change phosphorylation status of target proteins to regulate nitrate responses. Ca^{2+} is a ubiquitous second messenger in all eukaryotes and has been implicated in plant signaling pathways. However, its role in nitrate signaling was not addressed in detail prior to this work. We demonstrated the role of Ca^{2+} as a second messenger and evaluated global changes in protein phosphorylation patterns to understand early steps of the nitrate-signaling pathway of *Arabidopsis thaliana*. We used a combination of tools including molecular genetics, proteomics, transcriptomics and integrative bioinformatics approaches to identify new components and targets of the nitrate-signaling pathway. Below we provide a summary of results obtained for each Aim. Please note figures cited below are included in an annex file.

Aim 1. To use cellular and molecular tools to demonstrate that calcium acts as a second messenger in the signaling pathway activated by nitrate in *Arabidopsis thaliana*.

Understanding the signal transduction pathway involved in the nitrate response has been a long sought-after goal in plant research. Our laboratory and other groups around the world identified regulatory components involved in nitrate responses. However, we still have limited understanding of what happens between nitrate perception and plant responses such as changes in gene expression. Ca^{2+} is a key essential second messenger in signal transduction pathways in plants. In this aim, we addressed the role of calcium as a second messenger in the nitrate-signaling pathway of *Arabidopsis*. We showed Ca^{2+} is a second messenger in the nitrate signaling pathway of *Arabidopsis*. Using aequorin reporter plants, we showed in this proposal that nitrate treatments transiently increase cytoplasmic Ca^{2+} concentration. We found nitrate also induces cytoplasmic concentration of inositol 1,4,5-trisphosphate (IP₃). Increases in IP₃ and cytoplasmic Ca^{2+} levels in response to nitrate treatments were blocked by U73122, a pharmacological inhibitor of phospholipase C, but not by the nonfunctional phospholipase C inhibitor analog U73343. In addition, increase in cytoplasmic Ca^{2+} levels in response to nitrate treatments was abolished in mutants of the nitrate transporter NITRATE TRANSPORTER 1.1/*Arabidopsis thaliana* NITRATE TRANSPORTER 1 PEPTIDE TRANSPORTER FAMILY 6.3 (NRT1.1/NPF6.3). Gene expression of nitrate-responsive genes was severely affected by pretreatments with Ca^{2+} channel blockers or phospholipase C inhibitors. These results indicate that Ca^{2+} acts as a second messenger in the nitrate signaling pathway of *Arabidopsis*. Our results suggest a model where NRT1.1/AtNPF6.3 and a phospholipase C activity mediate the increase of Ca^{2+} in response to nitrate required for changes in expression of prototypical nitrate-responsive genes. Additional details regarding this aim were published in Riveras et al (2015) *Plant Physiology*.

Aim 2. To use cutting-edge proteomics tools to analyze global changes in protein concentration and phosphorylation patterns in response to nitrate treatments.

It is well established that many signaling pathways require modulation of protein abundance, localization or activity by changes in their phosphorylation patterns. In order to identify new regulatory factors involved in nitrate signaling in plants, in this Aim we generated a detailed phosphoproteomics profile with subcellular and temporal resolution in response to nitrate treatments in *Arabidopsis*.

Quantitative phosphoproteomic profile of Arabidopsis-roots in response to nitrate treatments.

As a first step to identify new regulatory factors involved in early signaling events in response to nitrate treatments, we performed quantitative time-course analyses of the *Arabidopsis* root phosphoproteome in response to nitrate treatments using chromatography coupled to tandem mass spectrometry (LC MS/MS). We grew *Arabidopsis* Col-0 plants for 14 days hydroponically, with a full nutrient solution containing 1 mM ammonium as the sole N source and were then treated with 5mM KNO₃ or 5mM KCl as control. Three independent biological replicates were performed for each treatment condition. A label-free high-performance liquid chromatography-tandem mass spectrometry (HPLC-MS/MS) method was applied to identify changes in phosphorylation levels at 0, 5 and 20 min after nitrate or control treatments. These time points were chosen because previous studies described rapid transient and persistent phosphorylation of proteins involved in N-starvation and resupply. Moreover, these experimental conditions allow for comparison with our transcriptomics studies using comparable plant material and growth conditions.

Total proteins from each sample were isolated and digested by trypsin. Phosphorylated proteins were identified after phosphopeptide enrichment using cerium oxide and subsequent HPLC-MS/MS analyses. The spectra were assigned to specific peptide sequences by MASCOT software. After normalization, phosphoprotein abundance was quantified by spectral counting. Using this experimental strategy, we identified and measured 2,048 phosphoproteins, based on 7,502 unique phosphopeptides. Most phosphopeptides were phosphorylated at a unique site (Figure 1A) and the relative distribution of each phosphorylated residue serine, threonine, or tyrosine (Figure 1B) was consistent with prior phosphoproteomic studies. As shown in Figure 1C & 1D, we identified phosphorylated proteins across several cellular functions and subcellular compartments based on Gene Ontology (GO) classification. No overrepresented GO categories were observed in comparison with the *Arabidopsis* genome, showing that our experimental strategy did not bias for protein functions or subcellular localization.

The phosphorylation levels of phosphopeptides corresponding to seventy-two proteins were found altered by nitrate treatments at 5 minutes, 70% of which (50 of 72) were induced (Figure 2A). A similar pattern was observed at 20 min, where nitrate increased levels for 78% of the phosphoproteins (138 out of 178). Only seven proteins increased their phosphorylation levels in response to nitrate at 5 and 20 min indicating a temporally separated phosphoproteome response to nitrate. The same trend was observed for down-regulation of phosphopeptides in our experiments (Figure 2B). These results indicate rapid and long-term phosphoproteomic changes in response to nitrate are mostly positive (i.e. increased phosphorylation) and include at least two components, a transient and rapid phosphorylation response followed by slower phosphorylation changes at later time points. Integrating phosphoproteomics and transcriptome time-course data in response to nitrate treatments revealed that the nature of the phosphoproteins in our data set differs significantly from genes implicated in transcriptome studies thus far. Only 2% of the genes coding for identified phosphorylated proteins were regulated by nitrate at the mRNA level. These results, identified many new components of the nitrate-response acting at the post-translational level that were previously unknown.

The new proteins identified in this Aim include signaling factors such as kinases and transcription factors. It also identified a number of proteins involved in protein binding, transporter activity and hormone metabolism. Although these initial experiments were performed using total protein fraction, we identified proteins across several cellular functions (Figure 3A & 3C) and subcellular compartments (Figure 3B & 3D) based on Gene Ontology classification (GO) and manual inspection. Based on the completeness and novelty of these results, we decided to shift our focus to functional analysis to identify interesting candidates for further studies rather than perform additional proteomics analysis.

In the rapid response (5 min), we found DNA or RNA binding, transcription factors and kinase activity overrepresented categories ($p < 0.05$) (Figure 3A). Consistent with these results, the cellular compartment "nucleus" was also enriched in nitrate-responsive phosphoproteins at 5 min (Figure 3B). Further analysis for components of signaling pathways and regulatory processes revealed that proteins involved in post-translational modifications, regulation of transcription and hormone signal transduction changed phosphorylation levels in response to nitrate at 5 min. These included kinases, calmodulin-binding receptor-like cytoplasmic kinase, G-signaling proteins and several transcription factors belonging to different families and hormone regulatory proteins. Interestingly, the auxin efflux-carriers PIN2 (At5g57090) and PIN4 (At2g01420) were found to be phosphorylated and changing phosphorylation levels in response to nitrate, potentially connecting

the effect of nitrate on auxin fluxes and plant developmental responses (see below for validation experiments).

Within the slow phosphorylation response to nitrate treatments (20 min), we observed different functional patterns as compared to the fast response. Transporter activity, protein binding, and hydrolase activity were overrepresented categories (Figure 3C). Moreover, the proteins differentially phosphorylated were enriched in cytoplasm and plasma membrane categories (Figure 3D). These results are consistent with the late response affecting transport and metabolic processes rather than signaling as observed at 5 min. Among known transport proteins we found the high affinity nitrate-transporter NRT2.1 (At1g08090), amino acid transporter (At1g47670), carbohydrate transporter (At2g25520), ammonium transporter AMT1-3 (At3g24300), K⁺-transporters KT1 (At4g32650) and a K⁺ Efflux antiporter (At1g01790) with increased levels of phosphorylation in response to nitrate after 20 min of treatment. Several phosphorylation sites potentially involved in nitrate transport and metabolism have been described by other groups. For instance, phosphorylation of Ser-501 in NRT2.1 was increased after 20 min of nitrate treatment. This site is localized in a C-terminal phosphorylation hotspot (Phosphat4.0) and was recently identified in nitrate-deprivation experiments with opposite phosphorylation response. Another example includes phosphorylation sites of AMT1.3 at Thr-464 and Ser-487 we found up-regulated by nitrate at 20 min. These sites have been described as important to regulate AMT1.3 transport. Furthermore, nitrate strongly promoted phosphorylation of the nitrate reductase NIA2 (AT1G37130) at the highly conserved and regulatory residue Ser-534 which is targeted by the kinase SnRK1. These observations validate our results and show our phosphoproteomics data provide many interesting new candidates for post-translational modification at the regulatory and metabolic level for future studies.

Linking nitrate-induced phosphorylation modifications to the NRT1.1/AtNPF6.3 transceptor-dependent signaling pathway

The nitrate transporter NRT1.1 (CHL1/NPF6.3) is the only known nitrate sensor in plants. In order to evaluate the role of this sensor in the phosphoproteome we profiled roots of *nrt1.1-null* mutant (*chl1-5*) plants treated with 5 mM KNO₃ or KCl as control. We measured transient changes in phosphorylation response to nitrate (5 min) using the same experimental design described in the previous section. We identified sixty-five phosphoproteins that change its phosphorylation levels in response to nitrate at 5 minutes. As expected, most of these phosphoproteins (43 of 65) were "up-regulated" following a similar pattern than the nitrate-responsive phosphorylation in *wild-type* plants (WT). Surprisingly, there was a very small overlap of the nitrate-phosphoproteome in WT plants and in *chl1-5* mutant (Figure 4A & 4B). Although 74% of the phosphoproteins were detected in both datasets, regulation by nitrate was only detected in wild-type plants. This result is consistent with our signaling model and highlights the importance of NRT1.1 (CHL1/NPF6.3) in the nitrate-signaling pathway.

In order to evaluate the functional impact of NRT1.1, we analyzed the functional categories represented among nitrate-responsive phosphoproteins in wild-type or *chl1-5* plants. Phosphoproteins in each dataset were functionally classified according to MapMan (Thimm et al., 2004), and the relative abundance of phospho-proteins within each functional category (secondary MapMan bins) was expressed as z-scores to identify specific signaling pathways or other processes that require a functional NRT1.1 (Figure 4). As shown in Figure 4C, we detected no significant changes in phospho-proteins involved in auxin signaling transduction, RNA-processing-splicing and phosphatases in *chl1-5* mutant. This result points to these biological processes as relevant regulatory processes downstream of NRT1.1 in the nitrate signaling-pathway.

Aim3. To validate new regulatory components in the nitrate signaling network.

The proteomics data generated in the previous Aim provided novel insights into the nitrate signaling pathways as well as the downstream targets that change gene expression. In this Aim we focus and validate three candidate proteins identified in Aims 1 and 2. Because of their central importance to nitrate signaling and biological process in the plant we chose to follow up on the role of PLC (particularly PLC2), PIN2 and AGO1 proteins. PLC2 was found differentially phosphorylated in our phosphoproteomics experiments. Moreover, PLC2 overexpressor lines showed root hair phenotypes which lead us to investigate the link between nitrate and root hair development (Canales et al 2017). In addition to PLC2, we also include other members of the family due to the fact we found *AtPLC1*, *AtPLC2*, *AtPLC4* and *AtPLC5* gene expression induced in response to nitrate treatments in root organs. We spent the last year and a half of our proposal preparing a myriad of genetic constructs to test the role of these PLCs in nitrate responses. We are currently evaluating the role of this phosphorylation, subcellular localization and possible

interaction with NRT1.1 transceptor in the nitrate response. Below we provide detailed results obtained for PIN2 and AGO1 protein phosphorylation.

(1) The role of PIN2 in the nitrate response of root system architecture.

Auxin is a key plant hormone involved in many aspects of growth and developmental responses. We and others, have shown auxin mediates root developmental responses to nitrate. Interestingly, auxin transport gene ontology functional categories were found overrepresented in our network analysis of nitrate-responsive phosphopeptides. Among the differentially expressed phosphopeptides, we identified the auxin efflux-carriers PIN2 as an attractive candidate to mediate signaling pathways and root developmental responses to nitrate. This auxin transporter showed decreased phosphorylation levels in response to nitrate treatments under our experimental conditions. To understand the function of PIN2 in the nitrate response, we first evaluated the nitrate response in a *pin2* loss-of-function mutant (*eir1.1*, Roman *et al.*, 1995). We subjected wild-type plants and the mutant *eir1-1* to nitrate treatments and evaluated root system architecture (RSA). Nitrate-treated wild-type plants develop shorter primary roots as compared to control-treated plants as described in previous studies from our laboratory (Figure 5). However, primary root length of *eir1-1* plants was not affected by nitrate treatments (Figure 5). These results indicate that PIN2 is important in modulating primary root growth in response to nitrate treatments in *Arabidopsis thaliana*. We also analyzed the number of initiating and emerging lateral roots using light microscopy in wild-type and *eir1-1* mutant plants after nitrate treatments. Nitrate treatments increased the density of lateral roots (both initiating and emerging) as compared with the control condition (KCl treatment) in wild-type plants. However, the lateral root response was altered in the *eir1-1* mutant, with decreased density of emerging and initiating lateral roots as compared to the wild-type phenotype. These findings indicate PIN2 also has a role in lateral root response to nitrate and highlight the importance of this auxin efflux-carrier for modulation of RSA in response to nitrate treatments.

To evaluate the impact of PIN2 phosphorylation in the nitrate response, we evaluated the impact of Ser439 phosphorylation by performing phospho-null and phospho-mimetic substitutions in the mutant *eir1-1* background. We prepared mutant versions of PIN2 with S439A (phosphorylation-deficient) or S439D (phosphorylation-mimetic) substitutions, and we transform *eir1-1* mutant plants. We evaluated the RSA response to nitrate in these plants as described above. As a control, we also complemented the *eir1-1* mutant with the wild-type version of PIN2 which rescued the RSA nitrate response defect of the *eir1-1* mutation (Figure 6). Interestingly, PIN2^{S439A} mutants partially rescued the RSA phenotype. Primary root growth was partially inhibited by nitrate treatments in PIN2^{S439A} plants as compared to wild-type plants (Figure 6). Similarly, lateral root density was also increased in nitrate-treated PIN2^{S439A} plants, with a similar response to wild-type plants. In contrast, primary root length and lateral root density after nitrate treatment was no different from control treatment in PIN2^{S439D} mutant plants. This phosphomimetic substitution abolished nitrate modulation of RSA in a similar manner to what observed in *eir1-1* mutant plants (Figure 6). This result suggests phosphorylation of PIN2 affects PIN2 function. Moreover, these results show that phosphorylation status of PIN2 in this residue is important for normal RSA response to nitrate in *Arabidopsis*.

(2) Function of ARGONAUTE1 phosphorylation in response to nitrate in *Arabidopsis thaliana*.

ARGONAUTE1 (AGO1) protein plays an important role in miRNA-mediated silencing. AGO1 binds directly to miRNAs and catalyze cleavage of target mRNAs leading to their degradation or translational repression of target mRNAs. We found AGO1 protein differentially phosphorylated in *Arabidopsis* roots after nitrate treatments. We identified a phosphorylation site at the Ser1001 residue and showed that AGO exhibited reduced phosphorylation in this residue in response to nitrate treatments. Interestingly, this serine residue is located right at the end of the PIWI domain, an RNase H-like domain that mediates the RNA cleavage activity of AGO1. This serine is conserved in the closely related AGO5 and AGO1 orthologues in other plants but not in mammalian Argonaute proteins. Although, this AGO1 phospho-site has been identified in several phosphoproteome studies in plants and this Ser residue is conserved throughout land plants, there is no information about its impacts on AGO1 activity or function. However, since phosphorylation is known to modulate protein activity, localization, or interaction, we hypothesized that it might play a role in AGO1 function and nitrate responses in plants.

To understand the function of AGO1 in general and specifically in the context of the nitrate response, we investigated morphological and molecular phenotypes in AGO1 phospho-null and

phospho-mimetic mutant plants under normal and nitrate-treatment conditions. We mutated the Ser1001 residue by site-directed mutagenesis to alanine (S1001A) or aspartic acid (S1001D). *AGO1* cDNA containing the S1001A or S1001D mutation was fused to 2000 base pairs of the endogenous *AGO1* promoter and introduced these constructs into the *ago1* null mutant (*ago1-1*). *Ago1-1* null mutant plants have severe morphological and developmental phenotypes with greatly disturbed plant architecture. Following an apparently normal embryo development, *ago1-1* mutant seedlings are characterized by their dark green hypocotyl and their unexpanded pointed cotyledons. The expression of wild-type *AGO1* cDNA into *ago1-1* mutants rescued the developmental defects. Interestingly, *AGO1*^{S1001A} and *AGO1*^{S1001D} cDNA complementation partially rescued *ago1-1* plants. This aminoacid change, S1001A or S1001D, did not affect *AGO1* mRNA or protein levels.

Considering the essential role that miRNAs play throughout plant growth and development, we analyzed developmental phase transitions during the entire plant life cycle in *AGO1*^{S1001A} and *AGO1*^{S1001D} mutants. Interestingly, mutants in which Ser-1001 was changed to alanine shows smaller shoot growth and a delay in flowering time but no alteration at previous developmental stages. On the other hand, phospho-mimetic *AGO1* produced fewer seeds than wild type and develop abnormal siliques, suggesting that *AGO1* phosphorylation has a role on fertility (Figure 7).

Since we found nitrate altered the phosphorylation pattern of *AGO1*, we evaluated the impact of *AGO1*^{S1001A} and *AGO1*^{S1001D} point-mutations for RSA response to nitrate. We observed an exacerbated primary root growth under N-limiting conditions in *AGO1*^{S1001D} plants as compared to *AGO1*^{S1001A} phospho-null mutants (Figure 8). Analysis of lateral root density in response to nitrate also showed defects for both S1001D and S1001A substitutions. These results indicate *AGO1* phosphorylation is important for proper root developmental response to nitrate under our experimental conditions (Figure 8).

To understand the molecular mechanism underlying these developmental phenotypes, we first investigated the impact of these mutations on miRNA biogenesis and *AGO1* loading. We found no significant differences in miRNA abundance nor *AGO1*-binding (Figure 9) at the sRNA transcriptome level. These results indicate *AGO1* sRNA loading and sRNA abundance are not affected by the S1001 mutations. Figure 10 shows selected microRNA examples measured by northern blot.

The most studied function of *AGO1* is arguably its slicer/RNase activity. Because the serine residue under scrutiny here is located right at the end of the catalytic domain, there is a possibility that this phosphorylation might impact *AGO1* slicer activity. To address this question, we examined *AGO1* slicer activity *in vitro*, using immunopurified *AGO1* from wild-type, *AGO1*^{S1001A} or *AGO1*^{S1001D} plants using *PHV* or *AP2* transcripts as tests (targets of miR165 and miR172 respectively). Figure 11 shows preliminary results obtained with this challenging assay in our laboratory. Based on this preliminary result, the phosphorylation identified in this proposal would impact *AGO1* catalytic activity. Additional experiments are underway to verify this highly significant result.

Small RNAs such microRNAs associate with *AGO* proteins and regulate cellular processes and plant responses to a broad of external stimuli, and therefore it is not surprising that these proteins are highly regulated. Our knowledge about miRNAs and its roles in the nitrate signaling pathway is still limited. Our results contributed to understand the impact of global post-transcriptional/translational modifications for an environmental response mediated by RISC.

COOPERACIÓN INTERNACIONAL

N° Proyecto: 1141097
Nombre Colaborador (a) Extranjero (a): SIOBHAN BRADY
Afiliación Institucional Actual: UNIVERSITY OF CALIFORNIA, DAVIS
Fechas de estadía **Desde :**21/09/2017 **Hasta :**26/09/2017

Describe las actividades realizadas y resultados obtenidos. Destaque su contribución al logro de los objetivos del proyecto. Si es pertinente, indique las publicaciones conjuntas generadas, haciendo referencia a lo informado en la etapa Productos. Agregue en la etapa anexos la información necesaria.

Dr. Siobhan Brady participated in the Seminar series: Eukaryotic Gene Regulation & Functional Genomics organized at P. Universidad Católica de Chile (September 22th, Santiago, Chile). Dr. Brady also participated in the Molecular Biosystems Conference: Eukaryotic gene Regulation and Functional Genomics, in Puerto Varas. This conference was a satellite activity to the Annual Meeting of the Chilean Society of Biochemistry and Molecular Biology (23 -25 september -2017). She talked about Transcriptional regulation of nitrogen metabolism. We had numerous discussions about our respective complementary projects.

N° Proyecto: 1141097
Nombre Colaborador (a) Extranjero (a): BRENDA ANDREWS
Afiliación Institucional Actual: UNIVERSITY OF TORONTO
Fechas de estadía **Desde :**23/09/2017 **Hasta :**30/09/2017

Describe las actividades realizadas y resultados obtenidos. Destaque su contribución al logro de los objetivos del proyecto. Si es pertinente, indique las publicaciones conjuntas generadas, haciendo referencia a lo informado en la etapa Productos. Agregue en la etapa anexos la información necesaria.

Dr. Brenda Andrews, participated in the Molecular Biosystems Conference: Eukaryotic gene Regulation and Functional Genomics, in Puerto Varas. This conference was a satellite activity to the Annual Meeting of the Chilean Society of Biochemistry and Molecular Biology. Dr. Brenda Andrews gave a keynote lecture entitled: Global mapping of genetic interactions reveals wiring diagrams of regulatory pathways and networks. She also gave a talk at the Annual Meeting of the Chilean Society of Biochemistry and Molecular Biology.

N° Proyecto: 1141097
Nombre Colaborador (a) Extranjero (a): JAVIER PAZ-ARES RODRIGUEZ
Afiliación Institucional Actual: CONSEJO SUPERIOR DE INVESTIGACIONES CIENTIFICAS/ CENTRO NACIONAL DE BIOTECNOLOGIA
Fechas de estadía **Desde :**25/11/2016 **Hasta :**02/12/2016

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Dr. Paz-Ares participated in the International Plant Biology Course at P. Universidad Católica de Chile (November 25th, Santiago, Chile). We used the same format as described for Dr. Benkova, but his presentations focused on Plant Nutrition, specifically phosphate nutrition. Dr. Paz-Ares also participated in the XI Conference of Plant Biology, organized by the Chilean Society for Plant Biologists (28 November -2 December 2016, Chillán). He delivered a plenary talk at the conference entitled "Genome wide analysis of target of PHOSPHATASE STARVATION RESPONSE REGULATOR 1 provides novel insights on transcription factor function and on plant nutrient physiology".

PRODUCTOS

ARTÍCULOS

Para trabajos en Prensa/ Aceptados/Enviados adjunte copia de carta de aceptación o de recepción.

Nº : 1
Autor (a)(es/as) : Undurraga, S.; Ibarra, C.; Fredes, I.; Gutiérrez, RA.
Nombre Completo de la Revista : Journal of Experimental Botany
Título (Idioma original) : Nitrate signaling and early responses in Arabidopsis roots
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Undurraga_2017.pdf
https://servicios.conicyt.cl/sial/index.php/investigador/f4_articulos/descarga/10548659/1141097/2017/106622/1/

Nº : 2
Autor (a)(es/as) : Armijo, G.; Gutiérrez, RA.
Nombre Completo de la Revista : Molecular Plant
Título (Idioma original) : Emerging players in the nitrate signaling pathway
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Nº : 3
Autor (a)(es/as) : Canales, JC.; Contreras-López, O.; Álvarez, JM.; Gutiérrez, RA.
Nombre Completo de la Revista : Plant Journal
Título (Idioma original) : Nitrate induction of root hair density is mediated by TGA1/TGA4 and CPC transcription factors in Arabidopsis thaliana
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Nº : 4
Autor (a)(es/as) : Gras, DE.; Vidal, EA.; Undurraga, SF.; Riveras, E.; Moreno, S.; Domínguez-Figueroa, J.; Alabadi, D.; Blázquez, M.; Medina, J.; Gutiérrez, RA.
Nombre Completo de la Revista : Journal of Experimental Botany
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OTRAS PUBLICACIONES / PRODUCTOS

N° : 1

Autor (a)(es/as) : Pastenes L, Valdivieso C, Di Genova A, Travisany D, Montecino M, Orellana, A.; González, M.; Gutiérrez, RA.; Allende, M.; Maass, A.; Mendez, M.

Título (Idioma original) : Global gene expression analysis provides insight into local adaptation to geothermal streams in tadpoles of the Andean toad *Rhinella spinulosa*

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N° : 2

Autor (a)(es/as) : Orlando Contreras-Lopez; Tomás Moyano, Daniela Soto, Rodrigo Gutiérrez

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CONGRESOS

Nº : 1

Autor (a)(es/as) : Gutiérrez, RA.

Título (Idioma original) : Phylogenomics and Systems Biology Approaches Reveals Conserved Adaptive Processes in Atacama Desert Plants

Nombre del Congreso : The Plant and Animal Genome (PAG)

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Nº : 2

Autor (a)(es/as) : Gutiérrez, RA.

Título (Idioma original) : Systems Biology to Dissect Nitrogen Responses in Arabidopsis thaliana.

Nombre del Congreso : 1st Latin American Workshop and Conference on Systems Biology

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N° : 3
Autor (a)(es/as) : Gutiérrez, RA.
Título (Idioma original) : Systems Biology to Dissect Nitrogen Responses in Arabidopsis thaliana.
Nombre del Congreso : Genomica funcional de Plantas 2017
País : ARGENTINA
Ciudad : Rosario
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N° : 4
Autor (a)(es/as) : Gutiérrez, RA.
Título (Idioma original) : Plant Nitrogen Responses and Chilean Natural Laboratories
Nombre del Congreso : XXII Congreso de la Sociedad Española de Fisiología Vegetal
País : ESPANA
Ciudad : Barcelona
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Nº : 5
Autor (a)(es/as) : Gutiérrez, RA.; Undurraga, S.; Soto, D.; Varala, K.; Eshel, G.; Araus, EV.; Nilo, R.; Díaz, F.; Carrasco, G.; Corazzi, G.
Título (Idioma original) : Phylogenomics and Systems Biology approaches reveals conserved adaptative processes in Atacama Desert plants
Nombre del Congreso : SEB, Annual Meeting 2017
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Nº : 6
Autor (a)(es/as) : Fredes, I.; Vega, A.; O'Brien, J.; Álvarez, JM.; Gutiérrez, RA.
Título (Idioma original) : Identifying the role of ARGONAUTE1 phosphorylation in the nitrate response in Arabidopsis thaliana
Nombre del Congreso : Molecular Biosystems? Conference on Eukaryotic Gene Regulation and Functional Genomics
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Nº : 7
Autor (a)(es/as) : Zapata-Romero, V.; Gutierrez, RA.

Título (Idioma original) : Marchantia polymorpha as a model system to dissect the Nitrogen Signaling Pathway in Plants
Nombre del Congreso : Molecular Biosystems? Conference on Eukaryotic Gene Regulation and Functional Genomics
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Nº : 8
Autor (a)(es/as) : Gutiérrez, RA.
Título (Idioma original) : Transcriptional regulatory networks in the nitrate response of Arabidopsis thaliana
Nombre del Congreso : Molecular Biosystems? Conference on Eukaryotic Gene Regulation and Functional Genomics
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Nº : 9
Autor (a)(es/as) : Fredes, I.; Vega, A.; Hernández, C.; Gutiérrez, RA.
Título (Idioma original) : IDENTIFYING THE ROLE OF ARGONAUTE1 PHO SPHORYLATION IN THE NITRATE RESPONSE IN Arabidopsis thaliana
Nombre del Congreso : XII Reunión de Biología Vegetal
País : CHILE

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Autor (a)(es/as) : Vega, A.; Agurto, M.; Dussarrat, T.; Canessa, P.; Gutiérrez, RA.
Título (Idioma original) : UNCOVERING PLANT HORMONE SIGNALING IN NITRATE - DEFENSE RESPONSE INTERACTION IN TOMATO
Nombre del Congreso : XII Reunión de Biología Vegetal
País : CHILE
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Nº : 11
Autor (a)(es/as) : Moyano, T.; Vidal, EA.; De Daruvar, A.; Gutiérrez, RA.
Título (Idioma original) : A PAIRWISE PROBABILISTIC FRAMEWORK TO INFER FUNCTIONAL GENE NETWORKS AND IDENTIFY KEY GENES IN RESPONSE TO PERTURBATIONS
Nombre del Congreso : XII Reunión de Biología Vegetal
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Nº : 12

Autor (a)(es/as) : Carrasco-Puga, G.; Hernandez-Castro, C.; Diaz, F.; Soto, D.; Latorre, C.; Gutierrez, RA.

Título (Idioma original) : INTERANNUAL PLANT BIODIVERSITY ESTIMATED FROM SOIL DNA AND SEEDLING EMERGENCE IN THE ATACAMA DESERT

Nombre del Congreso : XII Reunión de Biología Vegetal

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Autor (a)(es/as) : Serrano, A.; Meyer-Reguero, C.; Moyano, T.; Contreras, R.; Arce-Johnson, P.; Gutiérrez, RA.

Título (Idioma original) : SUGAR AND RIPENING, A TRANSCRIPTOMIC ANALYSIS TO UNDERSTAND THE REGULATORY PROCESS OF GRAPE BERRY DEVELOPMENT

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Nº : 14

Autor (a)(es/as) : Ibarra-Henriques, C.; Riveras, E.; Álvarez, JM.; Vega, A.; Gutiérrez, RA.

Título (Idioma original) : THE ROLE OF ATPLCS IN THE NITRATE SIGNALING PATHWAY OF Arabidopsis thaliana ROOTS

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Nº : 15

Autor (a)(es/as) : Zapata-Romero, V.; Gutierrez, RA.

Título (Idioma original) : Marchantia polymorpha AS A MODEL SYSTEM TO DISSECT THE NITROGEN SIGNALING PATHWAY IN PLANTS

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Título (Idioma original) : Hoffmannseggia doelli: CHARACTERIZATION OF AN EXTREMOPHILE PLANT FROM ATACAMA DESERT
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N° : 17
Autor (a)(es/as) : Armijo, G.; Medina, MP.; Kraiser, T.; Gras, D.; Zuniga, A.; González, B.; Gutiérrez, RA.
Título (Idioma original) : INTERACTION BETWEEN Arabidopsis thaliana AND Sinorhizobium meliloti FOR IMPROVED PLANT GROWTH AND NITROGEN NUTRITION
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Nº : 18
Autor (a)(es/as) : Undurraga, S.; Soto, D.
Título (Idioma original) : ECOLOGICAL GENOMICS IDENTIFIES PROCESSES REQUIRED FOR ADAPTATION IN ATACAMA DESERT
Nombre del Congreso : XII Reunión de Biología Vegetal
País : CHILE
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Nº : 19
Autor (a)(es/as) : Undurraga, S.; Daniela, Soto.
Título (Idioma original) : PHYLOGENOMICS AND SYSTEMS BIOLOGY APPROACHES REVEAL CONSERVED ADAPTIVE PROCESSES IN ATACAMA DESERT PLANTS
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Nº : 20
Autor (a)(es/as) : Undurraga, S.; Soto, D.
Título (Idioma original) : A PAIRWISE PROBABILISTIC FRAMEWORK TO INFER FUNCTIONAL GENE

NETWORKS AND IDENTIFY KEY GENES IN RESPONSE TO PERTURBATIONS

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Autor (a)(es/as) : Zapata-Romero, V.; Gutierrez, RA.

Título (Idioma original) : Marchantia polymorpha AS A MODEL SYSTEM TO DISSECT THE NITROGEN SIGNALING PATHWAY IN PLANTS

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Autor (a)(es/as) : Fredes, I.; Vega, A.; Hernández, C.; Gutiérrez, RA.

Título (Idioma original) : IDENTIFYING THE ROLE OF ARGONAUTE1 PHOSPHORYLATION IN THE NITRATE RESPONSE IN Arabidopsis thaliana

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Nº : 23
Autor (a)(es/as) : Aguilar, M.; Carrasco, G.; Díaz, F.; Latorre, C.; Gutiérrez, RA.
Título (Idioma original) : Hoffmannseggia doelli: CHARACTERIZATION OF AN EXTREMOPHILE PLANT FROM ATACAMA DESERT.
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Nº : 24
Autor (a)(es/as) : Armijo, G.; Medina, MP.; Kraiser, T.; Gras, D.; Zuniga, A.; González, B.; Gutiérrez, RA.
Título (Idioma original) : INTERACTION BETWEEN Arabidopsis thaliana AND Sinorhizobium meliloti FOR IMPROVED PLANT GROWTH AND NITROGEN NUTRITION
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Autor (a)(es/as) : Undurraga, S.; Soto, D.

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Nº : 26

Autor (a)(es/as) : Hernandez, C.; Carrasco-Puga, G.; Diaz, F.; Latorre, C.; Soto D.; Gutierrez, R.

Título (Idioma original) : INTERANNUAL PLANT BIODIVERSITY ESTIMATED FROM SOIL DNA AND SEEDLING EMERGENCE IN THE ATACAMA DESERT

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TESIS/MEMORIAS

N° : 1
Título de Tesis : Riqueza potencial de una comunidad vegetal andina del desierto de Atacama
Nombre y Apellidos del(de la) Alumno(a) : Gabriela Carrasco
Nombre y Apellidos del(de la) Tutor(a) : Rodrigo Gutiérrez ; Claudio Latorre
Título Grado : Pregrado
Institución : Pontificia Universidad Católica de Chile
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Ciudad : Santiago
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N° : 2
Título de Tesis : Marchantia polymorpha como modelo de estudio de nitrógeno en plantas
Nombre y Apellidos del(de la) Alumno(a) : Valentina Zapata
Nombre y Apellidos del(de la) Tutor(a) : Rodrigo Gutiérrez
Título Grado : Pregrado
Institución : Pontificia Universidad Católica de Chile
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N° : 3
Título de Tesis : Estudio de la asociación de bacterias con plantas y sus efectos positivos sobre el crecimiento vegetal
Nombre y Apellidos del(de la) Alumno(a) : María Paz Medina
Nombre y Apellidos del(de la) Tutor(a) : Rodrigo Gutiérrez
Título Grado : Pregrado
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N° : 4
Título de Tesis : Nitrato regula el crecimiento a través del proceso de endoreplicación en Arabidopsis thaliana
Nombre y Apellidos del(de la) Alumno(a) : Sebastian Moreno
Nombre y Apellidos del(de la) Tutor(a) : Rodrigo Gutiérrez
Título Grado : Doctorado
Institución : Pontificia Universidad Católica de Chile
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N° : 5
Título de Tesis : Modelamiento de estados del transcriptoma de Arabidopsis thaliana en el desarrollo y frente a perturbaciones
Nombre y Apellidos del(de la) Alumno(a) : Tomas Moyano
Nombre y Apellidos del(de la) Tutor(a) : Rodrigo Gutiérrez
Título Grado : Doctorado
Institución : Pontificia Universidad Católica de Chile
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Tesis_Tomas_Moyano.pdf
https://servicios.conicyt.cl/sial/index.php/investigador/f4_tesis_memorias/descarga/10548659/1141097/2017/82103/1/

Nº : 6
Título de Tesis : Function of ARGONAUTE1 phosphorylation in response to nitrate in Arabidopsis thaliana
Nombre y Apellidos del(de la) Alumno(a) : Isabel Fredes
Nombre y Apellidos del(de la) Tutor(a) : Rodrigo Gutiérrez
Título Grado : Doctorado
Institución : Pontificia Universidad Católica de Chile
País : CHILE
Ciudad : Santiago
Estado de Tesis : En Ejecución
Fecha Inicio : 02/03/2015
Fecha Término :
Envía documento en papel : no
Archivo Asociado :
Isabel_Fredes.pdf
https://servicios.conicyt.cl/sial/index.php/investigador/f4_tesis_memorias/descarga/10548659/1141097/2017/82104/1/

Nº : 7
Título de Tesis : Identification of PLC(s) involves in the nitrate-indices singling pathway in Arabidopsis thaliana roots
Nombre y Apellidos del(de la) Alumno(a) : Catalina Ibarra
Nombre y Apellidos del(de la) Tutor(a) : Rodrigo Gutiérrez
Título Grado : Doctorado
Institución : Pontificia Universidad Católica de Chile
País : CHILE
Ciudad : Santiago
Estado de Tesis : En Ejecución
Fecha Inicio : 01/03/2016
Fecha Término :
Envía documento en papel : no
Archivo Asociado :
Informe_Tesis_Catalina_Ibarra.pdf
https://servicios.conicyt.cl/sial/index.php/investigador/f4_tesis_memorias/descarga/10548659/1141097/2017/82113/1/

N° : 8
Título de Tesis : Caracterización de la respuesta a Nitrato de *Marchantia polymorpha*
Nombre y Apellidos del(de la) Alumno(a) : Susan Hitschfeld
Nombre y Apellidos del(de la) Tutor(a) : Rodrigo Gutiérrez; Fernan Federici
Título Grado : Doctorado
Institución : Pontificia Universidad Católica de Chile
País : CHILE
Ciudad : Santiago
Estado de Tesis : En Ejecución
Fecha Inicio : 01/03/2016
Fecha Término :
Envía documento en papel : no
Archivo Asociado :
Estado_avance_de_tesis_Susan_Hitschfeld.pdf
https://servicios.conicyt.cl/sial/index.php/investigador/f4_tesis_memorias/descarga/10548659/1141097/2017/82123/1/

N° : 9
Título de Tesis : Modulación de la exportación nuclear de transcritos durante la respuesta a nitrato en *Arabidopsis thaliana*
Nombre y Apellidos del(de la) Alumno(a) : Alejandro Fonseca
Nombre y Apellidos del(de la) Tutor(a) : Rodrigo Gutiérrez
Título Grado : Doctorado
Institución : Pontificia Universidad Católica de Chile
País : CHILE
Ciudad : Santiago
Estado de Tesis : En Ejecución
Fecha Inicio : 01/03/2016
Fecha Término :
Envía documento en papel : no
Archivo Asociado :
Informe_de_avance_Alejandr_Fonseca.pdf
https://servicios.conicyt.cl/sial/index.php/investigador/f4_tesis_memorias/descarga/10548659/1141097/2017/82124/1/

N° : 10
Título de Tesis : Características estructurales diferencialmente conservadas en *atNPF6.3* (NRT1.1/CHL1) determinan su función en la señalización por nitrato.
Nombre y Apellidos del(de la) Alumno(a) : Jonathan Morales
Nombre y Apellidos del(de la) Tutor(a) : Rodrigo Gutiérrez
Título Grado : Doctorado
Institución : Pontificia Universidad Católica de Chile
País : CHILE
Ciudad : Santiago
Estado de Tesis : En Ejecución
Fecha Inicio : 01/01/2018

Fecha Término :

Envía documento en papel : no

Archivo Asociado :

resumen_tesis_jmorales_20180205.pdf

https://servicios.conicyt.cl/sial/index.php/investigador/f4_tesis_memorias/descarga/10548659/1141097/2017/83039/1/

ANEXOS

Nº : 1

Archivo Asociado : Informe_Seguimiento_Etico_Bioetico__1141097.pdf

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Nº : 2

Archivo Asociado : Final_Report_Figures.key_.pdf

https://servicios.conicyt.cl/sial/index.php/investigador/f5_anexos/descarga/10548659/1141097/2017/83698/

Nº : 3

Archivo Asociado : Informe_Difusion_FONDECYT_1141097.pdf

https://servicios.conicyt.cl/sial/index.php/investigador/f5_anexos/descarga/10548659/1141097/2017/83842/

A continuación se detallan los anexos físicos/papel que no se incluyen en el informe en formato PDF.

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