Developmental processes regulated by small RNAs during Arabidopsis-Root knot nematode interaction. F.E. DÍAZ-MANZANO1, J. CABRERA1, M. BARCALA2, R. OLMO1, A.C. SILVA3, M.F. ANDRÉS2, I. MARTÍNEZ1, V. RUIZ-FERRER1, C. FENOLL1, C. ESCOBAR1. 1Facultad de Ciencias Ambientales y Bioquímica, Universidad de Castilla-La Mancha, Avenida de Carlos III, s/n, 45071, Toledo, Spain. 2Departamento Protección Vegetal, Instituto Ciencias Agrarias-CSIC, Calle Serrano, 115, 28006, Madrid, Spain. E-mail: Fernando.Diaz@uclm.es

Root knot nematodes (Meloidogyne spp.) currently cause major agricultural losses. They infect plants in the root elongation zone and penetrate intracellularly into the vascular cylinders, inducing galls containing nematode feeding cells, the giant cells (GCs). We studied the differential transcriptome of Arabidopsis GCs and galls after Meloidogyne spp. Infection, as compared to vascular cells, revealing high-repressed genes probably due to gene expression reprogramming during their differentiation. Sequencing of small RNAs (sRNAs) showed profiles consistent with a role of sRNAs in gene silencing. The 24 nt-sRNAs, known to be involved in epigenetic regulation, were highly induced in early formed galls (3 d post infection), and together with differentially regulated miRNAs could be mediating the large gene repression that occurs during early development of GCs/galls. We have studied the roles of miR390 and miR172, accumulated in galls at early infection stages. Loss of function Arabidopsis lines for both miRNAs showed reduced numbers of galls after nematode infection. The two miRNAs participate in plant developmental processes in different ways. TAS3 precursor is cleaved by miR390 triggering tasiRNAs biogenesis that inhibits ARF2-4, releasing repression of lateral root growth. In contrast, miR172 downregulates the AP2-like genes during flowering via a translational mechanism rather than by mRNA cleavage. We discuss the putative molecular networks induced by plant-nematodes in this biotic interaction through miR172 and miR390.

Role of the PTC1 protein phosphatase in stress response in Fusarium oxysporum f. sp. lycopersici. P.P. FERREIRA LEMOS and C. HERA. Departamento de Genética. CampusUniversitario de Rabanales. Universidad de Córdoba. 14071 Córdoba, Spain.

Type 2c Ser/Thr phosphatases (PTCs) are a class of protein phosphatases, conserved in eukaryotes. The PP2C proteins are involved in the regulation of many cellular functional processes, addressed by their role on MAPK cascades. Seven putative PTC proteins have been identified in Fusarium oxysporum f. sp lycopersici 4287, using the BLAST algorithm, with PTCs from Saccharomyces cerevisiae and Fusarium graminearum. The expression of these genes in different stress conditions and plant infection was evaluated by RT-qPCR. Upregulation of ptc1 was observed after cell wall stress, osmotic stress and plant infection, while downregulation was detected after invasive growth. A mutant strain, Δptc1, was obtained by the split marker strategy. The Δptc1 strain was more sensitive to SDS (0.125%) and menadione (20 μg mL−1) than the wild type, indicating possible roles of PTC1 in, respectively, cell wall/membrane stress (MPK1 pathway) and oxidative stress (HOG1 pathway). In addition, the Δptc1 strain showed greater tolerance to LiCl (0.15M and 0.30M) on different media pH (5, 7 and 8.5) than wild type, suggesting a role of PTC1 in lithium efflux mediated by the Nha1 (Na+/K+/Li+/Rb+ antiporter). The phosphorylation level of HOG1, MPK1 and FMK1 proteins was evaluated by western-blot; the Δptc1 strain showed greater phosphorylation level of HOG1 compared to the wild type. These results suggest an important role of PTC1 on the HOG1 pathway of Fusarium oxysporum f. sp lycopersici.

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Transientsilencing of the FaWRKY1 strawberry gene (Fragaria x ananassa) in fruit induces resistance to Colletotrichum acutatum infection. J.J. HIGUERA-SOBRINO1, F.J. MOLINA-HIDALGO1, I. ARJONA-GIRONA2, F. AMIL-RUIZ1, J. GARRIDO-GALA1, A. LEKHBOU1, J.A. MERCADO2, F. PLIEGO-ALFARO3, J. MUÑOZ-BLANCO4, C.J. LÓPEZ-HERRERA2, J.L. CABALLERO1. 1Departamento de Bioquímica y Biología Molecular, Edif. Severo Ochoa-C6, Plantain Baja-Ala Norte. Campus de Rabanales s/n. Universidad de Córdoba-14071, Córdoba, Spain. 2Departamento de Protección de Cultivos, Instituto de Agricultura Sostenible, C.S.I.C. C/Alameda del Obispo s/n, Apartado 4084, Córdoba, Spain. 3Departamento...
Anthracnose, caused by Colletotrichum acutatum, is responsible for significant yield losses in commercial strawberry production worldwide. For this reason, it is of interest to uncover the molecular basis underlying this strawberry/pathogen interaction. Previously, FaWRKY1 was identified as an important element mediating defence responses. This gene encodes an AtWRKY75-like transcription factor (type IIc), which is upregulated in strawberry following C. acutatum infection. In this study, Agrobacterium-mediated transient transformation was used to both silence and overexpress the FaWRKY1 gene in fruit, with the aim to clarify its function in the strawberry defense mechanism. Analyses of FaWRKY1-RNAi strawberry fruits showed resistance to C. acutatum infection, 5 d after inoculation with this pathogen. Overexpression of this gene in strawberry fruit showed increased susceptibility to C. acutatum. Molecular analysis is being carried out with these fruit samples to elucidate candidate genes transcriptionally regulated by FaWRKY1. Furthermore, in vitro DNA-binding assays have revealed a tentative consensus sequence [G/T][T/C]TGAC[T/C], containing the core sequence TGAC (W box), as the likely target sequence for FaWRKY1 binding. These analyses will strengthen genome-wide promoter target site prediction for FaWRKY1.

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Evidence that the putative movement protein (MP2) of Broad bean wilt virus 1 is a pathogenicity determinant. C. CARPINO1,2, I. FERRIOL1, L. ELVIRA-GONZÁLEZ1, L. RUBIO1,3, E. PERI2, S. DAVINO2,4, L. GALIPIENSO1,3,4.

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Broad bean wilt virus 1 (BBWV-1, genus Fabavirus, family Secoviridae) infects crops of economic importance, such as broad bean, pepper, tomato, spinach, and ornamental plants. The virus genome is constituted by two molecules of positive single stranded RNA, each encoding a polyprotein which is further processed by proteolytic cleavage. RNA1 encodes the proteins involved in viral replication and expression, while RNA2 encodes the movement protein (MP) and two coat proteins (LCP and SCP). RNA2 contains an alternative second start codon rendering a smaller putative movement protein, called MP2. To date, the BBWV-1 proteins related to pathogenicity are unknown. The roles of MP2 in symptom determination, post-transcriptional gene silencing (PTGS) and elicitation of hypersensitive response (HR) were examined. Expression of MP2 in Nicotiana benthamiana through Potato virus X (PVX) caused necrotic lesions, indicating that MP2 is a symptom determinant. Analysis of O$_2$ accumulation and necrosis staining revealed that this protein elicited the cellular HR. Transient expression of MP2 in N. benthamiana 16C, that constitutively expresses Green Fluorescent Protein (GFP), and a complementation assay with a vector based on Turnip crinkle virus sequence (TCV-sGFP) showed that this protein acts as a suppressor of PTGS.

Chlorophyll degradation pathway is linked to stomatal and photosynthetic dysfunctions observed in oats resistant to powdery mildew. G. MONTIL-LA-BASCÓN1, M. ROCA2, L.A.J. MUR3, PRATS E1.

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Cost of resistance is usually associated with the energy and nutritional penalties linked to induction of defenses. Currently, a mechanistic understanding of the sources of these costs is lacking, other than vague