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**EVIDENCE FOR THE INVOLVEMENT OF VP37 OF BROAD BEAN WILT VIRUS 1 IN THE INDUCTION OF PLANT SYMPTOMS AND POSTTRANSCRIPTIONAL GENE SILENCING.** C. Carpino<sup>1,2</sup>, I. Ferriol<sup>2</sup>, L. Elivira-Gonzalez<sup>2</sup>, L. Rubio<sup>2,3</sup>, E. Peri<sup>1</sup>, S. Davino<sup>1</sup>, L. Galipienso<sup>2,3,4</sup>. <sup>1</sup>Department of Agricultural and Forestry Science, University of Palermo, Piazza Marina 61, 90133 Palermo, Italy. <sup>2</sup>Instituto Valenciano de Investigaciones Agrarias (IVIA), Ctra. CV-315, 46113 Moncada, Valencia, Spain. <sup>3</sup>Euro-Mediterranean Institute of Science and Technology (IEMEST), Via Michele Miraglia 20, 90139 Palermo, Italy. <sup>4</sup>Departamento de Biotecnología, Escuela Técnica Superior de Ingeniería Agronómica y del Medio Natural, Universidad Politécnica de Valencia, Camino de Vera s/n, 46022 Valencia, Spain. E-mail: salvatore.davino@unipa.it

Broad bean wilt virus 1 (BBWV-1, genus *Fabavirus*, family *Secoviridae*) infects economically important crops (broad bean, pepper, tomato, spinach and ornamental plants) and is worldwide distributed. It is constituted by two positive ssRNA molecules that encode two polyproteins further processed by proteolytic cleavage. RNA-1 encodes proteins necessary for viral replication and expression, whereas RNA-2 encodes a movement protein (MP) and two coat proteins (LCP and SCP). RNA-2 contains an alternative start codon that renders a smaller putative protein (VP37). Until now, BBWV-1 pathogenicity determinants were not identified, and proteins related to symptomatology induction were unknown. In this work BBWV-1 putative VP37 was identified as the main responsible for virus-induced symptomatology in *Nicotiana benthamiana* and broad bean using a BBWV-1 infectious clone and a BBWV-1 mutant. Moreover, VP37 transient expression through a *Potato Virus X* (PVX) vector caused necrotic lesions in *N. benthamiana*, indicating that this protein acts as a pathogenicity determinant. Finally, transient expression of VP37 in *N. benthamiana* 16C, that constitutively expresses the 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 suppressor of post transcriptional gene silencing (PTGS).

Together our results demonstrate that VP37 putative protein is directly involved in the elicitation of BBWV-1 symptomatology in *N. benthamiana* and broad bean, and that this protein is a symptom determinant and acts as gene silencing suppressor.

**IDENTIFICATION OF A MULTIDRUG AND TOXIC EXTRUSION TRANSPORTER INVOLVED IN THE SECRETION OF INDOLE-3-ACETIC ACID AND ITS CONJUGATES IN PSEUDOMONAS SAVASTANOI.** M. Cerboneschi, C. Biancalani, S. Calamai, L. Bini, S. Tegli. Dipartimento di Scienze Produzioni Agroalimentari e dell'Ambiente (DISPAA), Laboratorio di Patologia Vegetale Molecolare, Università degli Studi di Firenze, Via della Lastruccia 10, 50019 Sesto Fiorentino (Firenze), Italy. Email: matteo.cerboneschi@unifi.it

*Pseudomonas savastanoi* (*Psv*) is the causal agent of olive and oleander knot disease, characterised by hyperplastic galls on the aerial parts, and whose development is dependent on a functional type III secretion system (TTSS) and on the expression of *brp* genes. Several phytohormones produced by *Psv*, i.e. the auxin 3-indoleacetic acid (IAA) and the cytokinin trans-zeatin, are also involved in knot formation. *Psv* synthesizes IAA from tryptophan, by the sequential activity of the enzymes IAAM and IAAH encoded by the homonymous genes. In *Psv* the *iaaL* gene is also present, coding for an enzyme for conjugation of IAA to lysine to give IAA-Lys. In the genome of the oleander strain *Psn23*, the *iaaM/iaaH* operon is located close to the *iaaL* gene, which upstream has an ORF encoding a putative multidrug and toxic compound extrusion (MATE) transporter (named *matE*). The IAA-driven inhibition of *brp* genes, as well as data from phenotype microarray analysis on *ΔiaaM*, *ΔiaaL*

and *ΔmatE* mutants, strongly suggested a functional link existing in *Psv* between IAA metabolism, TTSS and MATE. Here, through an integrated approach among bioinformatic analysis, structural biology and site-directed mutagenesis on *Psn23* MATE, and HPLC-MS analysis of bacterial auxin production, we found the transporter encoded by *matE* contributing to this picture, by mediating IAA and IAA-Lys efflux. This is the first report of a MATE responsible for auxin transport in bacteria. Because of its role on IAA homeostasis in *Psv*, these findings could also contribute to the development of novel anti-infectives targeting *Psv* MATE.

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**INTERFERENCES ON THE DETECTION OF XYLELLA FASTIOSA IN POLYGALA MYRTIFOLIA BY REAL-TIME PCR.** V. Chiatti<sup>1,2</sup>, N. Pucci<sup>1</sup>, S. Lucchesi<sup>1</sup>, V. Modesti<sup>1</sup>, M. Reverberi<sup>2</sup>, S. Loreti<sup>1</sup>. <sup>1</sup>Centro di ricerca per la patologia vegetale (CREA-PAV) via C.G. Bertero 22, 00156 Roma. <sup>2</sup>Dipartimento di Biologia Ambientale, Sapienza Università di Roma. E-mail: stefania.loreti@crea.gov.it

*Xylella fastidiosa* (*Xf*) is a harmful quarantine pathogen that colonizes the xylem vessels of a wide range of host plants. Recently introduced in Europe, it occurred in France on *Polygala myrtifolia* and in Apulia (Southern Italy) on *Olea europaea*. The disease named "quick decline syndrome of olive" has been associated with the strain CoDiRO, belonging to the subspecies *pauca* (variant "sequence type 53"). The outbreak on *P. myrtifolia* was instead caused by the subsp. *multiplex*. A national research plan has been established, in order to control the disease and perform monitoring activities in pest-free areas. The scope of our studies was to develop and harmonize diagnostic protocols of *Xf*, mainly on olive – being an economically valuable species – and on *P. myrtifolia*, which is affected by both the subspecies, and is in evaluation as "spy-indicator" plant specie in risky locations to support the early detection of *Xf*. Two different real-time PCR assays (based on TaqMan<sup>TM</sup> and EvaGreen<sup>TM</sup>) were used. The overall results proved the assays to be appropriate for testing olive samples. Further investigation was needed for *P. myrtifolia*, due to ambiguous results obtained with both assays on 38.7% of the samples. To understand the nature of the interference, sequencing of a non specific amplicon obtained by the real-time PCR assay was performed, with no relevant matches found in the BLAST database. The possibility to obtain indeterminate results by real-time PCR has to be considered when official analyses of *P. myrtifolia* are performed, to avoid the risk of misinterpretation of the final results.

**BIOLOGICAL ACTIVITY OF ALKALI PRE-TREATED ARUNDO DONAX EXTRACT TOWARDS DIFFERENT FILAMENTOUS FUNGI.** S. Cianchetta, M. Nota, S. Galletti. Consiglio per la ricerca in agricoltura e l'analisi dell'economia agraria, Centro di ricerca Agricoltura e Ambiente (CREA-AA), Via di Corticella 133, 40128 Bologna, Italy. E-mail: stefania.galletti@crea.gov.it

Giant reed (*Arundo donax* L.) is considered one of the most promising lignocellulosic species for bioenergy in the Mediterranean environment, for its high productivity and low input requirements. In the framework of the AGROENER project, funded by MiPAAF, a study is undergoing on the possibility of exploiting the biomass as substrate for oleagineous microorganisms, after a delignifying pre-treatment, for the production of 2<sup>nd</sup> generation biodiesel. The spectrophotometric analysis of an extract, obtained after alkaline pre-treatment of giant reed dry biomass, revealed the