whereas a putative D-cysteine desulfhydrase coding gene was found. Approx. 4,000 tetracycline-resistant colonies from an available Tn5 random insertion mutant bank were screened to find phenotypes defective in some of the traits mentioned above. A collection of 80 mutants were selected, including 34 showing reduced (or no growth) or colour change in medium supplemented with copper, ten impaired in biofilm formation, 18 unable to grow or with altered morphology in YEM medium, and 18 displaying reduced or no production of phytase. The molecular characterization of these mutants is currently being performed to identify the affected genes and to determine their involvement in (endophytic) colonization, biocontrol performance, and rhizosphere survival of strain PICF7.

This research is supported by grant P12-AGR667 (Junta de Andalucía, Spain), co-funded by the ERDF of the UE. Thanks are due to Antonio Valverde for this excellent technical assistance.

Characteristics of the biocontrol rhizobacterium *Pseudomonas chlororaphis* PCL1606. S. TIENDA, C. VIDA, A. DE VICENTE , F.M. CAZORLA. *Instituto de Hortofruticultura Subtropical y Mediterránea "La Mayora" (IHSM-UMA-CSIC), Departamento de Microbiología, Facultad de Ciencias, Universidad de Málaga, Spain. E-mail: sandratienda@uma.es*

The major disease affecting avocado crops in the Mediterranean area is white root rot, caused by *Rosellinia necatrix*. The biocontrol rhizobacterium *Pseudomonas chlororaphis* PCL1606 has been isolated from rhizospheres of healthy avocado trees, growing in an area affected by white root rot. As a main characteristic, PCL1606 showed strong *in vitro* antagonism against *R. necatrix* and other important soil-borne pathogens, mainly due to the production of the antimicrobial compound 2-hexyl, 5-propyl resorcinol (HPR). Production of other antifungal compounds has also been detected. PCL1606 persists and colonizes avocado roots, closely interacting and colonizing hyphae of *R. necatrix*, leading to negative effect on the fungus. These phenotypes, acting together, allowed PCL1606 to display biocontrol activity towards *R. necatrix* in avocado plants. We have observed that PCL1606 shows no plant growth promoting activities. The availability of the complete genome sequence of PCL1606 will allow identification of additional features involved in biocontrol by this bacterium.

This work is supported by National plan I+D+I MINECO (AGL2014-52518-C2-1-R; MINECO, Spain), and co-funded by FEDER (EU). S. Tienda is funded by a grant from FPI program MINECO.

Characterization of new mycoviruses in *Fusarium oxysporum* f. sp. *dianthi*. A.T. TRENAS [1,2], M.C. CAÑIZARES[2], A. VALVERDE-CORREDOR[1], C.G. LEMUS-MINOR[1], M.D. GARCÍA-PEDRAJAS[2], E. PÉREZ-ARTÉS[1]. *[1]Depto. Protección de cultivos, Instituto de Agricultura Sostenible, Consejo Superior de Investigaciones Científicas (IAS-CSIC), Alameda del Obispo s/n, 14004 Córdoba, España. [2]Depto. Protección de plantas, Instituto de Hortofruticultura Subtropical y Mediterránea "La Mayora", Universidad de Málaga-Consejo Superior de Investigaciones Científicas (IHSM-UMA-CSIC), 297550 Algarrobo-Costa, Málaga, España. E-mail: a.torres.trenas@csic.es.*

Mycoviruses that cause hypovirulence are potential biocontrol agents of their fungal hosts. In previous research, we characterized FodV1, a chryso-like mycovirus found in isolate Fod116 of *Fusarium oxysporum* f. sp. *dianthi* (Fod). The transference of FodV1 to a new Fod recipient isolate evidenced the induction of hypovirulence in the fungal host. We have analysed the prevalence of FodV1 as well as the incidence and diversity of mycoviral dsRNAs in a collection of 300 Fod isolates. RT-PCR using total RNA extracts and specific primers for the RdRp segment of FodV1, and subsequent sequence analysis, showed that mycovirus FodV1 was present in only three additional Fod isolates. Cellulose column chromatography analysis showed the presence of other dsRNA molecules in 40 isolates. These dsRNAs corresponded to at least five banding patterns, characteristic of different viral families, and three of them were selected for further characterization. Partial sequence data indicated that a monopartite 2.5 kb mycovirus corresponds to a mitovirus, and that a cuatripartite mycovirus shows high homology with Aspergillus foetidus dsRNA mycovirus, and probably corresponds to a new member of the family Alternaviridae. A third monopartite 9.5 kb mycovirus (FodV2) has been almost fully sequenced. This shows high homology with a number of previously described hypoviruses. To determine the putative hypovirulent nature of

FodV2, we transferred it by hyphal anastomosis to a new hygR-tagged recipient isolate, and analysed its effect on some hypovirulence-associated phenotypic traits. Results obtained indicated that FodV2 does not induce hypovirulence in its fungal host.

This research was supported by the Project AGL 2013-48980-R, from the Spanish Ministry of Economy and Competitiveness, co-funded by the European Union (FEDER funds).

Role of the gluconic acid production by the rhizobacterium *Rahnella aquatilis* in pH regulation and biocontrol of the vascular wilt fungus *Fusarium oxysporum*. D. PALMIERI[1], F. DE CURTIS[1], D. VITULLO[1], A. DI PIETRO[2], G. LIMA[1], D. TURRÀ[2]. [1]*Department of Agricultural, Environmental and Food Sciences, University of Molise, Via De Sanctis snc - 86100 Campobasso, Italy.* [2]*Department of Genetics, University of Cordoba, Campus Rabanales, Ed. Gregor Mendel - 14071 Cordoba, Spain. E-mail: davide.palmieri@studenti.unimol.it*

pH affects all aspects of life. Microbes have evolved efficient mechanisms of ambient pH adaptation and modification. In plant rhizospheres, secretions from roots promote the proliferation of microbes, which can alter the pH of this ecological niche. Previous research revealed that rhizosphere pH acts a key factor during infection of the vascular wilt fungus *F. oxysporum* f. sp. *lycopersici* (*Fol*) on its host plant tomato (*Solanum lycopersicum*). While non-infected roots acidify the extracellular environment, infection by *Fol* results in marked root alkalinization, which promotes fungal pathogenicity. We studied the role of pH modification by the soil-inhabiting Gram-negative bacterium *Rahnella aquatilis* (*Ra*) in its interaction with *Fol* in the tomato rhizosphere. Co-inoculation of tomato roots with *Ra* provided efficient protection from vascular wilt caused by *Fol*. *Ra* produced strong extracellular acidification, both in artificial media and in the tomato rhizosphere, most likely through production of gluconic acid from glucose through the enzyme glucose dehydrogenase (Gcd). Preventing rhizosphere acidification by *Ra*, either through application of a buffer solution or by targeted deletion of the bacterial Gcd gene, led to loss of the biocontrol activity against *Fol*. These results suggest that extracellular pH regulation plays a key role in the interaction between bacteria and fungi in the rhizosphere, with important consequences for plant health.

This research was supported through project BIO2013-47870-R from the Spanish Ministerio de Innovación y Competitividad (MINECO).

Effects of farnesol production by *Trichoderma* on the development of bean (*Phaseolus vulgaris*). S. MAYO[1], A. RODRÍGUEZ-GONZÁLEZ[1], O. GONZÁLEZ-LÓPEZ[1], A. LORENZANA[1], G. CARRO-HUERGA[1], M.P. CAMPELO[1], S. GUTIÉRREZ[2], P.A. CASQUERO[1]. [1]*Research Group of Engineering and Sustainable Agriculture, Natural Resources Institute, University of León, Av. Portugal 41, 24071 León, Spain.* [2]*Area of Microbiology, Research Group of Engineering and Sustainable Agriculture, University School of Agricultural Engineers, University of León, Ponferrada Campus, Av. Astorga s/n, 24401 Ponferrada, Spain. E-mail: pacasl@unileon.es*

Common bean (*Phaseolus vulgaris*) is the third most important food legume worldwide, surpassed only by soybean and peanut. *Trichoderma* (Teleomorph: *Hypocrea*) is a fungal genus found in the soil. These fungi are secondary, fast growing, opportunistic invasive organisms, which produce enzymes that degrade fungal cell walls, and induce production of compounds with antimicrobial activity. We evaluated the effect of farnesol production of *T. harzianum* (T34) on the development of bean. *In vivo* assays were performed with this isolate and two transformants (T34dpp1.2 and T34dpp1.3) which were overexpressing the *dpp1* gene. Bean seeds were coated with a spore suspension of each *Trichoderma* isolate. They were sown and maintained with a photoperiod of 16 h light, 25°C/16 °C (day/night), and 60% RH. Plants were removed 45 d after sowing, evaluated for: hypocotyl diameter, , root system length,and dry weights of shoots and roots. T34dpp1.3 and control plants (without fungi) were larger than plants inoculated plant with T34, in hypocotlyl diameter, root system length, and shoot dry weight. However, T34 did not present differences in comparison with T34dpp1.3 for root system dry weight root system, but T34dpp1.2 did.

This research was supported by the National project (AGL2012-40041-C02-02) (Ministry of Economy and Competitiveness) and by the Regional project (LE228U14) (Junta de Castilla y León).