XXIII Convegno Nazionale
Società Italiana di Patologia Vegetale - SIPaV

In occasione del 25° anniversario dalla fondazione

Piacenza, 4-6 ottobre 2017
Università Cattolica del Sacro Cuore
Via Emilia Parmense, 84

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Programma

Mercoledì 4 ottobre 2017

12.30-14.30	**Registrazione e affissione poster**	
14.30-15.00	Apertura lavori e saluti delle Autorità	
	Prima Sessione - Plant/microbe interaction	**Moderatori**: F. Favaron, E. Baraldi
15.00-15.30	*Relazione su invito* "A POPULATION GENETICS PERSPECTIVE ON PLANT DURABLE RESISTANCE: A CASE STUDY WITH DOWNY MILDEW OF GRAPEVINE"	Francois Delmotte, INRA Bordeaux, Francia
15.30-15.50	AN ECTOMYCORRHIZAL FUNGUS MAY DECREASE THE SUSCEPTIBILITY OF *PINUS SYLVESTRIS* TO THE NATIVE PATHOGEN *HETEROBASIDION ANNOSUM* BUT NOT TO THE EXOTIC *H. IRREGULARE*	***L. Giordano***, E. Zampieri, G. Lione, A. Vizzini, J.V. Colpaert, R. Balestrini, P. Gonthier
15.50-16.10	PROMOTER CHARACTERIZATION OF THE *VviATL156* GENE INVOLVED IN GRAPEVINE RESISTANCE TO *PLASMOPARA VITICOLA*	P. Ariani, D. Danzi, A. Regaiolo, A. Polverari, ***E. Vandelle***
16.10-16.30	STUDY OF THE MOLECULAR DIALOGUE BETWEEN GRAPEVINE INFLORESCENCE/BERRY AND *BOTRYTIS CINEREA* DURING THE INITIAL, QUIESCENT, AND EGRESSION INFECTION STAGES	Z.H. Mehari, S. Pilati, P. Sonego, G. Malacarne, U. Vrhovsek, K. Engelen, P. Tudzynski, M. Zottini, E. Baraldi, ***C. Moser***
16.30-16.50	Pausa caffè	
16.50-17.10	METABOLOMIC AND TRANSCRIPTOMIC PROFILES IN HEALTHY AND ONION YELLOW DWARF VIRUS INFECTED 'ROSSA DI TROPEA' ONIONS	***A. Tiberini***, F. Mercati, F. Araniti, A. Ciampa, G. Micali, S.B. Grand, A. Taglienti, M.R. Abenavoli, M.T. Dell'Abate, F. Sunseri, L. Tomassoli, G. Albanese
17.10-17.30	TRANSCRIPTOME PROFILING OF DIFFERENTIALLY EXPRESSED GENES IN STRAWBERRY AFTER PREHARVEST APPLICATION OF BENZOTHIADIAZOLE AND CHITOSAN	***L. Landi***, R.M. De Miccolis Angelini, S. Pollastro, E. Feliziani, F. Faretra, G. Romanazzi
17.30-17.50	THE pH REGULATION AS A NEW KEY MECHANISM OF BIOCONTROL OF THE VASCULAR WILT FUNGUS *FUSARIUM OXYSPORUM* BY THE RHIZOBACTERIUM *RAHNELLA AQUATILIS*	***D. Palmieri,*** F. De Curtis, D. Vitullo, A. Di Pietro, G. Lima, D. Turrà.
17.50-18.10	Premio SIPaV "Giovanni Scaramuzzi"	
	Opificio delle Idee	
18.10-18.20	Introduzione	M.L. Gullino
18.20-19.20	Presentazione contributi giovani in formazione selezionati	
	DEVELOPMENT OF A PCR-BASED DIAGNOSTIC ASSAY FOR THE DETECTION OF *COLLETOTRICHUM LUPINI* ON LUPIN SEEDS	***B. Caggiano***, D. Da Lio, G. Puntoni, G. Le Floch, R. Baroncelli , S. Pecchia

DECIPHERING THE CROSSTALK BETWEEN THE BIOCONTROL AGENT *TRICHODERMA GAMSII* T6085 AND THE PATHOGEN *FUSARIUM GRAMINEARUM*: A GENOME-WIDE TRANSCRIPTOMIC ANALYSIS	***A. Zapparata***, R. Baroncelli, G. Vannacci, S. Sarrocco
UNRAVELLING THE MECHANISMS OF *COLLETOTRICHUM LUPINI* HOST SPECIALIZATION	***D. Da Lio***, R. Baroncelli, C. Ranaldi, G. Puntoni, G. Vannacci, G. Le Floch, S. Sarrocco
USE OF TaqMan qPCR TO EVALUATE THE COLONISATION RATE OF BUNCH TRASH AND DEVELOPING BERRIES BY *BOTRYTIS CINEREA* IN VINEYARDS	***G. Fedele***, M. Si Ammour, E. González-Domínguez, C. Morcia, V. Terzi, V. Rossi
STUDIES ON KIWIFRUIT DECLINE, AN EMERGING ISSUE EVEN FOR FRIULI VENEZIA GIULIA (EASTERN ITALY)	***F. Savian***, M. Martini, S. Borselli, S. Saro, R. Musetti, N. Loi, G. Firrao, P. Ermacora
FIELD INVESTIGATION ON GARLIC DRY ROT	***L. Mondani***, G. Chiusa, P. Battilani
GRAPEVINE PINOT GRIS DISEASE: EPIDEMIOLOGICAL TRAITS	***G. Tarquini***, M. Martini, G.L. Bianchi, A. Loschi, N. Loi, P. Ermacora
EXTRAGENOMIC SEQUENCES HIGHLIGHT DIFFERENCES WITHIN *FUSARIUM VERTICILLIOIDES* STRAINS ISOLATED FROM ITALIAN *ZEA MAYS* KERNELS	***A. Grottoli***, G. Giuliano, M. Beccaccioli, M. Blandino, W. Sanseverino, R. Aiese Cigliano, V. Scala, M. Reverberi
STUDY ON THE INCREASED INCIDENCE OF *GRAPEVINE PINOT GRIS VIRUS* SYMPTOMS IN A VERMENTINO VINEYARD IN NORTHERN SARDINIA	***N. Schianchi***, V. Prota, G. Moro
COMPETITION FOR NUTRIENTS AND SPACE: A MECHANISM OF ACTION OF *AUREOBASIDIUM PULLULANS* STRAINS	***A. Di Francesco***, M. Mari
CHARACTERIZATION OF PROMOTER SEQUENCES OF *RAPID ALKALINIZATION FACTOR* (*RALF*) GENES IN *FRAGARIA* X *ANANASSA* INTERACTING WITH *COLLETOTRICHUM ACUTATUM*	***F. Negrini***, K. Ogrady, K.M. Folta, E. Baraldi
IN VITRO AND *IN VIVO* DEVELOPMENT OF THE PREDOMINANT MEMBERS OF THE *FUSARIUM HEAD BLIGHT* SPECIES COMPLEX OF WHEAT AND THEIR SECONDARY METABOLITE PRODUCTION	***F. Tini***, G. Beccari, D.M. Gardiner, M. Sulyok, L. Covarelli
CHARACTERIZATION OF THE ROLE OF A *RAPID ALKALINIZATION FACTOR* (*RALF*) GENE IN THE SUSCEPTIBILITY OF STRAWBERRY FRUITS TO *COLLETOTRICHUM ACUTATUM*	***M. Guidarelli***, M.C. Merino, D. de Biase, A. Pession, E. Baraldi

19.20-20.15	Tavole rotonde: Valorizzazione della ricerca	
	Pubblicazioni scientifiche	L. Rubino
	Brevetti	P. Battilani
	Spin-off	V. Rossi
20.15	Cena Buffet	

Giovedì 5 ottobre 2017

	Seconda Sessione: Botanical epidemiology and disease control	**Moderatori:** G. Romanazzi, T. Caffi
9.00-9.30	*Relazione su invito "*MODELLING CROP HEALTH AND CROP LOSSES: CONCEPTS, APPROACHES AND AVENUES*"*	Serge Savary, INRA Toulouse, Francia
9.30-9.50	THE LEAFHOPPER VECTOR *EUSCELIDIUS VARIEGATUS* RESPONDS DIFFERENTLY TO INFECTION WITH TWO PHYTOPLASMAS	***L. Galetto***, S. Abbà, M. Rossi, M. Vallino, M. Pesando, M. Pegoraro, D. Bosco, C. Marzachì
9.50-10.10	POSSIBILITIES OF COPPER REDUCTION IN CONTROL OF TOMATO BACTERIAL DISEASES	G. Giovanale, E. Fortunati, A. Mazzaglia, ***G.M. Balestra***
10.10-10.30	LONG-TERM ORGANIC MANAGEMENT MODIFIES SOIL MICROBIOTA THAT SUPPRESSES SOILBORNE PHYTOPATHOGENIC FUNGI AND VIRUSES	***G. Cesarano,*** M. Minutolo, D. Alioto, F. De Filippis, F. Scala, G. Bonanomi
10.30-11.50	Pausa caffè e Sessione Poster	
11.50-12.10	USE OF A POMEGRANATE PEEL EXTRACT TO CONTROL OLIVE ANTHRACNOSE	***L. Schena***, S. Pangallo, M.G. Li Destri Nicosia, G.E. Agosteo, F.V. Romeo, P. Rapisarda, A. Abdelfattah, S. Mosca, S. Scibetta, S. Minutillo, G. Magnano di San Lio, S.O. Cacciola.
12.10-12.40	ROLE OF ORNAMENTAL RUTACEOUS PLANTS IN LONG-DISTANCE DISPERSAL OF CITRUS QUARANTINE BACTERIA	***V. Catara***, G. Licciardello, A. Urso, P. Bella, G. Timpanaro, P. Caruso.
12.40-13.00	APPLICATION OF *TRICHODERMA* STRAINS AND THEIR BIOACTIVE METABOLITES FOR MORE SUSTAINABLE SOYBEAN PRODUCTION	***R. Marra***, N. Lombardi, G. d'Errico, M. Pascale, F. Lacatena, A. Pironti, A. Bottiglieri, P. Lombari, M. Lorito, S.L. Woo.
13.00-14.30	Pranzo di lavoro	
15.00-15.45	Resoconto Opificio delle Idee	M.L. Gullino
15.45-16.45	Presentazione degli Spin-off censiti dalla SIPaV	
16.45-17.00	Pausa caffè	
17.00-17.30	Publishing with Springer	Zuzana Bernhart, Executive Editor, Springer Nature, Dordrecht, Paesi Bassi
17.30-19.30	Assemblea Generale dei Soci SIPaV	
20.00-23.00	Cena sociale	

Venerdì 6 ottobre 2017

	Terza Sessione: Food safety and mycotoxins	**Moderatori:** A. Moretti, D. Spadaro
9.00-9.30	*Relazione su invito* "MYCOTOXIN AND FOOD SAFETY IN A CLIMATE CHANGE SCENARIO"	Sonia Marin, Universitat de Lleida, Spagna
9.30-9.50	NEW ACQUISITIONS ON THE BIOCONTROL OF FUSARIUM HEAD BLIGHT: FROM ECOLOGY TO CRISPR-CAS GENOME EDITING	***S. Sarrocco,*** L. Malfatti, S. Manfredini, P. Esteban, G. Puntoni, R. Bernardi, M. Haidukowski, A. Moretti, G. Vannacci
9.50-10.10	A PATHOGENICITY CLUSTER FOR EXPLOITING MAIZE KERNELS DEFENCES IN *ASPERGILLUS FLAVUS*	L. Antiga, S. La Starza, C. Miccoli, G. O'Brian, G.A. Payne, S. Xiaomei, S. D'Angeli, C. Fanelli, M.M. Altamura, ***M. Reverberi***
10.10-10.30	*ASPERGILLUS FUMIGATUS* IN ORGANIC SUBSTRATES: A NEW THREAT FOR HUMAN HEALTH	K. Santoro, ***S. Matić***, U. Gisi, D. Spadaro, M. Pugliese, M.L. Gullino
10.30-11.30	Pausa caffè e Sessione Poster	
11.30-11.50	AFLA-PISTACHIO: DEVELOPMENT OF A MECHANISTIC MODEL TO PREDICT AFLATOXIN CONTAMINATION OF GREEK PISTACHIO NUTS	M.D. Kaminiaris, ***M. Camardo Leggieri***, D.I. Tsitsigiannis, P. Battilani
11.50-12.10	YEAST VOLATILE ORGANIC COMPOUNDS REDUCE GROWTH AND OCHRATOXIN A PRODUCTION BY *ASPERGILLUS* SPP.	***M.G. Farbo***, P.P. Urgeghe, Z. Ul Hassan, A. Marcello, S. Jaoua, Q. Migheli
12.10-12.30	*FUSARIUM AVENACEUM*, AN EMERGING PATHOGENIC FUNGUS CAUSING PRE-HARVEST WET APPLE CORE ROT IN TRENTINO REGION	***V. Gualandri***, A. Branz, M. de Concini, G. Angeli
12.30 -13.00	Chiusura lavori	

Volume 99 (Supplement) October 2017

Journal of Plant Pathology

Formerly *Rivista di patologia vegetale* established in 1892

An International Journal of the Italian Society for Plant Pathology

XXIII NATIONAL MEETING OF THE ITALIAN SOCIETY FOR PLANT PATHOLOGY (SIPaV)

Piacenza, October 4-6, 2017

Abstracts of papers

EDIZIONI ETS, Pisa, Italy
Poste Italiane S.p.A. - Spedizione in Abbonamento Postale - D.L. 353/2003
(conv. in L. 27/02/2004 n° 46) art. 1, comma 1 - DCB AREA CENTRO 1/Pisa.

ISSN 1125-4653

The **Società Italiana di Patologia Vegetale, SIPaV, (Italian Society for Plant Pathology)** was established in 1992 following the dissolution of the Italian Society for Crop Protection (SIF) and the Italian Phytopathological Association (AFI). Its main aims are to promote research into different branches of plant pathology, to disseminate knowledge about plant diseases and their aetiological agents and to promote cooperation among experts working in the field of plant pathology, and partnership in fundamental and applied reasearch. The Society organizes meetings, gathers and distributes information about plant diseases, and maintains cooperation with other national and international scientific organizations and with national and local administrative authorities on problems involving plant health management.

The Society publishes a journal (Journal of Plant Pathology), which hosts articles by members and external contributors, a bulletin and other bibliographic material to exchange information among members.

The SIPaV is affiliated to the International Society for Plant Pathology (ISPP) and to the European Foundation for Plant Pathology (EFPP).

COUNCIL (2017-2019)

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PREMIO SIPaV
“GIOVANNI SCARAMUZZI”

PREMIO SIPAV "GIOVANNI SCARAMUZZI"

ON THE ROLE OF PHLOEM PROTEIN IN PLANT-PATHOGEN INTERACTION

L. Pagliari

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Phytoplasmas are prokaryotic plant pathogens mostly restricted to the sieve elements (SEs) of the phloem tissue. Diseases caused by these pathogens result in a huge impact on agriculture. At present, no effective control strategy has been developed, therefore the study of the mechanisms at the basis of plant responses to phytoplasma infection is needed to identify resistance or tolerance sources. Phytoplasmas are strictly dependent on metabolic compounds from their hosts and for this reason many attempts to culture them *in vitro* failed. Moreover, the necessity to control experimental and environmental conditions makes investigations in natural plant hosts particularly limiting. In that respect, the reliability of *Arabidopsis thaliana* as model plant for studying phytoplasma-plant interaction was proved, comparing macroscopic, histological and ultrastructural modifications induced by phytoplasma infection in Arabidopsis with those reported in natural host plants.

It has been supposed that the agglutination of SE protein filaments could limit phloem flow and consequently pathogen spread. Nevertheless, these hypotheses were based on indirect evidences and, up to now, the information about the physiological role of the SE protein filaments are still incomplete even in physiological condition. To elucidate the interaction between phytoplasmas and SE protein filaments, Arabidopsis mutant lines lacking one or two genes related to these proteins (*AtSEOR1*, *AtSEOR2*, *AtPP2-A1*) were used both in healthy and in infected conditions. Various microscopy techniques were performed, in order to combine fresh and embedded tissue observations, gaining information on both ultrastructural and physiological SE modifications. Agglutinated SE protein masses were observed only in infected SEs, supporting the hypothesis about their involvement in plant defence mechanism. Overexpression of *AtSEOR1*, *AtSEOR2* and *AtPP2-A1* genes, analysed by real-time RT-PCR experiments, confirmed these observations. SE protein filaments were observed also in mutant lines, indicating that in case of stressful condition, such as pathogen infection, the presence of *AtSEOR1* and *AtSEOR2* genes is not fundamental, contrary to what previously reported. Nevertheless, only filaments in their wild-type form blocked the phloem mass flow in case of phytoplasma infection.

These data were integrated with molecular results about phytoplasma titre quantification and no correlation between phloem impairment and pathogen concentration was found. The low phytoplasma titre in *AtSEOR1ko* lines indicated the possible involvement of this gene in plant defence mechanism. To see if and how defence mechanisms were activated, analyses of phytohormones, central regulators of plant immunity, were carried out. Results suggested a possible role of jasmonic acid and *cis*-12-oxo-phytodienoic acid in plant defence against phytoplasmas.

SESSION I

PLANT-MICROBE INTERACTION

Keynote Lecture

A POPULATION GENETICS PERSPECTIVE ON PLANT DURABLE RESISTANCE: A CASE STUDY WITH DOWNY MILDEW OF GRAPEVINE

F. Delmotte, F. Fabre, C. Couture, I. Demeaux, S. Richart-Cervera, L. Delière, Y. Dussert and C. Delmas

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Grapevine downy mildew, caused by the obligate biotrophic oomycete *Plasmopara viticola*, is considered to be one of the most destructive grapevine diseases worldwide. This pathogen, which is native to North America, was accidentally introduced into Europe in the 1870s. Grapevine (*Vitis vinifera*) cultivars are highly susceptible to the disease and currently require an intense chemical management programme to control the disease. During the last 20 years, there has been increased effort to develop partially-resistant grapevine varieties in Europe. Experimental systems based on these partially-resistant varieties showed a reduction by almost 90% of fungicides in vineyards. However, we now need to make sure that the efficacy of resistance persists in time despite the constant evolution of pathogen populations. Until recently, little was known about the genetic structure and diversity of *P. viticola* despite its economic importance. In order to provide elements of knowledge on the durability of grapevine resistance, we have assessed the genetic and phenotypic diversity of *P. viticola* populations. In the native area of the pathogen, we have investigated host plant specialization and host range expansion of *P. viticola* by combining a phylogenetic approach with pathogenicity tests. We then reconstructed the pathways of invasion of the pathogen from wild *Vitis* spp. in North America to wine-producing regions worldwide. In Europe, the rapid evolution of aggressive strains eroding partial resistance of grapevine was evidenced by cross-inoculation experiments. The availability of the genome sequence of *P. viticola* allowed to detect candidate genes for the adaptation to host partial resistance. Altogether, these results might have important implications for viticulture, including breeding for resistance and disease management.

SESSION I

ORAL PRESENTATIONS

AN ECTOMYCORRHIZAL FUNGUS MAY DECREASE THE SUSCEPTIBILITY OF *PINUS SYLVESTRIS* TO THE NATIVE PATHOGEN *HETEROBASIDION ANNOSUM* BUT NOT TO THE EXOTIC *H. IRREGULARE*. **L. Giordano[1,2], E. Zampieri[1], G. Lione[1], A. Vizzini[3,4], J.V. Colpaert[5], R. Balestrini[4], P. Gonthier[1].** *[1]University of Torino, Department of Agricultural, Forest and Food Sciences (DISAFA), Largo Paolo Braccini 2, I-10095 Grugliasco (TO), Italy. [2]University of Torino, Centre of Competence for the Innovation in the Agro-Environmental Field (AGROINNOVA), Largo Paolo Braccini 2, I-10095 Grugliasco (TO), Italy. [3]University of Torino, Department of Life Sciences and Systems Biology (DBIOS), Viale P.A. Mattioli 25, I-10125 Torino, Italy. [4]Institute for Sustainable Plant Protection, CNR, Torino Unit, Viale P.A. Mattioli 25, I-10125 Torino, Italy. [5]Universiteit Hasselt, Centrum voor Milieu-Kunde (CMK), Agoralaan Gebouw D, 3590 Diepenbeek, Belgium. E-mail: paolo.gonthier@unito.it*

In the last century, the intensification of global trade has greatly enhanced the likelihood of biological invasions resulting in increasing threats to native ecosystems. The North American root rot agent *Heterobasidion irregulare* Garbel. et Otrosina was accidentally introduced in central Italy in 1944 where the Eurasian congener *H. annosum* (Fr.) Bref. is also present. *H. irregulare* has become invasive colonizing pine and oak stands along 103 km of coastline west of Rome. Although many studies have focused on factors driving biological invasions, very little is known on the role played by native mycorrhizal fungi in modulating host tolerance to non-native pathogens. The aim of this study was to compare the level of susceptibility of *Pinus sylvestris* L. to *H. irregulare* and *H. annosum* between plants mycorrhized and non-mycorrhized with the native ectomycorrhizal fungus *Suillus luteus* (L.) Roussel. Inoculation experiments were performed with three pathogen genotypes per species on seven-month-old mycorrhized and non-mycorrhized seedlings. To assess the level of host susceptibility, seedlings were inspected every day and scored depending on their time to death. The resulting survival curves pointed out that mycorrhizae reduced significantly the level of host susceptibility to the native pathogen, but not to the exotic one. Besides, non-mycorrhized plants were equally susceptible to both pathogens. These findings suggest that a symbiont-mediated mechanism of tolerance might protect the host plant from a native pathogen, but may fail in the presence of a non-native one. In this model system, this mechanism may confer an additional competitive advantage to *H. irregulare*.

This work was supported by the Italian Ministry of Education, University and Research, within the FIRB program (grant number RBFR1280NN).

PROMOTER CHARACTERIZATION OF THE *VviATL156* GENE INVOLVED IN GRAPEVINE RESISTENCE TO *PLASMOPARA VITICOLA*. **P. Ariani, D. Danzi, A. Regaiolo, A. Polverari, E. Vandelle.** *Università degli Studi di Verona, Strada Le Grazie 15, 37135 Verona (VR), Italy. E-mail: elodiegenevieve.vandelle@univr.it*

Despite a lot of efforts to overcome the susceptibility of grapevine (*Vitis vinifera*) to pathogens, in particular *Plasmopara viticola*, by crossing with wild relatives, traditional breeding did not lead so far to significant results. Thus, to allow the development of new plant breeding techniques to improve premium cultivars, we are currently studying the resistance mechanisms evolved in naturally resistant American grapes. We selected *VviATL156* as candidate gene specifically upregulated in the resistant *V. riparia* upon downy mildew infection and generated stable transformed grapevines (*V. vinifera* cv. Shiraz) constitutively expressing it. *VviATL156* encodes an E3-ubiquitin ligase, belonging to the ATL gene family, the closest homolog of Arabidopsis *ATL2*, which is highly responsive to elicitors and hormones. Transgenic grapevines were more resistant to downy mildew. However, knowledge about the specific regulation of candidate genes is crucial to identify a truly promising gene, not severely affected by the genomic background in which it is introduced. In this context, the *VviATL156* regulative regions from both resistant and susceptible grapevine species were cloned and sequenced. Bioinformatics analyses allowed to define the core promoter structures and *cis*-acting element compositions. An extra TATA-box was predicted in the *V. riparia* sequence, together with some over-represented *cis*-acting elements, likely related to disease resistance. The promoters were then functionally characterized in stably transformed *Arabidopsis thaliana* plants, under physiological conditions and in response to hormones and pathogen infection. Moreover, promoter *trans*-activation by specific transcription factors was evaluated in a Dual Luciferase Assay experiment in transiently transformed *Nicotiana benthamiana*.

STUDY OF THE MOLECULAR DIALOGUE BETWEEN GRAPEVINE INFLORESCENCE/BERRY AND *BOTRYTIS CINEREA* DURING THE INITIAL, QUIESCENT, AND EGRESSION INFECTION STAGES. **Z.H. Mehari[1,4,5], S. Pilati[1], P. Sonego[1], G. Malacarne[1], U. Vrhovsek[1], K. Engelen[1], P. Tudzynski[2], M. Zottini[3], E. Baraldi[4], C. Moser[1].** *[1]Research and Innovation Centre, Fondazione Edmund Mach, 38010 San Michele all'Adige (TN), Italy. [2]Institute for Biology and Biotechnology of Plants, Westf. University of Muenster, Muenster, Germany. [3]Università di Padova, Italy. [4]Università di Bologna, Laboratorio di Biotecnologie Vegetali, Bologna, Italy. [5]Ethiopian Institute of Agricultural Research, A.A., Ethiopia. E- mail: claudio.moser@fmach.it*

Grapes quality and yield are affected by gray mould disease caused by the necrotrophic fungus *Botrytis cinerea*. Primary infections are mostly initiated at blooming by air-borne conidia but the fungus often remains quiescent from bloom until maturity, when it causes grey mould. Molecular analyses of the interaction between *B. cinerea* and the flower/berry of grapevine (*Vitis vinifera* L.) were carried out using confocal microscopy plus integrated transcriptomic and metabolic analysis of the host and the pathogen. Open flowers from fruiting cuttings of cv. Pinot Noir were infected with GFP-labeled *B. cinerea* and samples taken at 24 and 96 hours post inoculation (hpi), 4 and 12 weeks post inoculation (wpi) were studied. Penetration of the flower epidermis by *B. cinerea* at 24 hpi coincided with an increased expression of fungal genes encoding virulence factors and induced a rapid defense reaction in the flowers involving genes associated with the accumulation of PR proteins, stilbenoids, reactive oxygen species and cell wall reinforcement. At 96 hpi the transcriptional reaction appeared largely diminished both in the host and in the pathogen. Afterwards, infected berries continued their development without any visible symptom, although the presence of *B. cinerea* could be ascertained. Nonetheless, at the transcriptional level, both the fungus and the hard-green berries displayed to be transcriptionally active. At 12 wpi, the egressed *B. cinerea* expressed almost all virulence and growth-related genes enabling the pathogen to colonize the berries. In response to egression, ripe berries reprogrammed different defense responses, though ineffectively.

METABOLOMIC AND TRANSCRIPTOMIC PROFILES IN HEALTHY AND ONION YELLOW DWARF VIRUS INFECTED 'ROSSA DI TROPEA' ONIONS. **A. Tiberini[1], F. Mercati[2], F. Araniti[1], A. Ciampa[4], G. Micali[1], S.B. Grande[1], A. Taglienti[3], M.R. Abenavoli[1], M.T. Dell'Abate[4], F. Sunseri[1], L. Tomassoli[3], G. Albanese[1].** *[1]Università degli Studi Mediterranea di Reggio Calabria, Dipartimento di AGRARIA, Località Feo di Vito, 89122 Reggio Calabria, Italy. [2]Consiglio Nazionale delle Ricerche, Istituto di*

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A research project SI.ORTO (SIR-MIUR grant – SIORTO-RBSI149LD5) has been activated to evaluate the effects of *Onion yellow dwarf virus* (OYDV, genus *Potyvirus*) on nutraceutical compounds accumulation in onion cv. Rossa di Tropea. This cultivar of southern Italy (Calabria), granted by the European Union with the Protected Designation Origin (IGP) trademark, is known for its mild to sweet flavor and its richness in flavonols and anthocyanins. OYDV was found responsible for an alarming agronomic decline of 'Rossa di Tropea' caused by severe leaf symptoms and stunting. Therefore, a metabolomic and transcriptomic profiling approach was used for analyzing differentially expressed genes (DEGs) and metabolites (DEMs) due to OYDV infection. By gas chromatography-mass spectrometry (GC-MS), a primary metabolism profiling was investigated by comparing healthy vs. OYDV-infected onion samples, collected at three time points (bulb harvesting, leaves drying and after bulbs storage) during an experimental trial conducted in Calabria. Several metabolites, connected with sugar and amino-acid metabolism and the TCA cycle, decreased in OYDV-infected bulbs compared with control plants at the first testing time. A pronounced increase of the same metabolites was then detected. Further, magnetic resonance micro-imaging (MRI) of whole bulbs highlighted a structural alteration of OYDV-infected bulbs as compared with healthy ones. In parallel, on the same material, the whole transcriptome determined with a RNAseq approach showed a mean of *ca*. 26,000 DEGs related to OYDV infection at the three analyzed time points. Preliminary results highlighted a OYDV-mediated modulation of important pathways in agreement with metabolic profiles. This research represents a first and integrated study on metabolic modulation in a plant-virus pathosystem ('Rossa di Tropea' onion–OYDV).

TRANSCRIPTOME PROFILING OF DIFFERENTIALLY EXPRESSED GENES IN STRAWBERRY AFTER PREHARVEST APPLICATION OF BENZOTHIADIAZOLE AND CHITOSAN.

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The application of resistance inducers exploiting natural protection of plants is an alternative approach to the use of synthetic fungicides for the control of pre- and postharvest decay of strawberries. However, the mechanisms behind the specific resistance inducer are not completely understood. The global transcriptional changes in strawberry fruits were investigated, using RNA-Seq technology, 6, 12 and 24 h after preharvest application to the canopy of benzothiadiazole and chitosan. Overall, 5,062 and 5,210 differentially expressed genes (fold change ≥ 2) were identified in the fruits treated with benzothiadiazole and chitosan, respectively, as compared with the control, only 20% of these genes being induced by both elicitors. The functional enrichment analysis highlighted different gene modulation over time for transcripts associated with photosynthesis and heat-shock proteins, according to the elicitor. Several genes associated with the plant immune system, biotic and abiotic stresses, and SAR, were elicited by the treatments. In addition, the reprogramming of protein metabolism was observed in fruits treated with both elicitors, which led to increased storage proteins. The RNA-Seq data were confirmed using RT-qPCR for 12 selected genes. This study established that the two elicitors affect cell pathways associated with plant defences in different ways, and suggests a role for chloroplasts as the primary target in the modulation of the plant defence responses, which actively communicate signals through changes in redox status. The genes identified in the present study can represent markers to better elucidate plant/pathogen/resistance inducer interactions, and to plan innovative sustainable disease management strategies.

THE pH REGULATION AS A NEW KEY MECHANISM OF BIOCONTROL OF THE VASCULAR WILT FUNGUS *FUSARIUM OXYSPORUM* BY THE RHIZOBACTERIUM *RAHNELLA AQUATILIS*.

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Microbes have evolved efficient mechanisms of environmental pH modification for a better growth and adaptation in the rhizosphere. Plant roots exudates promote the proliferation of microbes, which can alter the rhizosphere pH. The rhizosphere pH is a key factor for infection of the vascular wilt fungus *F. oxysporum* f. sp. *lycopersici* (*Fol*) to tomato plants (*Solanum lycopersicum*). In fact, infection by *Fol* results in a marked root alkalinization which promotes fungal pathogenicity, while acidification usually occurs in non-infected roots. Our work investigated the role of pH modification by the soil-inhabiting Gram-negative bacterium *Rahnella aquatilis* (*Ra*) in its interaction with *Fol* in tomato rhizosphere. Co-inoculation of tomato roots with *Ra* provided efficient protection from *Fol*. *Ra* produced strong extracellular acidification, both in artificial media and in tomato rhizosphere, most likely via the production of gluconic acid from glucose by the enzyme glucose dehydrogenase (Gcd). The addition of a specific buffer solution or deletion of the bacterial Gcd gene, both preventing rhizosphere acidification, led to the loss of the biocontrol activity of *Ra* against *Fol*. Our findings suggest that extracellular pH regulation is a crucial factor in the interaction between some biocontrol bacteria and pathogenic fungi in the rhizosphere, with important consequences in biocontrol optimization and plant health.

SESSION “OPIFICIO DELLE IDEE”

YOUNG RESEARCHERS

DEVELOPMENT OF A PCR-BASED DIAGNOSTIC ASSAY FOR THE DETECTION OF *COLLETOTRICHUM LUPINI* ON LUPIN SEEDS. **B. Caggiano[1], D. Da Lio[1], G. Puntoni[1], G. Le Floch[2], R. Baroncelli[2], S. Pecchia[1].** *[1]Università di Pisa, DISAAA-a, Via del Borghetto 80, 56124 Pisa, Italy. [2]University of Western Brittany, LUBEM Technopôle, 29280 Plouzané-Brest, France. E-mail: susanna.pecchia@unipi.it*

Lupins anthracnose is a destructive seed- and airborne disease affecting stems and pods, caused by *Colletotrichum lupini*. The pathogen is a member of the *C. acutatum* species complex, and contrasts with other members of the latter by its host specificity and by its apparent low genetic variability. Primary seed infections as low as 0.01-0.1% can cause very severe infections. Under this conditions, one the most effective management strategies is the development of a robust and sensitive seed detection assay to screen seed lots before planting. PCR-based detection systems exhibit higher levels of sensitivity than conventional techniques but when applied to seed tests they require the extraction of PCR-quality DNA from target organisms in backgrounds of saprophytic organisms and inhibitory seed-derived compounds. To overcome these limitations a new detection protocol for *C. lupini* based on a biological enrichment step followed by a PCR assay was developed. Several enrichment protocols were compared and a 72 h incubation of the seeds with yeast malt broth amended with ampicillin, streptomycin and lactic acid was the most efficient technique to increase *C. lupini* biomass before DNA extraction. A species specific *C. lupini* primer pair was developed, based on rDNA IGS region sequences. The specificity was evaluated against 23 different *Colletotrichum* species and 21 different non target organisms isolated from seeds of *Lupinus albus* cv. Multitalia, *L. luteus* cv. Mister and *L. angustifolius* cv. Tango. The protocol described here enabled the detection of *C. lupini* in samples artificially infected with less than 1/1,000 infected seed.

DECIPHERING THE CROSSTALK BETWEEN THE BIOCONTROL AGENT *TRICHODERMA GAMSII* T6085 AND THE PATHOGEN *FUSARIUM GRAMINEARUM*: A GENOME-WIDE TRANSCRIPTOMIC ANALYSIS. **A. Zapparata, R. Baroncelli, G. Vannacci, S. Sarrocco.** *Università di Pisa, Dipartimento di Scienze Agrarie, Alimentari e Agro-ambientali, Via del Borghetto 80, 56124 Pisa. E-mail: antonio.zapparata@gmail.com*

Fusarium graminearum is one of the most relevant pathogen associated with Fusarium Head Blight (FHB) in Europe, causing yield losses and mycotoxin accumulation on wheat grain. During infection stages *F. graminearum* produces deoxynivalenol (DON, type B trichothecene), a virulence factor and a protein synthesis inhibitor threatening animal and human health. The biocontrol agent *Trichoderma gamsii* T6085 has been successfully applied on wheat crops at anthesis, during two following growing seasons, leading to a reduction of both incidence and severity of FHB. Furthermore, T6085 tolerates high concentration of DON (50 ppm) and shows antagonistic traits against a mycotoxigenic isolate of *F. graminearum*, such as mycoparasitism. A comparative genomic analysis of the genome of T6085, together with other fungal organisms, revealed expansions in gene families related to mycoparasitism, such as glutamic and serine peptidases and specific carbohydrate-degrading enzymes. In this study, we performed a molecular characterization of the crosstalk (before contact) between T6085 and *F. graminearum* through a genome-wide transcriptomic analysis. Mapping reads against the sequenced genomes of both organisms and following downstream statistical analysis allowed the identification of differential expressed genes in self- and nonself-interactions. The results here described will contribute to the development of T6085 as biocontrol agent by elucidating the molecular mechanisms underpinning the crosstalk between this beneficial isolate and the mycotoxigenic *F. graminearum*.

UNRAVELLING THE MECHANISMS OF *COLLETOTRICHUM LUPINI* HOST SPECIALIZATION. **D. Da Lio[1], R. Baroncelli[1,2], C. Ranaldi[1], G. Puntoni[1], G. Vannacci[1], G. Le Floch[2], S. Sarrocco[1].** *[1]Università di Pisa, DISAAA-a, Via del Borghetto 80, 56124 Pisa, Italy. [2]University of Western Brittany, LUBEM Technopôle, 29280 Plouzané-Brest, France. E-mail: daniele.dalio@hotmail.com*

Colletotrichum lupini is a plant pathogen responsible for anthracnose disease on lupin (*Lupinus albus*), where it causes severe symptoms leading to severe yield losses. *C. lupini* is a member of the *C. acutatum* species complex and, differently from the other members of this complex, it possesses a high specificity for its host, making it an interesting evolutionary model for host-pathogen interaction studies. In order to determine the host specificity of *C. lupini* compared to the other related species included in the same complex, an efficient protocol to infect lupin seeds was developed and tested using 30 *Colletotrichum* spp. isolates, including 10 *C. lupini* strains. To follow the colonization process and to establish a host penetration timing, a reference isolate of *C. lupini* (IMI504893) was transformed using *A. tumefaciens* in order to obtain a GFP marked strain. Five transformants were obtained and characterized using both morphological and physiological assays and a phenotypic microarray test (Biolog) in order to detect metabolic differences between the wild type and the GFP marked strains. The selected transformed GFP strain was artificially inoculated on lupin and observed in detail through fluorescence microscopy observations. Additionally, several genomes of *C. lupini*, including the reference and the transformed strains, were sequenced using NGS techniques. Data provided by genome sequencing and the definition of the infection timings will be used for further transcriptomic analyses to extensively define the *C. lupini*/lupin interaction process.

USE OF TaqMan qPCR TO EVALUATE THE COLONISATION RATE OF BUNCH TRASH AND DEVELOPING BERRIES BY *BOTRYTIS CINEREA* IN VINEYARDS. **G. Fedele[1], M. Si Ammour[1], E. González-Domínguez[1], C. Morcia[2], V. Terzi[2], V. Rossi[1].** *[1]Department of Sustainable Crop Production, Facoltà di Scienze Agrarie, Alimentari e Ambientali, Università Cattolica del Sacro Cuore, Via Emilia Parmense 84, 29122 Piacenza, Italy. [2]CREA-GB Consiglio per la ricerca in agricoltura e l'analisi dell'economia agraria-centro di Ricerca Genomica e Bioinformatica, Via San Protaso 302, 29017 Fiorenzuola d'Arda, Italy. E-mail: vittorio.rossi@unicatt.it*

Botrytis cinerea is one of the main grapevine pathogens. The complexity of its life cycle and the existence of different infection pathways has caused growers to rely heavily on routine application of fungicides at four growth stages: flowering (A), pre-bunch closure (B), véraison (C) and before harvest (D). Recently, a weather-driven, mechanistic model was developed to predict the severity of Botrytis bunch rot epidemics. The model was validated against 21 epidemics by using a discriminant function analysis (DFA), with 81% accuracy. The DFA showed that the infection periods predicted by the model during flowering and shortly after flowering had a key role in determining the severity of Botrytis bunch rot on mature bunches. Experiments were then conducted to understand the contribution of early-season infections on the final disease severity. Colonization by *B. cinerea* was assessed in bunch trash and in developing grape berries subjected to different fungicide treatments by using both classical mycological assays and DNA quantification

through PCR. Relative quantification of *B. cinerea* DNA directly from *Vitis vinifera* tissues was performed by duplex qPCR with TaqMan chemistry. The qPCR assay was based on a probe designed on *B. cinerea* intergenic spacer (IGS) region and a probe designed on *V. vinifera* resveratrol synthase gene I. The inhibitory effect of grape DNA on amplification of fungal DNA was successfully evaluated by adding increasing amounts of *B. cinerea* DNA into *V. vinifera* DNA. A fungal colonization rate was then calculated to compare the effect of fungicide treatments performed in A, B, C or D. The obtained results, combined with the epidemiological model, provide new information on how to schedule fungicides treatments for controlling *B. cinerea* in vineyards.

STUDIES ON KIWIFRUIT DECLINE, AN EMERGING ISSUE EVEN FOR FRIULI VENEZIA GIULIA (EASTERN ITALY). F. Savian[1], M. Martini[1], S. Borselli[1], S. Saro[2], R. Musetti[1], N. Loi[1], G. Firrao[1], P. Ermacora[1]. *[1]University of Udine, Department of Agriculture Food Environment and Animal Sciences, Via Delle Scienze 206, 33100 Udine, Italy. [2]ERSA-FVG, Regional Agency for Rural Development of Friuli Venezia Giulia, Via Sabbatini 5, 33050 Pozzuolo, Italy. E-mail: francesco.savian@uniud.it*

A new syndrome affecting kiwifruit (kiwifruit decline, KD) was described for the first time in 2012 in Veneto; in 2014 KD reached Friuli Venetia Giulia and in 2015 Piedmont. The affected plants show a collapsed root system with no feed roots. As a result, after heat waves, the plants go through a sudden, irreversible and fast dieback, usually leading to death in only few weeks. With the aim of understanding the aetiology and some epidemiological traits of KD, two approaches were used. Firstly, the identification of affected fields was carried out by regional territorial agency ERSA-FVG in a survey during 2015 and 2016 summers. We interviewed the farmers, whose fields were affected, to gather information about disease appearance and spread patterns, agricultural practices and orchard characteristics. Secondly, we performed a classic isolation of fungi from roots of affected plants on PDA. Fungal isolates were grouped according to morphological features, and successively identified by molecular tools based on PCR/RFLP, sequencing and BLAST analyses of the ITS region. Genera *Fusarium*, *Ilyonectria* and *Pythium* were identified more frequently, but so far no correlation was found between fungal species and sampling sites. These preliminary results suggest that some opportunistic fungi might be involved in the disease besides abiotic stress factors (especially waterlogging). As further research, we will set up experiments to reproduce the disease symptoms and we will apply remote sensing techniques in the fields for an early and more accurate diagnosis of KD.

FIELD INVESTIGATION ON GARLIC DRY ROT. L. Mondani, G. Chiusa, P. Battilani. *Università Cattolica del Sacro Cuore, Department of Sustainable Crop Production (DI.PRO.VE.S.), Via Emilia Parmense 84, 29122 Piacenza. E-mail: paola.battilani@unicatt.it*

Fusarium proliferatum was signalled worldwide since 2002 as the main causal agent of garlic dry rot. According to literature, other *Fusarium* spp. and nematodes (*Ditylenchus dipsaci*) could contribute to the disease. Moreover, white varieties are reported as more susceptible compared to red ones. Dry rot is considered a postharvest disease, but relevant incidence of symptomatic bulbs at harvest was also reported. The aim of this study was to investigate infection time and agents involved in garlic dry rot from field to table. Field sampling was organised in Piacenza province (north Italy), area of production of white garlic (PGI). Six field units were selected for sampling, three of them with an history of relevant dry rot. Soil was sampled before sowing. Garlic plants were collected in three growth stages: BBCH 15 (5th leaf clearly visible), BBCH 45 (50% of the expected bulb diameter reached), BBCH 49 (dead leaves, dry bulb top, complete growth). Soil serial dilutions and colony forming units (CFU) count were performed, as well as nematode counting. Direct isolation from symptomatic and asymptomatic plants was managed, so as the identification at species level for a selected set of fungal strains. PCR was applied to confirm fungi and nematode identification. *Ditylenchus dipsaci* was not detected in soil in autumn. Regarding fungi, the largely dominant species were *F. proliferatum* and *F. oxysporum*, isolated since BBCH 15, increasing in incidence from early growth stages to harvest. *F. proliferatum* seems confirmed as the candidate most relevant causal agent of dry rot, infecting bulbs early in field.

Work supported by PSR 2014-2020-16.1.01, Focus Area 2A, "Guidelines to control Fusarium dry rot in Piacenza white garlic".

GRAPEVINE PINOT GRIS DISEASE: EPIDEMIOLOGICAL TRAITS. G. Tarquini[1], M. Martini[1], G.L. Bianchi[2], A. Loschi[1], N. Loi[1], P. Ermacora[1]. *[1]Department of Agricultural, Food, Environmental and Animal Sciences. University of Udine. via delle Scienze, 206, I-33100 Udine, Italy. [2]ERSA, Plant Protection Service, via Sabbatini 5, 33050, Pozzuolo del Friuli (UD), Italy; E-mail: paolo.ermacora@uniud.it*

A new *Trichovirus*, named *Grapevine Pinot gris virus* (GPGV) was discovered in 2012 by NGS approach in Pinot gris grapevine with symptoms of chlorotic mottling and leaf deformation. Despite reports are increasing worldwide, the aetiology of GPG disease remains still unclear since the virus was detected both in symptomatic and asymptomatic plants. The aim of this work was to investigate epidemiological traits of GPG disease, considering both its spread in field conditions and its transmission by grafting in controlled conditions. In spring 2014, ninety virus-free Pinot gris cuttings were planted in a vineyard located in Farra d'Isonzo (GO) with high incidence of symptomatic plants and surveyed for symptoms expression for four consecutive vegetative seasons. Field observations revealed that symptoms appear in early spring and after a stage of scarce vegetation plants recover, producing new asymptomatic tissues, which make difficult to identify symptomatic leaves. RT-qPCR analyses were performed at different times of growing season assessing GPGV presence in symptomatic and asymptomatic plants. Results showed that: i) GPGV detection in spring revealed the highest percentage of infected plants; ii) the number of RT-qPCR positive plants decreased in July and September; iii) GPGV-infected plants were mostly asymptomatic (77%), while 23% were symptomatic.

In greenhouse conditions, GPGV-infected scions collected from symptomatic and asymptomatic plants were grafted on virus-free Pinot gris cuttings. Minimum incubation period for symptoms expression was 3 months, symptoms presence and RT-qPCR for GPGV agreed in 13 samples out of 21. Further observations and analyses will be carried out.

EXTRAGENOMIC SEQUENCES HIGHLIGHT DIFFERENCES WITHIN *FUSARIUM VERTICILLIOIDES* STRAINS ISOLATED FROM ITALIAN *ZEA MAYS* KERNELS. A. Grottoli[1], G. Giuliano[1], M. Beccaccioli[1], M. Blandino[2], W. Sanseverino[3], R. Aiese Cigliano[3], V. Scala[4], M. Reverberi[1]. *[1]Department of Environmental Biology, University of Rome "Sapienza", Rome, Italy. [2]Department of Agricultural, Forest and Food Sciences, Università degli Studi di Torino, Italy. [3]Sequentia-Biotech, Barcelona, Spain. [4]CREA-PAV, 00136 Roma, Italy. E-mail: alessandro.grottoli@uniroma1.it*

Fusarium is a genus including ubiquitous plant-pathogenic fungi causing severe crop losses. In the last years, several novel species of

Fusarium have been described *via* molecular phylogenetics analysis; nonetheless, most of these are not formally distinguished. Even due to this uncertainty, *Fusarium* genus is commonly divided into species complexes; the species are grouped by physiological, biological, ecological and genetic similarity. *Fusarium fujikuroi* species complex (FFSC) is one of the largest complexes. Although species within the FFSC complex are closely related, individuals, even within the same species, may have distinct phenotypic traits like mycotoxins production and pathogenicity. In a previous study, bioinformatics analysis performed on an Italian wild type *Fusarium verticillioides* (Sacc.) Nirenberg strain ITEM 10027 (*Fv*) indicated about 2 mega bases of genome, not present on the reference database (*Fv* 7600). We analyzed this "extra" region using an in-house bioinformatics pipeline. We found that gene sequences within this region were highly related to genes of *Fusarium fujikuroi* (Sawada) Wollenw. and other FFSC related species. Gene sequences within this region were used to develop molecular markers and were used to classify about 200 *Fv* isolated from *Zea mays* L. kernels collected from the 2013 crops coming from the main areas of the northern Italian maize cultivated fields. Interestingly, different profiles emerged among the various samples of *Fv* despite these isolates derive frequently from the same location.

STUDY ON THE INCREASED INCIDENCE OF *GRAPEVINE PINOT GRIS VIRUS* SYMPTOMS IN A VERMENTINO VINEYARD IN NORTHERN SARDINIA. N. Schianchi, V. Prota, G. Moro. *Università degli Studi di Sassari, Dipartimento di Agraria, Via De Nicola 9, 07100 Sassari. E-mail: nschianchi@uniss.it*

Grapevine Pinot gris virus (GPGV, genus *Trichovirus*, family *Betaflexiviridae*) is a plant pathogen recently found in several grapevine cultivars in many Italian regions, showing leaf deformation, chlorotic mottling and stunting symptoms. In order to determine the presence of GPGV in symptomatic grapevines, a ten year old 'Vermentino' vineyard, situated in Olmedo (Alghero, northern Sardinia) was monitored over three years (2014-2016). The monitoring involved a parcel of 147 plants and allowed to group them as follows: 35 plants that showed typical symptoms of GPGV, 36 plants with no typical symptoms and 76 asymptomatic plants. Diagnostic tests, carried out from the grapevines under observation, showed a high percentage (70%) of GPGV-infected vines among the asymptomatic ones. This represents an important problem regarding the disease control. In the spring of 2017, the work was aimed at confirming the presence of symptoms in the 35 plants observed as symptomatic in the previous year, all of which were shown to be infected by laboratory tests, but also to verify possible symptoms on the 76 asymptomatic vines in 2016. Field observations made on the parcel of 147 plants confirmed the presence of symptoms on symptomatic infected grapevine of the previous year. Interestingly, a significant increase (61%) in the number of symptomatic plants among the asymptomatic plants in 2016 was reported.

COMPETITION FOR NUTRIENTS AND SPACE: A MECHANISM OF ACTION OF *AUREOBASIDIUM PULLULANS* STRAINS. A. Di Francesco, M. Mari. *CRIOF, Department of Agricultural Science, University of Bologna, Via Gandolfi 19, 40057 Cadriano, Bologna, Italy. E-mail:alessand.difrancesc3@unibo.it*

Aureobasidium pullulans strains L1 and L8 were evaluated in order to elucidate how the competition for nutrients and space was involved in their activity against *Monilinia laxa*, the causal agent of peach brown rot. The competition for nutrients was studied by co-culturing pathogen conidia and antagonists in different conditions of nutrient availability and avoiding contact between them. Both antagonists prevented *M. laxa* conidia germination depending on culture substrate. In fact, L1 and L8 showed the lowest inhibition of conidial germination in peach juice at 5%, with a reduction of 12.6% and 13.9%, respectively. HPLC amino acid analysis of peach juice revealed that the addition of the yeast modified their composition: asparagine was completely depleted soon after 12 h of incubation and aspartic acid content markedly increased. Pure asparagine and aspartic acid were tested by *in vitro* trials at the concentrations found in peach juice. Asparagine stimulated pathogen growth; conversely, medium amended with aspartic acid significantly inhibited the conidia germination and mycelial development of the pathogen. Scanning electron microscopy revealed that both strains showed the capability to compete with *M. laxa* for space (starting 8 h after treatment), colonizing the wound surface and inhibiting pathogen growth. Our study showed that L1 and L8 strains could compete with *M. laxa* for nutrients and space; this mode of action may play an important role in the antagonistic activity of the yeast.

CHARACTERIZATION OF PROMOTER SEQUENCES OF *RAPID ALKALINIZATION FACTOR* (*RALF*) GENES IN *FRAGARIA* × *ANANASSA* INTERACTING WITH *COLLETOTRICHUM ACUTATUM*. F. Negrini[1], K. Ogrady[2], K.M. Folta[2], E. Baraldi[1]. *[1] University of Bologna, Dipsa (Department of Agricultural Science), Viale Fanin 46, 40127, Bologna, Italy. [2] Horticultural Sciences Department, University of Florida, 1301 Fifield Hall, Gainesville, FL 32611, USA. E-mail: francesca.negrini6@unibo.it*

Rapid alkalinization factor (*RALF*) genes are ubiquitous in plant kingdom and encode for small peptides that cause a rapid increase of apoplastic pH trough the interaction with its receptor FERONIA. *RALF* genes are involved in many developmental processes from fertilization to cell root growing, and recently they have been identified also as strong suppressors of plant immunity signals. Additionally, *RALF* genes are found in the genome of many plant fungal pathogen species that use an alkalinization strategy to infect the host. Here these genes play a crucial role as pathogenicity genes and are determinant for the infection. *RALF* genes from strawberry (*Fragaria* × *ananassa*) were found upregulated in red fruits infected with the anthracnose pathogen *Colletotrichum acutatum*. To investigate on the signaling mechanisms underneath the *RALF* gene upregulation, the 5' upstream sequence of strawberry *RALF* gene was analyzed using PlantPAN bioinformatic tool in order to predict the cis-acting and transcription factor binding motifs. To assess the determinant region for transcription regulation, progressive truncated sequence 5' upstream of *RALF* coding sequences were isolated and fused to eGFP and GUS reporter genes. The so obtained constructs will be used in transient expression of *Fragaria* × *ananassa* fruit to evaluate the expression level trough qRT-PCR and/or GUS staining upon different exogenous stimuli associated with pathogen perception and response. Evaluating the *RALF* expression regulation by the pathogen will allow to identify candidate editing targets to prevent strawberry pathogenesis from alkalinizing pathogens.

***IN VITRO* AND *IN VIVO* DEVELOPMENT OF THE PREDOMINANT MEMBERS OF THE *FUSARIUM* HEAD BLIGHT SPECIES COMPLEX OF WHEAT AND THEIR SECONDARY METABOLITE PRODUCTION. F. Tini[1], G. Beccari[1], D.M. Gardiner[2], M. Sulyok[3], L. Covarelli[1].** *[1] Dipartimento di Scienze Agrarie, Alimentari e Ambientali, Università degli Studi di Perugia, Borgo XX Giugno 74, 06121, Perugia, Italy. [2] Queensland Bioscience Precinct, CSIRO Agriculture and Food, Brisbane, 4067 QLD, Australia. [3] Department of Agrobiotechnology (IFA-Tulln), University of Natural Resources and Applied Life Sciences, Vienna (Boku), Konrad*

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Fusarium head blight (FHB) is one of the most important fungal wheat diseases worldwide. FHB is caused by at least 17 *Fusarium* species, however, just a few of them are considered prominent members of the complex. A study was carried out to investigate the relationships between the three most important members of the FHB community in central Italy [*F. graminearum* Schwabe, *F. poae* (Peck) Woll. and *F. avenaceum* (Fr.) Sacc.] both *in vitro* and *in planta* and the possible role of secondary metabolites, as analyzed by LC-MS, during interaction. Potato Dextrose Broth and bread wheat heads (cv. A416) were inoculated with conidial suspensions of the three *Fusarium* species in all their possible combinations, including a *F. graminearum Tri5*-KO mutant strain. After 6 days at 25°C on an orbital shaker for liquid cultures and 7 days at 20°C in a growth chamber for inoculated heads, fungal biomass of each species was quantified by real time quantitative-PCR using species-specific primers. In general, a reduction of the total biomass with the increase of the species in the growth medium occurred. *F. avenaceum* showed the lowest development reduction in the presence of the other two species, both in double and triple combination. *F. graminearum* showed a significant development reduction, but *F. poae* was the species that showed the highest growth reduction in the presence of the other two FHB pathogens. The results about both *in vitro* and *in planta* qPCR assays and the presence of secondary metabolites during fungal interactions are discussed.

CHARACTERIZATION OF THE ROLE OF A *RAPID ALKALINIZATION FACTOR* (*RALF*) GENE IN THE SUSCEPTIBILITY OF STRAWBERRY FRUITS TO *COLLETOTRICHUM ACUTATUM*.

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The fungal pathogen *Colletotrichum acutatum*, the causal agent of strawberry (*Fragaria* × *ananassa*) anthracnose, can infect strawberry fruit hosts both at unripe and ripe stages causing anthracnose symptoms only on red ripe fruits. In order to understand the molecular basis of the high susceptibility of red ripe strawberry fruits, the role of a gene encoding for a *Rapid alkalinization factor* (*RALF*), a secreted peptide causing a rapid apoplastic alkalinization, has been investigated. The variation of the expression of *RALF* was monitored by using qRT-PCR at the early time points (8, 16, 20, 24 hours post inoculation, hpi) of the interaction in both white and red strawberry fruits inoculated with *C. acutatum*. The expression of *RALF* was found to increase exclusively in ripe susceptible inoculated fruits starting from 20 to 24 hpi suggesting a role for this gene in modulating the susceptibility of red strawberries. For this reason, an *Agrobacterium*-mediated transient transformation was used for silencing and overexpressing the *RALF* gene in ripe and unripe strawberry fruits, respectively. *RALF*-silenced ripe fruits did not show any decrease in the susceptibility with respect to control fruits. The overexpression of *RALF* in white unripe inoculated strawberries determined a slight decrease in the resistance of these fruits, but further investigation are needed to clearly confirm a role for *RALF* expression in fruit susceptibility, independently from the use of *Agrobacterium* and/or agroinfiltration procedure.

SESSION II

BOTANICAL EPIDEMIOLOGY AND DISEASE CONTROL

Keynote Lecture

MODELLING CROP HEALTH AND CROP LOSSES: CONCEPTS, APPROACHES AND AVENUES

S. Savary

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An overview of concepts and modelling approaches in both plant disease epidemiology and crop loss (yield loss) analysis is presented. In both these fields, epidemiology and crop loss analysis, we discuss the process of model development, including model simplification. We emphasize model simplification as a main avenue to model genericity. We also discuss the usefulness of considering differing evaluation criteria depending on the stage of model development, and thus depending on modelling objectives.

Crop health refers to a very wide range of organisms that may negatively affect crop growth and performances. A generic framework, that allows addressing this diversity, is therefore required for crop health modelling. Two concepts are particularly important in this framework: first, the concept of damage mechanism, with its different processes leading to impairment of crop physiology and crop growth, and second, the concept of injury level, from potential to attainable, and to actual.

The damage mechanisms constitute coupling points between the dynamics of injuries (such as the dynamics of an epidemic), and crop growth (represented in a crop loss model). The concept of crop health syndrome can be operationalized in models as a set of injury functions over time, each function representing the dynamics of an injury (such as, for example, the time-course of an epidemic). Crop health in a given production situation thus is represented by the set of such injury functions, which in turn can be used as drivers for crop loss models, via the damage mechanisms coupling points.

As is the case for crop yield levels, three sets of super-imposed factors are identified that determine three levels of injuries: potential injury (defined by climate, and crop and pest genotypes); attainable injury (determined by the previous set of factors, and limited by crop management); and actual injury (determined by the two previous sets of factors, and reduced by direct pest and disease management instruments). For example, epidemiologists may consider potential, attainable, or actual epidemics. These three levels of injuries (of epidemics) are helpful to model potential (uncontrolled), attainable (limited), and actual (reduced) yield losses caused by diseases and pests.

SESSION II

ORAL PRESENTATIONS

THE LEAFHOPPER VECTOR *EUSCELIDIUS VARIEGATUS* RESPONDS DIFFERENTLY TO INFECTION WITH TWO PHYTOPLASMAS. **L. Galetto[1], S. Abbà[1], M. Rossi[1], M. Vallino[1], M. Pesando[1] M. Pegoraro[1], D. Bosco[1,2], C. Marzachì[1].** *[1]CNR Istituto per la Protezione Sostenibile delle Piante, Strada delle Cacce 73, 10135 Torino, Italy. [2]Università degli Studi di Torino, DISAFA, Largo Braccini 2, 10095 Grugliasco (TO), Italy. E-mail: luciana.galetto@ipsp.cnr.it*

Phytoplasmas are plant pathogenic bacteria spread by hemipteran insects. They can establish either pathogenic or symbiotic relationships with their vectors. The leafhopper *Euscelidius variegatus* is an efficient natural vector of chrysanthemum yellows phytoplasma (CY) and a laboratory vector of Flavescence dorée phytoplasma (FD). These two genetically unrelated phytoplasmas have opposite effects on their common vector species: CY improves its fitness, while FD has a negative effect. To elucidate the molecular bases of these different responses, RNAseq analysis of *E. variegatus* infected with the two phytoplasmas was performed, providing the first *de-novo* sequence assembly for a phytoplasma insect vector. The insect genes differentially expressed between the two conditions (CY *vs* FD) with a functional annotation were related mainly to immune response, movement and energy production. The differential expression of 10 of these genes was confirmed by RT-qPCR, and several biological experiments were carried out to further support the molecular evidences. Consistently with RNAseq results, measures of the phenoloxidase enzymatic activity, melanization and body pigmentation indicated a higher level of activation of the immune response in *E. variegatus* upon FD infection compared to CY infection, confirming the entomopathogenic effect of FD. In line with transcriptomic data, higher CO_2 production and faster movements in CY-infected than in FD-infected insects indicate that the long coevolution between CY phytoplasma and *E. variegatus* could possibly have manipulated the vector behavior to increase its transmission.

POSSIBILITIES OF COPPER REDUCTION IN CONTROL OF TOMATO BACTERIAL DISEASES. **G. Giovanale[1], E. Fortunati[2], A. Mazzaglia[1], G.M. Balestra[1].** *[1]University of Tuscia, Department of Agricultural and Forestry Science (DAFNE), Via S. Camillo De Lellis snc, 01100 Viterbo, Italy. [2]University of Perugia, Civil and Environmental Engineering Department, UdR INSTM, Strada di Pentima 4, 05100 Terni, Italy. E-mail: balestra@unitus.it*

Xanthomonas axonopodis pv. *vesicatory* (*Xav*) *and Pseudomonas syringae* pv. *tomato* (*Pst*) are able to determine economic losses for tomato crops causing serious damage in greenhouse and open field on all airborne organs, including fruits. Their present control is linked to the health of the seeds, appropriated cultural practices and copper preventive treatments. The recent EU guidelines have led to a drastic reduction in Cu^{++} use over the next few years. Alternative plant protection strategies are so requested for conventional and in particular for organic tomato farmers. Here, the objective was to verify the possibility to reduce the copper salts (hydroxide, oxychloride, sulphate) doses currently suggested to contrast both bacteria. *In vitro* and *in planta* tests for *Xav* and *in vitro*, *in planta* and in open field tests for *Pst* were performed. Additionally, botanical extracts and different essential oils, alone and/or in combination with reduced amounts of copper salts, were tested to evaluate the possibility to decrease copper dosage field doses used up to now. The results obtained pointed out the possibility to reduce the amount of copper salts doses maintaining the effectiveness, by reducing the multiplication of *Pst* and *Xav* populations and their damages on tomato plants.

Research funded by MIPAAF, Alt Rame in Bio project.

LONG-TERM ORGANIC MANAGEMENT MODIFIES SOIL MICROBIOTA THAT SUPPRESSES SOILBORNE PHYTOPATHOGENIC FUNGI AND VIRUSES. **G. Cesarano, M. Minutolo, D. Alioto, F. De Filippis, F. Scala, G. Bonanomi.** *Department of Agricultural Sciences, University of Naples Federico II, via Università 100, Portici 80055 (Naples), Italy. E-mail: gaspare.cesarano@unina.it*

Vegetable cultivation under plastic tunnels provides high-quality crop yields but requires high external input and, in the long-term, negatively affects soil fertility. The use of organic amendments has been proposed as an effective approach for soil quality recovery. Here, with a two-year long mesocosms experiment, we compared the effects of organic management, based on crop residues mixed with biochar, ordinary mineral fertilizers and fumigation. Soil quality was assessed by determining physical and chemical soil parameters, whereas microbial community functioning and structure were assessed by high-throughput sequencing of bacterial and eukaryotic rRNA gene markers and BIOLOG EcoPlates™. Disease suppression was evaluated on four pathosystems: lettuce-*Sclerotinia sclerotiorum*; lettuce-*Fusarium oxysporum* f. sp. *raphani*; tomato-*Rhizoctonia solani*; tomato-*Tomato spotted wilt virus*. The use of synthetic fertilizers, in the long-term, lowered crop production and soil pH, soil organic carbon content and aggregation stability but increased salinity. Diversity and richness of bacteria and eukaryotes were lower in the synthetic than in the organic amendments. The addition of organic amendments promoted the growth of *Acidobacteria* and *Gemmatimonadetes*. On the contrary, members of *Actinobacteria* and *Proteobacteria* were more abundant in the soil treated with synthetic fertilizers. Compared with the use of synthetic fertilizers, the application of organic amendments reduced the disease incidence in lettuce-*F. oxysporum* and lettuce-*S. sclerotiorum* pathosystems. On the contrary, in tomato-*R. solani* pathosystem the highest disease suppression was observed with the application of synthetic fertilizers and, for organic management, with the application of alfalfa at high single dose. Finally, the use of organic amendments effectively reduced the disease incidence in tomato artificially inoculated with *Tomato spotted wilt virus*.

USE OF A POMEGRANATE PEEL EXTRACT TO CONTROL OLIVE ANTHRACNOSE. **L. Schena[1], S. Pangallo[1], M.G. Li Destri Nicosia[1], G.E. Agosteo[1], F.V. Romeo[2], P. Rapisarda[2], A. Abdelfattah[1], S. Mosca[1], S. Scibetta[1], S. Minutillo[1], G. Magnano di San Lio[1], S.O. Cacciola[3].** *[1]Dipartimento di Agraria, Università Mediterranea di Reggio Calabria, Località Feo di Vito, 89122 Reggio Calabria, Italy. [2]Consiglio per la ricerca in agricoltura e l'analisi dell'economia agraria (CREA)-Centro di Ricerca Olivicoltura, Frutticoltura e Agrumicoltura (CREA-OFA), Corso Savoia 190, 95024 Acireale (CT). [3]Dipartimento di Agricoltura, Alimentazione e Ambiente, Università degli Studi, Via S. Sofia 100, 95123 Catania, Italy. E-mail: lschena@unirc.it*

A pomegranate peel extract proved to be very effective in controlling olive anthracnose. It had a strong fungicidal activity against the major causal agent of the disease in southern Italy (*Colletotrichum acutatum sensu stricto*), was effective in preventive and curative trials, and induced resistance in treated olive tissues. In field trials, the extract almost completely inhibited the development of rot and was significantly more effective than copper, which is traditionally used to control the disease. The highest level of protection was achieved by applying the extract in the early phases of the disease outbreaks. The analysis of epiphytic populations showed a strong antimicrobial activity of the extract, which sharply reduced both fungal and bacterial populations associated to olive drupes. According to metabarcoding analyses, *C. acutatum s.s.* had a relative abundance of 34% and was much more abundant than

Colletotrichum godetiae (0.1%), supporting previous contentions on a population shift from the latter to the former pathogen in southern Italy. These findings have important practical implications on the development of effective control strategies.

ROLE OF ORNAMENTAL RUTACEOUS PLANTS IN LONG-DISTANCE DISPERSAL OF CITRUS QUARANTINE BACTERIA. V. Catara[1], G. Licciardello[1], A. Urso[1], P. Bella[2], G. Timpanaro[1], P. Caruso[3]. *[1]Dipartimento di Agricoltura, Alimentazione e Ambiente, Università degli Studi di Catania, Via S. Sofia 100, 95123 Catania, Italy. [2]Dipartimento di Scienze Agrarie, Alimentari e Forestali, Università degli Studi di Palermo, Viale delle Scienze, Ed. 4, 90128 Palermo, Italy. [3]Consiglio per la ricerca in agricoltura e l'analisi dell'economia agraria, Centro di Ricerca per l'Olivicoltura, frutticoltura e agrumicoltura, Corso Savoia 190, 95024 Acireale, Italy. E-mail: vcatara@unict.it*

Three major bacterial diseases, Citrus Bacterial Canker (CBC) caused by *Xanthomonas citri* pathovars, Citrus Variegated Chlorosis (CVC) caused by *Xylella fastidiosa* subsp. *pauca* and Huanglongbing (HLB) associated to three '*Candidatus* Liberibacter' species could threaten citrus industry in the EU and Mediterranean countries. Plants (or part of plants) represent the long-distance dispersal across continents of these quarantine bacterial pathogens and their vectors. Plants other than those used for citrus fruit production could be carefully evaluated, *e.g.* ornamental plants and other vegetal material for food or non-food uses in the *Rutaceae* family only partially covered by European Council directives. To implement data useful for the pest risk assessment on the introduction in the EU-Med area of these pathogens through the ornamental plant pathway and to increase awareness on the risk posed by the current measures to prevent entry or the passenger pathway, different data sets were analyzed. The host status of 25 rutaceous ornamental species toward strains of the *X. citri* pathotypes for which very scarce or old data were available was experimentally updated in the framework of ORPRAMed project in collaboration with international institutions. In addition, CBC, CVC and HLB data related to natural and artificial plant hosts, were crossed to the interceptions for plant health reasons of imported rutaceous plants and plant products in the last 15 years according to EUROPHYT and with the trade flows in vegetal material for non-food uses for those countries where these pathogens are present.

APPLICATION OF *TRICHODERMA* STRAINS AND THEIR BIOACTIVE METABOLITES FOR MORE SUSTAINABLE SOYBEAN PRODUCTION. R. Marra[1], N. Lombardi[2], G. d'Errico[1], M. Pascale[1], F. Lacatena[1], A. Pironti[1], A. Bottiglieri[1], P. Lombari[1], M. Lorito[1,2], S.L. Woo[2,3]. *[1]Università degli Studi di Napoli Federico II, Dipartimento di Agraria, Via Università 100, 80055 Portici (NA), Italy. [2]Consiglio Nazionale delle Ricerche-Istituto per la Protezione Sostenibile delle Piante (CNR -IPSP), Via Università 133, 80055 Portici (NA), Italy. [3]Università degli Studi di Napoli Federico II, Dipartimento di Farmacia, Via Montesano 49, 80131 Napoli, Italy. E-mail: robmarra@unina.it*

Soybean represents one of the most cultivated food crops worldwide and has substantial economic importance for industrial applications. Novel biotechnological approaches may be used to increase crop production and reduce chemical inputs in agriculture. *Trichoderma* spp. are plant symbionts, capable of producing multiple positive effects on plants, such as disease control, enhanced growth and production, increased resistance. Recently, it has been demonstrated that *Trichoderma* produces bioactive metabolites (BAM) that can be involved in the beneficial interactions established with the plants. In this work, we investigated the effects of selected *Trichoderma* strains and their BAM, harzianic acid (HA), 6-pentyl-α-pyrone (6PP) and hydrophobin1 (HYTLO1), applied singly or in combinations, on the growth, yield and nutrient uptake of soybean plants in greenhouse and field experiments. A significant growth promotion effect was observed on plants treated with HA and 6PP, when used alone or combined with *T. harzianum* KV906 in greenhouse. In a field trial, there was a 39% increase in soybean productivity (seed weight) in plants treated with 6PP, in comparison to the water control, with effects also on the seed mineral, protein and lipid contents. The treatment modified the fatty acid profile of soybean, *i.e.* by increasing the level of unsaturated (oleic, linolenic and 11-eicosenoic acids) and saturated (stearic acid) fatty acids compared to the control. These results highlight the advantage of using plant biostimulants based on natural compounds and microbes to enhance plant growth and production, as well as improve macro-/micro-nutrient composition of products derived from an important oilseed crop.

SESSION III

FOOD SAFETY AND MYCOTOXINS

KEYNOTE LECTURE

MYCOTOXIN AND FOOD SAFETY IN A CLIMATE CHANGE SCENARIO

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Mycotoxins are a group of naturally occurring secondary metabolites produced by certain moulds. They can grow on a variety of different crops and foodstuffs including cereals, nuts, spices, dried fruits, apple juice and coffee, often under warm and humid conditions.

Recent studies show a general increasing incidence of aflatoxin and deoxynivalenol in cereals and nuts, due to, among other causes, climate change. During postharvest of raw materials, some control measures are required to minimise the mycotoxin problem. Firstly, further mycotoxin accumulation (aflatoxin, in particular) must be avoided through suitable water activity and temperature monitoring during transport and storage steps. At this point, predictive mycology can help to stablish suitable control measures. In particular, probability models for growth or aflatoxin production may provide a decision support on storage times, or storage or transport conditions. Secondly, as no suitable decontamination methods exist which can be applied to foods, it is important to assess the decontamination potential of the food processing operations. In the case of cereals, cleaning, sorting and milling to flour have been shown to reduce mycotoxin presence to some extent, while for nuts, cleaning, sorting and roasting do reduce the initial levels of aflatoxins. Additionally, cereals may be processed to several foods, in particular, deoxynivalenol contaminated wheat is processed into bakery products or pasta which are among the most consumed cereal products in Mediterranean diets. Boiling reduces widely DON concentration in pasta, because it is highly soluble in water and it is transferred to broth, but there is not thermodegradation process associated to it. Apart from the parent toxins, like deoxynivalenol, in the later years there is increased awareness of the existing so-called modified mycotoxins, which are either matrix-associated or modified in their chemical structure and may, consequently, elude conventional analysis because of impaired extraction efficiency. Moreover, the modifed mycotoxin concentrations can sometimes surpass the parent mycotoxin concentration.

Bread production, in particular, involves dough fermentation and baking. Baking, in general, leads to certain deoxynivalenol reduction; however the extent of such reduction depends on temperature level, time, size of loaves, and enzyme levels. On the other hand, dough fermentation involves a high enzymatic activity which results in modified deoxynivalenol (matrix associated) release to the unmodified form. Specific studies have been carried out which demonstrate the impact of enzyme addition to certain cereal foods in the final levels of mycotoxins. Moreover, baking results in a dramatic increase in deoxynivalenol-3-glucoside, a modified mycotoxin which has been shown to be cleaved by intestinal bacteria and thus become bioavailable as DON.

Thus there is a need for in-depth knowledge of the impact of all the food processes which deal with commonly contaminated raw materials, in order to help the food industry to achieve its food safety objectives.

SESSION III

ORAL PRESENTATIONS

NEW ACQUISITIONS ON THE BIOCONTROL OF FUSARIUM HEAD BLIGHT: FROM ECOLOGY TO CRISPR-CAS GENOME EDITING. **S. Sarrocco[1], L. Malfatti[1], S. Manfredini[1], P. Esteban[1], G. Puntoni[1], R. Bernardi[1], M. Haidukowski[2], A. Moretti[2], G. Vannacci[1].** *[1]Università di Pisa, Dipartimento di Scienze Agrarie, Alimentari e Agro-ambientali, Via del Borghetto 80, 56124 Pisa. [2]ISPA-CNR, Via Amendola, 122/O, 70126 Bari, Italy. E-mail: sabrina.sarrocco@unipi.it*

Since the first promising results obtained by the use of the two beneficial isolates *Trichoderma gamsii* T6085 and *Fusarium oxysporum* 7121 against *Fusarium graminearum*, the mechanisms involved in this multitrophic approach for the biocontrol of Fusarium Head Blight (FHB) on wheat have been further investigated. Applications of one or more biocontrol agents during the two critical phases of the disease (crop residues and spikes at anthesis) could result in a good disease control, due to exploitative and interference competition mechanisms potentially used by beneficial organisms to control FHB causal agents and to reduce the risks connected with mycotoxin contamination. In order to investigate the ecology of *T. gamsii* T6085 and *F. oxysporum* 7121, the two antagonists and the pathogen have been used as inoculants of two different natural substrates, wheat and rice, thus resulting in a significant reduction of *F. graminearum* growth and trichothecenes production. The three isolates have been also inoculated on wheat straw: both *T. gamsii* and *F. oxysporum* significantly reduced *F. graminearum* growth and perithecia production on cultural debris. A tool to study fungal behaviour in depth is actually provided by the CRISPR-Cas technique, a genome editing approach with enormous potentiality to be used in biocontrol and crop pathology studies. Recently, CRISPR-Cas9 has been applied to the isolates here described to edit genes putatively involved in plant/pathogen and antagonist/pathogen interaction in order to further investigate aspects connected with the biocontrol of FHB.

A PATHOGENICITY CLUSTER FOR EXPLOITING MAIZE KERNELS DEFENCES IN *ASPERGILLUS FLAVUS*. **L. Antiga[1], S. La Starza[1], C. Miccoli[2], G. O'Brian[3], G.A. Payne[3], S. Xiaomei[3], S. D'Angeli[1], C. Fanelli[1], M.M. Altamura[1], M. Reverberi[1].** *[1]Department of Environmental Biology, University of Rome "Sapienza", Rome, Italy. [2]Università degli Studi del Molise, sede Campobasso, via Francesco De Sanctis 1, 86100, Campobasso, Italy. [3]Department of Plant Pathology, North Carolina State University, 851 Main Campus Drive, Raleigh 27695 – 7567, NC, USA. E-mail: massimo.reverberi@uniroma1.it*

Aspergillus flavus is a saprophytic cosmopolitan fungus, capable of infecting crops in pre- and post-harvest exploiting different secondary metabolites, including aflatoxins. The latter are held in high regard as carcinogenic and genotoxic in animals and humans, even though they have no effect on host plants. In mining the genome of *A. flavus* for identifying secondary metabolite clusters putatively involved in the pathogenesis process, our attention has turned to the cluster 32 containing some fungal effectors such as salicylate hydroxylase, quercetinases and necrosis/ethylene inducing proteins (NepA). In order to understand how this cluster works during the disease, we conducted histological and histochemical experiments in *A. flavus* pin bar-infected maize caryopses. The same samples were analyzed for (i) the expression of specific genes inside the cluster (*e.g. salOH*, *NepA*), (ii) the production of salicylate and the presence of its dehydroxylated form, *i.e.* cathecol, by LC-MS/MS. Within this frame, several mutants of *A. flavus* impaired or enhanced in specific functions (*e.g.* cluster 32 overexpression, NepA KO and OE strains) were checked for their ability to cause disease in maize caryopses. A scenario emerged in which fungal progression through living tissues (*e.g.* aleuron) is accompanied by a significant rise in the level of fungal effectors, such as SalOH and NepA, and by a degradation of SA that, in turn, appears strategic for the fungus to bypass caryopses defences and attenuate programmed cell death phenomena naturally occurring in the aleurone layer of maturating kernels.

***ASPERGILLUS FUMIGATUS* IN ORGANIC SUBSTRATES: A NEW THREAT FOR HUMAN HEALTH.** **K. Santoro[1,2], S. Matić[1], U. Gisi[1], D. Spadaro[1,2], M. Pugliese[1,3], M.L. Gullino[1,2].** *[1]Università degli Studi di Torino, Centro AGROINNOVA, Largo Braccini 2, 10095 Grugliasco (TO), Italy. [2]Università degli Studi di Torino, DISAFA, Largo Braccini 2, 10095 Grugliasco (TO), Italy. [3]AgriNewTech srl, Via G. Quarello 15/A, 10135, Torino (TO), Italy. E-mail: slavica.matic@unito.it*

Aspergillus fumigatus is an ubiquitous saprophytic fungus present in soil and decaying organic materials, which can cause serious diseases in humans and animals. Aspergilloses are treated with demethylation inhibitor (DMI) fungicides, but recently resistant isolates appeared in the medical and also environmental area. The present study focused on molecular characterization and quantification of *A. fumigatus* in different environmental samples and determination of sensitivity to medical and agricultural DMI fungicides. *A. fumigatus* presence was low in soil and meadow/forest organic matter but high concentrations (10^3 to 10^7 cfu/g) were detected in substrates subjected to elevated temperatures, such as compost and silage. High genetic diversity of *A. fumigatus* isolates from compost was found based on simple sequence repeats (SSR) analysis, even within the same fungal population. The *cyp51A* gene, associated to DMI sensitivity, was sequenced and several mutations were found. Nevertheless, all isolates were fully sensitive to the tested DMI fungicides, indicating that the observed mutations did not cause DMI resistance. Only one *A. fumigatus* isolate obtained from compost showed reduced sensitivity to one medical fungicide. In this isolate, six mutations in the *cyp51A* gene were found but none of them corresponded to the mutations L98H, Y121F, and T289A known to code for DMI resistance. This study suggests that *A. fumigatus* isolates from compost and commercial growing substrates cannot be considered as potential carriers for DMI resistance in the environment.

AFLA-PISTACHIO: DEVELOPMENT OF A MECHANISTIC MODEL TO PREDICT AFLATOXIN CONTAMINATION OF GREEK PISTACHIO NUTS. **M.D. Kaminiaris[1], M. Camardo Leggieri[2], D.I. Tsitsigiannis[1], P. Battilani[2].** *[1]Agricultural University of Athens, Department of Crop Science, Laboratory of Phytopathology, Iera Odos 75, 11855 Athens. [2]Università Cattolica del Sacro Cuore, Department of Sustainable Crop Production (DI.PRO.VE.S.), Via Emilia Parmense 84, 29122 Piacenza. E-mail: paola.battilani@unicatt.it*

Pistachios are cultivated worldwide for their high nutritional value and their good flavour. In Greece, the main pistachio variety is *Pistachia vera* cv. *Aegina*. One of the main regions of pistachio cultivation in Greece is Aegina Island, located close to Athens and the pistachio nuts cultivated there are registered as P.D.O. (Product of Designation of Origin). During the last decades, several surveys on Greek pistachio nuts indicated high contamination with aflatoxin B_1 (AFB_1), therefore aflatoxins are considered a major problem for the crop. In Europe, a legislation is in force and 12 µg/kg of AFB_1 is the fixed limit. The ultimate goal of the current study was to develop a mechanistic, weather-driven model, to predict *Aspergillus flavus* growth and AFB_1 contamination in pistachios on a daily base from nut setting until harvest. The planned steps were: i) to

develop a prototype model based on AFLA-maize, ii) to collect meteorological and AF contamination data in Aegina, iii) to run the model and elaborate a probability function to estimate the likelihood to overcome the legal limit and iv) to manage a preliminary validation. AFLA-pistachio model was developed; the validation was carried out using data collected in 2014 and 2015 as model input and around 70% of pistachio orchards were correctly classified by the model in respect to the legal limit. Results were very promising and AFLA-pistachio model seems to be a useful tool for stakeholders to follow the dynamic of AFB_1 contamination risk throughout the pistachio growing season.

Authors acknowledge the Agricultural Association of Pistachio Producers of Aegina for kindly supplying aflatoxin contamination data and the National Asteroscope of Athens for kindly supplying meteorological data.

YEAST VOLATILE ORGANIC COMPOUNDS REDUCE GROWTH AND OCHRATOXIN A PRODUCTION BY *ASPERGILLUS* spp. M.G. Farbo[1], P.P. Urgeghe[1], Z. Ul Hassan[2], A. Marcello[1], S. Jaoua[2], Q. Migheli[1]. *[1]Dipartimento di Agraria, Università degli Studi di Sassari, Viale Italia 39, I-07100 Sassari, Italy. [2]Department of Biological and Environmental Sciences, College of Arts and Sciences, Qatar University, P.O. Box: 2713, Doha, Qatar. E-mail: mgfarbo@uniss.it*

Ochratoxin A (OTA) is classified as a group 2B carcinogen by the World Health Organization. Some species of *Aspergillus* are the main source of OTA in warm and tropical regions, particularly *Aspergillus carbonarius* (Bainier) Thom. is considered one of the most relevant OTA producers in food and feed. Inhibiting the growth of OTA-producing fungi on sensitive commodities is by far the most reliable method to prevent OTA contamination of food and feed. Aim of this study is to evaluate the biocontrol ability of selected yeast strains against OTA producing *A. carbonarius* and *Aspergillus ochraceus*. We have previously demonstrated that four low or non-fermenting yeasts are able to control the growth and sporulation of OTA-producing *A. carbonarius* both *in vitro* and on detached grape berries. Two yeast strains (*Candida intermedia* 253 and *Candida friedrichii* 778) were most effective in reducing mycelial growth, sporulation and *in vitro* OTA production by *A. carbonarius* and *A. ochraceus*. This biological effect is at least partly due to the release of volatile organic compounds (VOCs). Exposure to yeast VOCs affects gene expression in *A. carbonarius*, as confirmed by downregulation of polyketide synthase, non-ribosomal peptide synthase, and the regulatory genes *laeA* and *veA*. HS-SPME/GC-MS analysis identified about 20 compounds, belonging to different chemical classes, such as alcohols, aldehydes, hydrocarbons and terpenes. Further studies will aim at testing single purified VOCs in order to identify the most effective compounds responsible for the inhibition of fungal growth and OTA production by *Aspergillus* spp.

This publication was made possible by NPRP grant # 8-392-4-003 from the Qatar National Research Fund (a member of Qatar Foundation). The findings achieved herein are solely the responsibility of the authors.

***FUSARIUM AVENACEUM*, AN EMERGING PATHOGENIC FUNGUS CAUSING PRE-HARVEST WET APPLE CORE ROT IN TRENTINO REGION. V. Gualandri, A. Branz, M. de Concini, G. Angeli.** *Fondazione Edmund Mach, Center for Technology Transfer, Via E. Mach 1, 38010 S. Michele all'Adige, Trento. E-mail: valeria.gualandri@fmach.it*

Trentino region is leading producer of apple (*Malus domestica*) in Italy with 9750 ha and more than 500.000.000 kilogram grown in 2015. The most common cultivars are Golden Delicious, Red Delicious and Renetta Canada. During 2011-2015 pre-harvest monitoring showed elevated presence of symptoms of core rot on apple fruits of cv. Golden Delicious, Fuji, Renetta Canada and Red Delicious. *Botryosphaeria* spp. and *Fusarium* spp. are the main fungi involved in the aetiology of this disease. Several species of the genus *Fusarium* can cause wet core on apple fruit. As wet core of apple is undetectable until the fruit is cut or consumed, it affects consumer confidence, constituting an economical problem for growers and a safety issue due to the potential production of mycotoxins. In this study we identified several *Fusarium* isolates from rotten in apple fields in Trentino Region (northen Italy). *Fusarium* identity was further investigated by sequence comparison of the ITS (primers ITS4/ITS5 and FA-ITSR/FA-ITSF). BLASTn analysis revealed high identity to *Fusarium avenaceum*. Therefore, both molecular and morphological observations indicated that the principal pathogenic *Fusarium* reported to cause wet core rot on apple fruits in Trentino region was *F. avenaceum*. This research focused on the molecular characterization of *Fusarium* strains involved in the wet core of apple fruit in Trentino region in order to increase knowledge on the biology and the infection mechanisms for an efficient control of the disease.

POSTERS

RNAi STRATEGIES TO INDUCE GENE SILENCING IN THE PHYTOPLASMA VECTOR *EUSCELIDIUS VARIEGATUS*.
S. Abbà[1], L. Galetto[1], M. Vallino[1], M. Turina[1], M. Pesando[1], M. Rossi[1], M. Pegoraro[1], D. Bosco[1,2], C. Marzachì[1]. *[1]CNR Istituto per la Protezione Sostenibile delle Piante, Strada delle Cacce 73, 10135 Torino, Italy. [2]Università degli Studi di Torino, DISAFA, Largo Braccini 2, 10095 Grugliasco (TO), Italy. E-mail: simona.abba@ipsp.cnr.it*

In this study, gene silencing techniques mediated by RNA interference (RNAi) are applied to the leafhopper *Euscelidius variegatus*, a natural vector of chrysanthemum yellows phytoplasma and an efficient vector of Flavescence dorée phytoplasma under laboratory conditions. Phytoplasmas are plant pathogenic bacteria transmitted by phloem-feeding insects belonging to the order *Hemiptera* and are associated with many diseases that cause severe economic impacts on many crops worldwide. The purpose of the work was to shed light on insect genes potentially involved in the interaction with phytoplasmas, and, more specifically, in the acquisition and transmission mechanisms of these pathogens. In particular, the ability of actin and ATP synthase β double stranded RNAs (dsRNAs) to induce RNAi effects after abdominal injection is being tested. Both genes are known to be involved in the interaction between phytoplasmas and their insect hosts, so an efficient perturbation of their expression is likely to cause a modification in the epidemiological cycle of this disease. The possibility of using insect viruses as dsRNA delivery tools for virus-induced gene silencing is being also explored. During a RNA-seq project of *E. variegatus*, a contig with high sequence similarity to known picorna-like viruses was in fact identified among the assembled insect transcripts. We are currently constructing an infectious clone of this virus, named *Euscelidius variegatus* virus 1 (EVV-1), which would represent a groundbreaking tool to be used directly as a biocontrol agent, providing additional tools to integrate or even replace insecticide treatments towards a more sustainable agriculture.

TERRITORY SURVEILLANCE OF *PANTOEA STEWARTII* subsp. *STEWARTII* FOR *ZEA MAYS* SEED PRODUCTION.
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Pantoea stewartii subsp. *stewartii* is indigenous to the Americas and has been introduced to other parts of the world with maize seeds. It causes the disease called Stewart's wilt. The main host is *Zea mays* (maize), especially sweet corn, but dent, flint, flour and pop corn cultivars can also be infected. In the Americas, *Chaetocnema pulicaria* Melsheimer (Coleoptera: Chrysomelidae) is the only known efficient vector and the main overwintering site of the bacterium. The pathogen may be transmitted in seed, and has occasionally been found to be able to overwinter in soil, manure or maize stalks. As part of the surveillance of the territory carried out in Emilia-Romagna on seed crops, two corn fields with symptoms of *Pantoea stewartii*, confirmed by laboratory analysis, were detected in 2015 and 2016. It was given official communication to the Italian Ministry of Agriculture and Forestry and prescribed the destination for other use of the seeds collected from the concerned fields. In 2017, a national drug monitoring program was developed, which was shared and adopted by the Italian regions. The program takes into account the EPPO protocol and at the end of the first year the results will be evaluated in terms of diagnostic technique and monitoring outcomes.

PLANT DECLINE ETIOLOGY IN POPLAR SHORT-ROTATION COPPICE PLANTATIONS.
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Poplar short rotation coppice represents a very good opportunity for the production of bioenergy as a strategy against global changes. In recent years, numerous plantations in central Italy showed worrying stump mortality of uncertain cause. Investigations have therefore been conducted to determine the etiology and symptomatology of this phenomenon. The external symptoms begin with leaf yellowing and sometimes necrosis, microphyllia, and reduced shoots growth, followed by withering and death of the stump. Upon sectioning of the stump, we detected browning of internal tissues of roots and crown, gradually deepening in. In severe cases, we observed wood decay in the central part of the stump, eventually deepening into the root system. Cultures from the edge of necrotic tissue on PDA yielded isolates of the same fungus. Sequence analysis of the Internal Transcriber Spacer region (ITS1-5.8S-ITS2) and of the β-tubulin gene showed that the isolates belong to the genus *Cryptosphaeria*. Species identification is still ongoing. Eventually, wood decay organisms belonging to the genera *Pholiota* and *Collibia* colonized the tissues as secondary invaders. Future goals will be to identify the species of the pathogen, define the disease predisposing factors and identify possible control measures.

FUNGI ASSOCIATED TO HAZELNUTS POSTHARVEST.
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Hazelnuts are important products used for direct consumption or processing, harvested once per year, and commonly stored for long periods before consumption. Several diseases are reported to this fruit, with discolour, rot and visible mould on kernels. Knowledge on these diseases and their role on product loss is still poor, as well as their linkage to the crop growing period and/or post-harvest is not clearly defined. The aim of this study was to describe fungi associated to hazelnuts, both in healthy and diseased nuts, during post-harvest, drying and storage conditions and how they act in time. Hazelnut were grown in Turkey, in two different fields, one of them with severe powdery mildew attack. Nuts were dried to 6% humidity in good and bad conditions. Sampling was managed after 15 day drying in good conditions or after 12 and 24 day drying when bad conditions were applied. Regarding storage, hazelnuts were sampled at the beginning and after 6 months. For each sample, associated fungi were obtained with direct plating. The most isolated fungi belonged to *Penicillium* spp., followed by *Cladosporium*, *Pestalotiopsis*, *Phomopsis* (above 10% mean incidence), *Aspergillus*, *Fusarium* (below 10%) and *Alternaria*, *Colletothricum*, *Rhizopus* (very limited occurrence). Regarding the origin of the orchards, the incidence of *Aspergillus* and *Fusarium* was significantly higher where powdery mildew was signalled. Bad drying conditions significantly increased *Cladosporium* spp. incidence. The increment in drying period, caused by bad drying conditions, produced an increase of *Aspergillus* and *Penicillium* incidence; some other fungi

increased (i.e. *Colletothricum*), especially when nuts were dried in good conditions (i.e. *Trichotecium*).

INVESTIGATING THE ROLE OF PLANT PRODUCED β-AMINOBUTYRIC ACID (BABA) IN DEFENSE AGAINST PATHOGENS. **I. Baccelli[1], G. Glauser[2], B. Mauch-Mani[1].** *[1]University of Neuchâtel, Institute of Biology, Rue Emile-Argand 11, 2000 Neuchâtel, Switzerland. [2]University of Neuchâtel, Neuchâtel Platform of Analytical Chemistry (NPAC), Avenue de Bellevaux 51, 2000 Neuchâtel, Switzerland. E-mail: ivan.baccelli@unine.ch*

β-aminobutyric acid (BABA) is a non-protein amino acid that, when applied to plants, can induce resistance through priming. BABA was believed to be xenobiotic until recently, when in our laboratory its endogenous production was demonstrated to occur in *Arabidopsis* and some crops, like maize and wheat. Further analyses revealed that BABA levels increase following pathogen infection (necrotrophic, biotrophic and hemibiotrophic pathogens) and abiotic stress (salinity and submergence), but the role of plant-produced BABA remains to be established. In order to investigate the biological significance of endogenous BABA variations during plant-pathogen interactions, we analyzed BABA levels in *Arabidopsis* plants after infections with virulent, avirulent (AvrRpt2), and non-pathogenic (hrpA) strains of *Pseudomonas syringae* pv. *tomato* DC3000, as well as after treatment with peptide elicitors (flagellin 22 and AtPep2). Our results show that the production of BABA is controlled by the molecular recognition of pathogen presence and endogenous BABA levels are accumulated differently according to resistance or susceptibility to disease. A mutant screening also led to the identification of a high producer of constitutive levels of BABA: the *Arabidopsis* mutant constitutive expresser of pathogenesis-related genes 5 (cpr5).

SPHINGOLIPIDS IN *FUSARIUM VERTICILLIOIDES* – *ZEA MAYS* INTERACTION. **M. Beccaccioli[1], M. Salustri[1], M. Reverberi[1], C. Fanelli[1], M. Ludovici[2], V. Scala[3].** *[1]Department of Environmental Biology, University of Rome "Sapienza", Roma, Italy. [2]IFO S. Gallicano, via E. Chianesi 53, 00144 Roma, Italy. [3]CREA-DC, via G. Bertero 32, 00136 Roma, Italy. E-mail: valeria.scala@crea.gov.it*

The sphingolipidome, *i.e.* the whole ensemble of sphingolipids, undergoes perturbation during the interaction between *Zea mays* caryopses and the mycotoxigenic fungus *Fusarium verticillioides*. Fumonisins, mycotoxins produced by *F. verticillioides*, inhibit the activity of ceramide synthase in plant host cell, leading to accumulation of sphingoid bases and depletion of complex sphingolipids. Sphingoid bases accumulation should trigger, in turn, programmed cell death (PCD) activating the MPK6-salycilic acid pathway, while depletion of complex sphingolipids could cause cell death because of membrane and vesicle impairment. Maize sphingolipidome is perturbed also in the early phases of infection, when the cell membrane goes through a rearrangement leading to the formation of defense-related lipid rafts enriched in complex sphingolipids. The level of variation of several sphingolipids, fumonisins and salicylic acid was evaluated by multiple reaction monitoring (MRM) approach in liquid cromatography-mass spectometry (LC-MS/MS) in maize ears artificially infected with *F. verticillioides* at different times post inoculation. We compared this metabolic profile with the expression of genes related with PCD and defense onset and execution. We hypothesize that *F. verticillioides* could jeopardize the sphingolipids metabolism of *Z. mays* at the early phases of infection to favor its progression into the caryopses.

A LIPIDOMIC APPROACH TO EVALUATE *FUSARIUM VERTICILLIOIDES*-MAIZE INTERACTION. **M. Beccaccioli[1], V. Scala[2], A. Grottoli[1], M. Ludovici[3], M. Reverberi[1].** *[1]Department of Environmental Biology, University of Rome "Sapienza", Rome, Italy. [2]CREA-PAV, 00136 Roma, Italy. [3]Laboratory of Cutaneous Physiopathology and Integrated Center of Metabolomics, San Gallicano Dermatologic Institute (IRCCS), Rome, Italy. E-mail: marzia.beccaccioli@uniroma1.it*

During host-pathogen interaction lipids play a crucial role, since they are not just a component of membrane but they can contribute to cell signaling. Some lipids and their metabolizing enzymes play a role in the regulation of fungal pathogenicity. Lipid metabolism involves various organelles, such as endoplasmic reticulum, mitochondria, peroxisomes, and lipid droplets, and the enzymes present in these districts govern important steps in the different lipid metabolic pathways. In this study, we considered the fatty acids (FA), the building blocks for the synthesis of the membrane lipids that also have the ability to act as signal molecules. Actually, fatty acids are the precursor of oxylipins, an oxidised fatty acid able to drive signals. We studied the lipid metabolism of *Fusarium verticillioides*. We analysed the gene expression of some enzymes implicated in the synthesis of LCFA (long chain fatty acids) and VLCFA (very long chain fatty acids), and of those involved in the oxylipin production during mycelia vegetative growth and the interaction with the host (kernels of *Zea mays*). Through a lipidomic approach we determined which FA and oxylipins are most involved in these stages of fungal life, as saprobiont and as pathogen. A crucial alteration of these FA classes emerged, suggesting that these molecules are responsible for the regulation of the fungal growth and host interaction.

PRELIMINARY RESULTS ON THE LEVEL AND DURATION OF WINTER WHEAT SUSCEPTIBILITY TO FUSARIUM HEAD BLIGHT CAUSAL AGENTS. **G. Beccari[1,2], L. Covarelli[1], F. Tini[1], M. Sulyok[3], C. Cowger[2].** *[1]Department of Agricultural, Food and Environmental Sciences, University of Perugia, Borgo XX Giugno 74, 06121, Perugia, Italy. [2]USDA, Department of Entomology and Plant Pathology, North Carolina State University, 27695, Raleigh, USA. [3]Department of Agrobiotechnology (IFA-Tulln), University of Natural Resources and Life Sciences, Vienna, Konrad Lorenzstraße 20, A-3430, Tulln, Austria. E-mail: giovanni.beccari@progetti.unipg.it*

A greenhouse experiment was conducted at NCSU (Raleigh, USA) to assess winter wheat susceptibility over time to the Fusarium head blight (FHB) causal agents *F. graminearum*, *F. avenaceum* and *F. poae*. Plants of the winter wheat cultivar Dyna-Gro Shirley, highly susceptible to FHB, were grown in pots. Anthesis time was considered as when 30-50% of anthers were extruded. At 0, 3, 6 or 9 days after anthesis (daa), heads were spray-inoculated with each of the three strains and incubated under mist irrigation for 72 hours. FHB severity was visually assessed for each head at 21 days after inoculation (dai). Fungal biomass and fungal secondary metabolites were quantified in the kernels harvested at physiological maturity by q-PCR and LC-MS/MS, respectively. The aggressiveness of the species, as measured by severity scoring at 21 dai and by q-PCR, was: *F. graminearum* > *F. avenaceum* >> *F. poae*. Overall, *F. graminearum* caused more severe FHB symptoms and higher deoxynivalenol accumulation at the earlier-middle infection timings (3 and 6 daa). Similarly, *F. avenaceum* biomass, accompanied by production of large quantities of the related mycotoxins such as enniatins and moniliformin, was higher at the same infection timings. Conversely, *F. poae* showed more severe disease and nivalenol accumulation at later infection timings (9 daa). *F. poae* may have required more extruded anthers for infection or resistance to this species was lower as the grain-filling process advanced. It is hypothesized that when

favorable conditions first occur at late anthesis, *F. poae* may have a relative lead in provoking the disease.

FUSARIUM SPECIES AND SECONDARY METABOLITES ASSOCIATED WITH DURUM WHEAT GRAINS FROM THREE DIFFERENT ITALIAN CLIMATIC AREAS. **G. Beccari[1], M.T. Senatore[1,2], L. Pedini[1], F. Tini[1], A. Prodi[2], P. Nipoti[2], V. Balmas[3], M. Sulyok[4], L. Covarelli[1].** *[1]Department of Agricultural, Food and Environmental Sciences, University of Perugia, Borgo XX Giugno 74, 06121 Perugia, Italy. [2]Department of Agricultural Sciences, Alma Mater Studiorum University of Bologna, Viale G. Fanin, 44, 40127 Bologna, Italy. [3]Department of Agriculture, University of Sassari, Via E. De Nicola 9, I-07100 Sassari, Italy. [4]Department of Agrobiotechnology, University of Natural Resources and Life Sciences, Vienna, Konrad Lorenzstraße 20, A-3430 Tulln, Austria. E-mail: lorenzo.covarelli@unipg.it*

Durum wheat grain samples, collected in three different Italian climatic areas (Emilia-Romagna, Umbria and Sardinia), were analyzed to: 1) isolate the mycoflora infecting the seeds on modified potato dextrose agar and by the deep-freezing blotter method; 2) identify the composition of the *Fusarium* head blight (FHB) species complex by *TEF1-α* region sequencing; 3) quantify the *Fusarium* species associated with FHB by q-PCR directly in the kernels; 4) determine fungal secondary metabolites in the grains by LC-MS/MS. The mycoflora was mainly represented by *Alternaria* and *Fusarium* species. The highest *Fusarium* spp. incidence was found in the samples from Emilia-Romagna, followed by those harvested in Umbria and in Sardinia. The FHB complex was mainly represented by *F. poae* in all the examined areas, while *F. graminearum* showed the highest incidence in the Emilia-Romagna samples followed by those from Umbria. This species was not detected in the samples from Sardinia. q-PCR assays allowed the detection of other *Fusarium* species, such as *F. langsethiae* and *F. sporotrichioides*, which were not found by the used isolation methods, showing that this technique may give an important contribution for an exhaustive description of the FHB complex. Secondary metabolites were correlated to the fungal community detected. In fact, deoxynivalenol (DON) was mainly found in the Emilia-Romagna samples, in which the DON producing species were more frequent. Enniatins and beauvericin were detected in the samples collected in Emilia-Romagna and Umbria. This study gives a comprehensive overview on the FHB complex by different analytical techniques and on the associated secondary metabolites.

SUSCEPTIBILITY OF _PAPAVERACEAE_ SPECIES TO _FUSARIUM OXYSPORUM_ f. sp. _PAPAVERIS_. **D. Bertetti[1], M.L. Gullino[1,2], A. Garibaldi[1].** *[1]Università degli Studi di Torino, Centro AGROINNOVA, Largo Braccini 2, 10095 Grugliasco (TO), Italy. [2]Università degli Studi di Torino, DISAFA, Largo Braccini 2, 10095 Grugliasco (TO), Italy. E-mail: domenico.bertetti@unito.it*

In 2011, *Fusarium oxysporum* f. sp. *papaveris* was reported on Iceland Poppy (*Papaver nudicaule*) for the first time in Italy. Successively, the susceptibility of eleven *Papaveraceae* species was evaluated. Nine *Papaver* species commonly grown for gardens and cut flowers were artificially inoculated together with *P. rhoeas* and *Chelidonium majus* naturally diffused in Italian flora. Five single-spore strains of the pathogen were used in the tests and the roots of the plants were dipped in the conidial suspensions prepared for each of them. On the basis of the disease index (D.I.) obtained by the final evaluation of symptoms, each species was assigned to one of the following susceptibility classes: R = Resistant (D.I. 0-5); PR = Partially Resistant (D.I. 6-20); MS = Moderately Susceptible (D.I. 21-50); S = Susceptible (D.I. 51-75); HS = Highly Susceptible (D.I. 76-100). *Papaver atlanticum*, *P. dubium*, *P. glaucum*, *P. pseudocanescens*, *P. nudicaule*, *P. rupifragum* and *Papaver* "Daneborg" were susceptible or highly susceptible to the pathogen; *P. trinifolium* and *P. orientale* were moderately susceptible; *C. majus* and *P. rhoeas* showed a susceptibility ranging from moderate to high. About the origin of *F. oxysporum* f. sp. *papaveris*, the susceptibility of *C. majus* and *P. rhoeas* might indicate that the pathogen is present in the spontaneous flora in Italy and/or that wild *Papaveraceae* species are a potential source of inoculum; the high temperatures as those registered in the glasshouse cultivation where the disease first appeared in Italy might help the development of symptoms that are limited in the sub-polar areas in which *P. nudicaule* is indigenous.

LOOP-MEDIATED ISOTHERMAL AMPLIFICATION ASSAY FOR THE RAPID DETECTION OF _CURTOBACTERIUM FLACCUMFACIENS_ pv. _FLACCUMFACIENS_ FOR QUARANTINE AND FIELD APPLICATIONS. **C. Biancalani[1], S. Calamai[1], M. Cerboneschi[1], E. Osdaghi[2], S. Tegli[1].** *[1]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. [2]Department of Plant Protection, Shiraz University, Shiraz, Iran. Email: carola.biancalani@unifi.it*

Curtobacterium flaccumfaciens pv. *flaccumfaciens* (*Cff*) is a Gram-positive bacterium, and the causal agent of a systemic vascular disease known as bacterial wilt of beans (*Phaseolus vulgaris*). Bean seedlings rapidly die after infection, while older plants can survive the attack and produce mature seeds, infected by *Cff* via the vascular system. These seeds are generally asymptomatic, with rare yellow, orange, or purple pigmentation on white bean seeds, depending on the *Cff* strains/variants. Within or on infected seeds *Cff* remains viable for decades, and seeds are the main means of dissemination of *Cff* over short and long distances. This bacterium is present since the early 20th century in North America, where it still causes high economic losses, and is endemic in many bean-producing countries. Although *Cff* is a quarantine pathogen for Europe (EPPO A2 list), so far the control of imported seeds for *Cff* is not mandatory, and inspections often consist just on visual examination. Recently, the isolation of *Cff* from diseased soybean in Germany dramatically focused on the threat posed by its further spread. Therefore, it is urgently needed to implement rapid, easy and specific tests for *Cff*, having the potential to be routinely used at the ports of entry. A loop-mediated isothermal amplification (LAMP) assay for *Cff* was developed, targeting a nucleotide sequence exclusive of *Cff* strains pathogenic on bean. This LAMP test has a sensitivity of 10 pg/reaction, and the entire procedure takes only about 1.5 h.

We gratefully acknowledge Ente Cassa di Risparmio di Firenze (project 2015.0999) and University of Florence (PIA 2017-2018) for the financial support.

ORGANIC FARMING PROMOTES THE DEVELOPMENT OF BENEFICIAL SOIL MICROBIOTA THAT INCREASES _RHIZOCTONIA SOLANI_ DISEASE SUPPRESSION. **G. Bonanomi[1], G. Cesarano[1], V. Antignani[2], C. Di Maio[1], F. De Filippis[1], F. Scala[1].** *[1]Department of Agricultural Sciences, University of Naples Federico II, via Università 100, 80055 Portici (Naples), Italy. [2]Department of Biology, Division of Natural Science, Bob Jones University, Greenville, USA, 29614. E-mail: giuliano.bonanomi@unina.it*

Intensive farming in agriculture raises serious concerns about the environmental sustainability due to the widespread use of

agrochemicals. In this work, we assessed the impact of organic and conventional soil management under intensive farming on soil suppressiveness against the soilborne pathogen *Rhizoctonia solani*. Three farms were considered: two of them practiced organic cultivation (for 10 and 20 years, respectively), while the other one practiced conventional cultivation. Soil suppressiveness was assessed in a greenhouse bioassay with lettuce (*Lactuca sativa*). Soil microbiome was characterized by combining BIOLOG EcoPlates™ with high-throughput sequencing of bacterial and eukaryotic rRNA gene markers. *R. solani* disease suppression was higher in organic than in conventional farming soil, but this property was lost in all soils after sterilization. Functional biodiversity was significantly higher in the two organic soils and was predictive of the suppressiveness towards *R. solani*. High-throughput sequencing of bacterial and eukaryotic rRNA gene markers revealed that organic management dramatically alters soil microbial composition and diversity. Surprisingly, soil bacterial diversity was lower in organic than in the conventional farms. On the other hand, soil *Metazoa* and especially *Anellida* of the family *Enchytraeidae* were almost absent in soils from the conventional farm, but were abundant in soils under organic management. Analysis, carried out at the genus level for the most abundant bacterial and eukaryotic microbial species, showed that 58.7% of the genera had a statistically significant correlation with suppressiveness, with the genus *Flavisolibacter*, *Massilia*, *Pseudomonas*, *Ramlibacter*, *Rhizophus*, and the earthworms belonging to the *Enchytraeidae* family positively correlated with the disease suppression. According to our analyses, the overall microbial taxonomic diversity was unrelated to suppressiveness.

HOW CLIMATE CHANGE COULD INFLUENCE MYCOTOXIN PRODUCTION IN LEAF VEGETABLE? P. Bosio[1], I. Siciliano[1], G. Gilardi[1], M.L. Gullino[1,2], A. Garibaldi[1]. *[1]Università degli Studi di Torino, Centro AGROINNOVA, Largo Braccini 2, 10095 Grugliasco (TO), Italia. [2]Università degli Studi di Torino, DISAFA, Largo Braccini 2, 10095 Grugliasco (TO), Italia. E-mail: ilenia.siciliano@unito.it*

Climate change will likely affect mycotoxins in agricultural products. The behaviour of mycotoxin production of *Myrothecium roridum* on cultivated rocket, of *M. verrucaria* on spinach, and for *Alternaria* sp. on rocket cabbage and cauliflower have been evaluated under phytotron conditions. Different temperature (from 14-18°C to 26-30°C) and CO_2 concentration (775-870 or 1550-1650 mg/m^3 of CO_2) combinations were tested. Verrucarin A and roridin E were produced by both *Myrothecium* sp. tested under all the evaluated temperatures and CO_2 conditions. For rocket, the maximum level of verrucarin A was found at 14-18°C and 1550-1650 mg/m^3 of CO_2, and the maximum roridin E production was detected at 26-30°C with 1550-1650 mg/m^3 of CO_2. For spinach, the maximum levels of verrucarin A and roridin E were found at a temperature of 26-30°C and a CO_2 level of 1550-1650 mg/m^3. Tenuazonic acid, alternariol, alternariol monomethyl ether and tentoxin produced by different *Alternaria* strains were analysed for each climate condition on rocket cabbage and cauliflower. Higher temperature influences environmental conditions and different factors involved in plant-pathogen interaction. Temperature was the main factor involved in disease severity, while host plants and strains were found to be the factors mostly influencing mycotoxin production. A large variability in the production of mycotoxins among the different host plants was observed. The spread of new pathogens induced by climate change could represent a risk for human health considering the ability of these pathogens to produce mycotoxins.

DOWNY MILDEW EPIDEMICS ON PARTIALLY RESISTANT GRAPEVINE: A SYSTEMS ANALYSIS. F. Bove[1], L. Willocquet[2], S. Savary[2], T. Caffi[1], V. Rossi[1]. *[1]Università Cattolica del Sacro Cuore, Dipartimento di Produzioni Vegetali Sostenibili, DI.PRO.VE.S., Via E. Parmense 84 - 29122 Piacenza, Italy. [2]Institut National de la Recherche Agronomique (INRA) UMR1248 AGIR, 24 Chemin de Borde Rouge, Auzeville, F-31326 Castanet-Tolosan. France. E-mail: federica.bove@unicatt.it*

Process-based simulation modelling can be a useful approach in leading phenotyping studies. The application of this approach is most interesting in the case of diseases of perennial crops such as grapevine, where experimental manipulation of plant material can be difficult. A process-based approach has been used to acquire knowledge on grapevine downy mildew (DM), caused by the oomycete *Plasmopara viticola*. A generic simulation model is here designed to analyze DM epidemics on both leaves and bunches, including various components of partial host resistance, such as infection efficiency, sporulation, infectious period and latency period. The proposed model is based on numerical integration and involves five different processes occurring during host–pathogen interactions: (i) crop growth, development and senescence; (ii) primary and secondary infection; (iii) infection on leaves; (iv) infection transmission to clusters; and (v) effect of genetic and ontogenic resistance. The simulation software STELLA® was used to encode the model. Specifications of the model are described and simulated outputs are presented. Knowledge gaps and insights for further research are highlighted.

Federica Bove carried out this work within the Doctoral School Agrisystem of the Università Cattolica del Sacro Cuore (Italy).

MinION SEQUENCER: A NEW TOOL FOR A FAST DISSECTION OF VIRAL INFECTION. M. Calassanzio, F. Morigi, E. Selleri, C. Lanzoni, A. Prodi, C. Ratti. *Dipartimento di Scienze Agrarie, Area Patologia Vegetale, Università di Bologna, Bologna, Italy. E-mail: claudio.ratti@unibo.it*

MinION is a portable and real time DNA and RNA sequencer developed by Oxford Nanopore Technologies. It consists of a chip with incorporated nanopores that measure the changes in electrical conductivity generated by the passage of DNA strands through a biological pore. The current in the nanopore is measured by a sensor several thousand times per second, and the data streams are passed to a microchip called the Application-Specific Integrated Circuit (ASIC). Data processing is carried out by the MinKNOW software, which deals with data acquisition and analysis. With MinION genomic fragments can be sequenced up to 50 kb with single-strand read accuracy better than 92%. The aim of this work was to use the MinION device to identify all viruses infecting plant samples and filamentous fungi. DNA libraries were generated from dsRNA, total RNA or TNA from purified virions from tobacco, rose, tomato, ficus, cabbage or *F. culmorum* strains and subject to analysis in pools of 10-15 samples. The reads generated by short runs (2 hours) using the developed protocols range from 100 to 4000 bp in size with the main distribution around 400 bp. BLAST analysis of the assembled contigs provided sufficient information to identify various mycoviruses and phytoviruses in the analyzed samples. Nanopore DNA strand sequencing proved to satisfy our purpose offering a universal device allowing the rapid identification of known and unknown pathogens that could substitute complex and time consuming traditional approaches.

EVIDENCE FOR THE INVOLVEMENT OF VP37 OF *BROAD BEAN WILT VIRUS 1* IN THE INDUCTION OF PLANT SYMPTOMS AND POSTRANSCRIPTIONAL GENE SILENCING. C. Carpino[1,2], I. Ferriol[2], L. Elivira-Gonzalez[2], L. Rubio[2,3], E. Peri[1], S. Davino[1], L. Galipienso[2,3,4]. *[1]Department of Agricultural and Forestry Science, University of Palermo, Piazza Marina 61, 90133 Palermo, Italy. [2]Instituto Valenciano de Investigaciones Agrarias (IVIA), Ctra. CV-315, 46113 Moncada, Valencia, Spain. [3]Euro- Mediterranean Institute of Science and Technology (IEMEST), Via Michele Miraglia 20, 90139 Palermo, Italy. [4]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 *hrp* 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 *hrp* 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.

We gratefully acknowledge EU Commission (project LIFE12 ENV/IT/336 "AFTER Cu") and Ente Cassa di Risparmio di Firenze (project 2015.0999) for their financial support.

INTERFERENCES ON THE DETECTION OF *XYLELLA FASTIOSA* IN *POLYGALA MYRTIFOLIA* BY REAL-TIME PCR. V. Chiatti[1,2], N. Pucci[1], S. Lucchesi[1], V. Modesti[1], M. Reverberi[2], S. Loreti[1]. *[1]Centro di ricerca per la patologia vegetale (CREA-PAV) via C.G. Bertero 22, 00156 Roma. [2]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™ and EvaGreen™) 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 2nd generation biodiesel. The spectrophotometric analysis of an extract, obtained after alkaline pre-treatment of giant reed dry biomass, revealed the

presence of phenolic compounds. After neutralization, the extract was assayed *in vitro* at different concentration (from 10 up to 80% in PDA plates) for biological activity towards different filamentous fungi, evaluating the colony growth inhibition. *Sclerotium rolfsii*, *Phanerochaete crysosporium* and *Trametes versicolor* were the most sensitive among the isolates tested, being inhibited (>95%) already at 30% dose. In addition, sclerotia formation by *S. rolfsii* was also completely inhibited already at 20% dose. *Phytophthora cactorum* and *Penicillium digitatum* were inhibithed at 50% dose, while *Sclerotinia sclerotiorum*, *Botrytis cinerea*, *Trichoderma harzianum* and *T. gamsii* only showed >90% inhibition at the maximum dose (80%). *Fusarium verticillioides*, *Alternaria alternata* and *Phoma betae* were the least sensitive, showing inhibition of 20-29% as a maximum. Finally, an abnormal growth was observed for *Pythium ultimum*, with sparse hyphae, almost completely inhibited at 80% dose. A fungicidal effect was observed for *S. rolfsii* at 70% dose, while the maximum dose was also lethal for *T. versicolor* and *P. cactorum*.

STREPTOMYCETES AGAINST FUSARIA: LIMITING TOXIN PRODUCTION AND FUNGAL GROWTH. E.M. Colombo, A. Kunova, C. Pizzatti, E. Burrone, P. Cortesi, M. Saracchi, M. Pasquali. *Università degli Studi di Milano, DeFENS - Dipartimento di Scienze per gli Alimenti, la Nutrizione e l'Ambiente, via Celoria 2, 20133 Milano, Italy. E-mail: elenamaria.colombo@unimi.it*

Fusarium Head Blight is a disease caused by different *Fusarium* species on small-grain cereals, reducing yield and affecting quality of the grains. In addition, grains can be contaminated by mycotoxins, among them trichothecenes, which can lead to several problems for human and animal health. Within an integrated pest management strategy, the use of biocontrol agents is a promising approach. The aim of this research was to characterize and select potential biocontrol agents belonging to the genus *Streptomyces*, isolated from soil or roots of various crops, in order to discover strains able to inhibit the growth and toxin production of eight strains (covering geographic and toxigenic diversity) of *Fusarium graminearum* and *F. culmorum*. The antagonistic activities of 47 isolated bacterial strains against the pathogenic fusaria were firstly investigated by a dual-culture plate method, using different media. Eighteen strains exhibited >50% growth inhibition in at least one media. The higher inhibition percentages ranged, depending on the medium, between 55 and 79%. The most promising isolates were identified by sequencing of 16S rDNA. Eventually, the strains showing the most consistent antagonistic activity were chosen to assess their plant growth promoting activity and their influence on the disease severity in wheat seedlings. The identification of the molecules involved in toxin regulation and fungal inhibition is foreseen. The obtained results are promising and confirm the interest to investigate natural diversity of actinomycetes for identifying novel methods to reduce mycotoxin production in the cereal-*Fusarium* spp. pathosystem.

LOOKING FOR AN EFFECTIVE GENETIC RESISTANCE AGAINST WHEAT LEAF SPOT GROUP PATHOGENS. B. Corsi[1], S. Holdgate[1], I. Mackay[1], the Efecta Wheat Consortium[2], P. Kock Appelgren[3], J. Gerard Hehir[3], J. Cockram[1]. *[1]NIAB, Huntington Road, Cambridge, CB3 0LE, United Kingdom. [2]Lise Nistrup Jørgensen, Annemarie Justesen (AARHUS, Denmark), Min Lin, Morten Lillemo (NMBU, Norway), Andrea Ficke (Norwegian Institute of Bioeconomy Research, Norway), Pao Theen See, Richard Oliver (Curtin University, Australia), Judith Turner (FERA, UK), Melanie Stadlmeier, Volkher Mohler, Lorenz Hartl (LfL, Germany). [3]Teagasc, Oak Park Crops Research Centre, Carlow, Republic of Ireland. E-mail: james.cockram@niab.com*

The wheat leaf spot group (LSG) of necrotrophic pathogens [*Parastagonospora nodorum* (*Pn*) cause of Septoria nodorum blotch; *Zymoseptoria tritici* (*Zt*) cause of Septoria tritici blotch and *Pyrenophora tritici-repentis* (*Ptr*), cause of tan spot] have been the cause of the most common foliar disease in UK wheat over the last twenty-eight years. The implementation of effective genetic disease resistance represents an important route towards increasing sustainable wheat production. As part of the ERA-CAPs funded project 'EfectaWheat' we are using an 8-parent Multi-parent Advanced Generation Inter-Cross (MAGIC) genetic mapping population, genotyped with an Illumina 90k single nucleotide polymorphism (SNP) array, to investigate the genetics of LSG resistance in wheat. Representative UK *Pn* and *Ptr* isolates were used to inoculate MAGIC field trials in 2016 and 2017; for *Zt* a natural infection was used instead. Disease severity and additional phenotypic data were scored. The combined datasets will be used in genetic analyses to identify QTLs and possible relationships between wheat genetic loci controlling effectors- and pathogen-mediated host resistance/susceptibility. Ultimately, this information will provide useful tools for the development of improved wheat cultivars with increased genetic resistance to LSG pathogens, helping to support sustainable wheat production in Europe.

EVALUATION OF "ANCIENT" SOFT AND DURUM WHEAT VARIETIES FOR THEIR TOLLERANCE TO *SEPTORIA TRITICI* BLOTCH (STB). L. Cottafavi, F. Cavina, M.T. Senatore, P. Nipoti, A. Prodi. *Department of Agricultural Sciences, Alma Mater Studiorum University of Bologna, Viale G. Fanin 44, 40127 Bologna, Italy. E-mail: antonio.prodi@unibo.it*

Septoria tritici blotch (STB) is one of the most damaging diseases of wheat and it is caused by *Zymoseptoria tritici* (also known as *Septoria tritici*). This study was conducted to evaluate the STB tolerance of several "ancient" Italian varieties of soft (Andriolo, Frassineto, Inallettabile, Verna) and durum (Senatore Cappelli, Timilia, Russello) wheat comparing their STB response with those of common commercial varieties: Tirex and Lepido (durum wheat); Bolero (soft wheat). In greenhouse experiments, plants were artificially inoculated by *Z. tritici* strains previously isolated from durum and soft wheat (strains Zs.D1 and Zs.S1, respectively). In literature it is reported that the majority of durum wheat varieties are highly resistant to most *Z. tritici* isolates from soft wheat and vice versa. The results obtained in this experiment confirmed this hypothesis only for the soft wheat varieties. They showed a high level of tolerance to Zs.D1 strain except Verna. Among the soft "ancient" tested varieties, Andriolo was the more resistant (no disease) followed by Inallettabile (5.6% of incidence) and Frassineto (11.1% of incidence). The soft commercial variety Bolero showed a lower incidence than durum varieties but higher than "ancient" soft wheat varieties, except Verna. All tested varieties, including "ancient" ones, inoculated with Zs.S1, demonstrated a high disease incidence. Among them Senatore Cappelli showed the lowest incidence (45.6%) and also a low gravity (8.1%), similar to the durum commercial varieties Tirex (5.6%) and Lepido (11.9%). This is the first preliminary work regarding the response to STB of "ancient" Italian wheat varieties.

DEVELOPMENT OF A RAPID PCR NUCLEIC ACID LATERAL FLOW IMMUNOASSAY (PCR-NALFIA) BASED ON rDNA IGS SEQUENCE ANALYSIS FOR THE DETECTION OF *MACROPHOMINA PHASEOLINA* IN SOILS. D. Da Lio, G. Puntoni, S. Pecchia. *Università di Pisa, DISAAA-a, Via del Borghetto 80, 56124 Pisa, Italy. E-mail: susanna.pecchia@unipi.it*

Macrophomina phaseolina is a soil- and seed-borne generalist fungal pathogen with a global distribution and can infect more than 500 plant species. In infected tissues the fungus produces microsclerotia which enable it to survive in soil for 2-15 years, act as primary inoculum source and are needed for the correct identification of the pathogen. The disease occurrence and severity are directly related to the population of viable microsclerotia in soil. The 'Nucleic Acid Lateral Flow Immunoassay' using a generic 'Lateral Flow Device', combined with PCR employing labelled primers to detect a specific amplicon, enables to circumvent the use of electrophoresis, making the diagnostic procedure faster and easier. This study describes the development of the species-specific primers MP102F/MP102R for *M. phaseolina* based on the intergenic spacer (IGS) of the rDNA sequence analysis. The primer specificity was checked and confirmed using 20 isolates of the pathogen and other 16 non-target fungi. Microsclerotia of *M. phaseolina* (1, 10, 100 and 200) were manipulated under a stereomicroscope and their DNA was extracted from microsclerotia alone or mixed with different types of soil. The resulting DNA, used in the PCR-NALFIA assay, provided positive results for all the samples tested. A semi-quantitative grey-scale reference card was developed using intervals corresponding to microsclerotia soil number. For this purpose, the normalized pixel grey volumes obtained after a densitometric analysis of the test line intensity generated by the LFD strips were used. Patent application relating to the method here described is pending.

TOWARDS A SUSTAINABLE STRATEGY FOR *XYLELLA FASTIDIOSA* CONTROL. G. D'Attoma[1,2], M. Morelli[2], S. Ciccо[3], M. Saponari[2], P. Saldarelli[2]. *[1]Università degli Studi di Bari Aldo Moro, Dipartimento di Scienze del Suolo, della Pianta e degli Alimenti, 70126 Bari, Italy. [2]CNR-Istituto per la Protezione Sostenibile delle Piante (IPSP), 70126 Bari, Italy. [3]CNR-Istituto di Chimica dei Composti Organo Metallici (ICCOM), 70126 Bari, Italy. E-mail: giusy.dattoma@ipsp.cnr.it*

Xylella fastidiosa (*Xf*) is a xylem-limited bacterium, regulated as a quarantine pest, that is causing a devastating disease on olive crops in the southern area of Apulia (Italy) and whose potential spread in the Mediterranean area poses a severe threat to EU agriculture and landscape environment. *Xf* virulence is related to the expression of a cluster of *rpf* (regulation of pathogenicity factors) genes responsible for a signalling system based on small fatty acid molecules called DSF (Diffusible Signalling Factor). Since DSF regulation is involved in pathogenicity traits of *Xf* and biofilm formation, a "pathogen-confusion" strategy, based on the alteration of DSF levels *in planta*, has been proposed to contrast bacterial infection. In grapevine, the strategy is based on the transgenic expression of the *rpfF* gene, which encodes the DSF-synthase. We are attempting to express the *rpfF* gene of the olive-infecting *Xf* strain CoDiRO in the heterologous *Escherichia coli* system. The gene product has been successfully detected by Western blot analysis in cell protein extracts. Chemical characterisation by Gas Chromatography-Mass Spectrometry analysis of the DSF molecules produced by this expression system, in addition to those naturally produced by *Xf* CoDiRO, are underway. Concurrently, a TMV-based vector has been engineered to harbour the same *rpfF* gene and induce its transient expression *in planta*. Biologically active transcripts of the vector have been inoculated to *Nicotiana tabacum* and *N. benthamiana* plants, to establish a model system on herbaceous hosts. *RpfF* expression was successfully proved by Western blot analysis, whereas movement and systemic colonisation of plant tissues were evaluated by RT-PCR assays. The same viral vector harbouring GFP in replacement of *rpfF* is used as a control. Following inoculation with *Xf* CoDiRO bacterial cells the system is now being tested to monitor the persistence of DSF expression and its efficacy to lower disease susceptibility or movement of bacterial cells behind the point of inoculation.

A COMBINED APPLICATION OF A BIOPOLYMER AND *TRICHODERMA* – A PROMISING TOOL TO CONTROL ROOT-KNOT NEMATODES. G. d'Errico[1], F. Lacatena[2], N. Lombardi[1], G. Murolo[3], C. Gigliotti[3], G. Manganiello[1], M. Malinconico[4], P. Mormile[5], S. Bolletti Censi[3], A. Vassetti[1], F. Vinale[1,2]. *[1]Consiglio Nazionale delle Ricerche- Istituto per la Protezione Sostenibile delle Piante (CNR-IPSP), Via Università 133, 80055 Portici (NA). [2]Università degli Studi di Napoli Federico II- Dipartimento di Agraria, Via Università 100, 80055 Portici (NA). [3]Marea Scarl, Via Vittoria Colonna 14, 80121 Napoli. [4]Consiglio Nazionale delle Ricerche - Istituto dei Polimeri, Compositi e Biomateriali (IPCB), Via Campi Flegrei 34, 80078 Pozzuoli (NA). [5]Consiglio Nazionale delle Ricerche - Istituto di Scienze Applicate e Sistemi Intelligenti(ISASI), Via Campi Flegrei 34, 80078 Pozzuoli (NA). E-mail: francesco.vinale@ipsp.cnr.it*

In recent years, there has been growing interest in the use of biopolymers for a wide range of agricultural applications. A polysaccharide-based compound may be a good carrier of biological agents or act as a barrier against pathogens due to its ability to form films coating plant structures. In the present study we evaluate the combination of a biopolymer and *Trichoderma* strains for the management of disease caused by the root-knot nematode *Meloidogyne incognita* on tomato. Previous experiments have indicated that *Trichoderma* species are able to act both as crop protection agent and plant growth promoters. A greenhouse test was performed by dipping the roots in an aqueous solution of the biopolymer, which were then air-dried prior to planting in either naturally infested or sterilized soil. Spore suspensions of selected *Trichoderma* strains were watered at the time of transplant and every two weeks for the next two months. Preliminary results indicate that root treatment with the polysaccharide mixture combined with the *Trichoderma* soil amendment, significantly improves plant growth and development in comparison to controls. Evaluation is ongoing to determine if the effect is due to inhibition of *M. incognita*, demonstrated by a decreased nematode infestation and reduction in galling, and/or increased plant general fitness. The applied biopolymer may function as a physical barrier against the nematode and/or as a substrate supporting the colonization of the rhizosphere by plant growth-promoting beneficial microbes.

Work supported by MAREA project (MIUR PON03PE_00106_1).

PEPTAIBOLS: NATURALLY OCCURRING PEPTIDES AS BIOFUNGICIDES. M. De Zotti[1], L. Sella[2], A. Bortolotto[1,2], I. Elmaghraby[2], F. Favaron[1]. *[1]Department of Chemistry, University of Padova, Via Marzolo 1, 35131 Padova, Italy. [2]Department of Land, Environment, Agriculture and Forestry, University of Padova, Viale dell'Università 16, 35020, Legnaro (PD), Italy. E-mail: marta.dezotti@unipd.it*

Fungi belonging to the genus *Trichoderma* are distributed worldwide and have been successfully used in field trials against many crop pathogens. They produce peptaibols, a peculiar family of peptides, as part of their defense system against other microorganisms. Such secondary metabolites are known for their plant-protection properties: they (i) possess antimicrobial activity, (ii) act as stimulants of plant defences and growth, (iii) elicit plant production of volatiles to attract natural enemies of herbivorous insects. Moreover, peptides are ecofriendly compounds that are degraded by enzymes to nontoxic amino acids. The large molecular diversity of

this group of peptaibols is particularly interesting to design new active substances against plant pathogens. Several newly synthesized peptides, analogs of a natural peptaibol, have been tested, alone or in combination, *in vitro* against some important plant pathogens such as *Plasmopara viticola*, *Botrytis cinerea*, *Penicillum italicum*, *P. digitatum*, *P. expansum* and the bacterium *Pectobacterium carotovorum*. We found significant differences in the biocidal activity among the synthesized peptaibols and some of them resulted particularly effective against these pathogens at low micromolar concentrations.

TWO NEW *PHYTOPHTHORA* SPECIES PATHOGENIC TO CITRUS IN VIETNAM. M. Evoli[1,2], F. La Spada[1], F. Aloi[1], B. Scanu[3], D. Ruano-Rosa[4], M. Horta Jung[5,6], S. Wright[7], A. Pane[1], G.E. Agosteo[2], L. Schena[2], G. Magnano Di San Lio[2], T. Jung[5,6], S.O. Cacciola[1]. *[1]Department of Agriculture, Food and Environment, University of Catania, Catania, Italy. [2]Dipartimento di Agraria, Università Mediterranea di Reggio Calabria, Feo di Vito, Reggio Calabria, Italy. [3]Dipartimento di Agraria, University of Sassari, Viale Italia 39, 07100 Sassari, Italy. [4]Department of Crop Protection, Institute for Sustainable Agriculture (IAS), Spanish National Research Council (CSIC), Alameda del Obispo s/n, P.O. Box 4084, 14080 Córdoba, Spain. [5]Phytophthora Research Center Mendel University, Zemedelska 1, 613 00 Brno, Czech Republic. [6]Phytophthora Research and Consultancy, Am Rain 9, Nußdorf, Germany. [7]Department of Electronics, Mathematics and Natural Sciences, University of Gavle, Gavle, Sweden. E-mail: olgacacciola@unict.it*

In December 2012, during a survey aimed at identifying *Phytophthora* species infecting tropical fruit crops in the Mekong River Delta (southern Vietnam), two new species, *Phytophthora mekongensis* and *P. prodigiosa*, of ITS clade 2 and 9, respectively, were recovered from fruits of pomelo (*Citrus grandis*) with symptoms of brown rot and from pomelo and 'King' mandarin (*Citrus nobilis*) trees on volkamer lemon (*C. volkameriana*) rootstock. Both species were separated from closely related species of the same clades by sequencing the ITS1-5.8S-ITS2 region of the rDNA and the cytochrome oxidase subunit 1 (COX1). In pathogenicity tests, *P. mekongensis* and *P. prodigiosa* incited brown rot on fruits of various *Citrus* species, including pomelo, grapefruit, lemon, bergamot and sweet orange 'Tarocco' and 'Valencia'. Moreover, on artificially inoculated stems of sweet orange (*C. sinensis*) and lemon (*C. limon*) *P. mekongensis* induced bleeding cankers and was as virulent as *P. citrophthora* and *P. nicotianae* isolates obtained from citrus trees in southern Italy. By contrast, *P. prodigiosa* and heterologous *P. nicotianae* isolates from mandevilla (*Mandevilla* x *amoena*) and lavender (*Lavandula* sp.) were weakly pathogenic on stems of citrus trees. The identification of two new species pathogenic to citrus in southern Vietnam has practical implications for quarantine, as export of citrus fruit to other countries where these *Phytophthora* species have not been reported could be subjected to restrictions.

CHITOSAN NANOPARTICLES ARE BETTER THAN RAW CHITOSAN IN THE CONTROL OF *FUSARIUM GRAMINEARUM*? A COMPARATIVE *IN VITRO* AND *IN PLANTA* STUDY. F. Faoro[1], M. Fattizzo[1], S. Gobbi[1], V. Picchi[2]. *[1]Department of Agricultural and Environmental Sciences, University of Milan, 20133 Milano, Italy. [2]CREA-IT, Research Centre for Engineering and Agro-food Processings, 20133 Milano, Italy. E-mail: franco.faoro@unimi.it*

Chitosan (Cs), a deacetylated chitin derivative, besides being a well known plant resistance inducer is also a fungitoxic compound widely used in pre- and post-harvest fungal infections. Recently, nanoparticles prepared from chitosan (Cs-NPs) have proved to be fungitoxic as well, with the added value to be potential carriers of agrochemicals. However, it is not known if they are also able to induce plant defence mechanisms. In this study we attempted to control a *Fusarium graminearum* strain GFP-engineered both *in vitro* and *in planta* (durum wheat), by treatment with Cs, Cs-NPs and Cs-NPs loaded with the antioxidant N-acetyl cysteine (NAC), with the aim to shed light on the mechanisms of Cs-NP activity. Cs-NP were prepared by ionotropic gelation from a 0.5% Cs solution containing or not 0.3 mg/ml of NAC. Cs, Cs-NP and Cs-NP-NAC ability to inhibit mycelium growth and sporidia germination *in vitro* were tested at different concentrations. Furthermore, they were applied to durum wheat ears during flowering, before or after the inoculation of spikelets with *F. graminearum*. In the *in vitro* experiments, all treatments showed a reduction of radial mycelial growth in respect to control, without significant differences among them. However, *in planta* results were less clear as Cs-NP-NAC, contrary to Cs and Cs-NPs were not able to inhibit fungal infection. Possibly, the antioxidant activity of Cs-NP-NAC hampers the capacity of Cs and unloaded Cs-NPs to control the fungus, by neutralizing the Cs-induced micro-oxidative bursts responsible for the activation of plant defence.

BIOINFORMATIC ANALYSIS OF THE p23 PROTEIN OF A WORLDWIDE COLLECTION OF *CITRUS TRISTEZA VIRUS* REVEALS MINOR DIFFERENCES OF NUCLEOTIDE SEQUENCES. R. Ferraro[1], G. Scuderi[2], A. Catara[1], G. Licciardello[2]. *[1]Parco Scientifico e Tecnologico della Sicilia, ZI Blocco Palma I, Via V. Lancia 57, 95121 Catania, Italy. [2]Agrobiotech, ZI Blocco Palma I, Via V. Lancia 57, 95121 Catania, Italy. E-mail: glicciardello@agrobiotech.it*

Some isolates of *Citrus tristeza virus* (CTV) induce a seedling yellows (SY) reaction on certain citrus hosts. In view of a proficient management of this disease, a bioinformatic analysis has been carried out comparing seven full genomes of CTV strain, selected in Sicily, with 43 genomes of the virus, representative of five genotypes, available in the NCBI database. A Maximum Likelihood (ML) method was applied to compare predicted complete sequences of p23 protein and its three regions previously identified for differentiation of isolates (24-29, 50-54 and 78-80) using MEGA 6.0 software, based on the isoelectric point (pI) and molecular weight (Mw), calculated with pI/Mw tool from server ExPASy. There are significant differences in the mean value of pI among VT-like, T30-like and T36-like groups, respectively: 6.10, 8.59 and 4.87 were observed only in the first region (24-29 aa). Moreover, the Sicilian isolate Mac39 (asymptomatic VT-like) differed from the VT isolates inducing severe SY only for one mutation at position 54 (from valine to isoleucine). Mac25, a T36-like isolate, showed a p23 protein similar to the VT-like group according to the first and second aminoacidic regions, while in the third region had Gly^{78} (G), Leu^{79} (L) and Lys^{80} (K) like atypical isolates. The minor nucleotide differences detected in the large number of CTV genome analyzed indicate that the seedling yellows phenotype is not determined by a specific factor of p23, and confirms the hypothesis that specific host factors are involved in the phenotypic expression.

GLIOTOXIN BIOSYNTHESIS IN *TRICHODERMA* spp. L. Fiorini[1], A. Gianoncelli[1], M. Bertuzzi[1]. R. Baroncelli[2], S. Sarrocco[3], G. Vannacci[3], E. Gobbi[1]. *[1]Università degli Studi di Brescia, Piattaforma di Microbiologia Agro-alimentare ed Ambientale, Dipartimento di Medicina Molecolare e Traslazionale, Viale Europa 11, 25123 Brescia (BS), Italy. [2]Laboratorie Universitaire de Biodiversité et Ecologie Microbienne, Université de Bretagne Occidentale, Brest, France. [3]Università di Pisa, Dipartimento di Scienze Agrarie, Alimentari e*

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Fungal biological control agents (BCAs) are a sustainable alternative for crops protection. Several BCAs are reported to produce metabolites that have antibiotic, fungicidal, insecticidal or antiviral properties. However, some of these metabolites, such as gliotoxin (GT), can be toxic to animals, humans included. Gliotoxin is a toxin produced by the "Q" strains of *Trichoderma virens*, with a role in the mycoparasitism of other fungi. Beside its potential use in biocontrol applications, GT is also involved in the virulence of the opportunistic human pathogen *A. fumigatus*. Thus, apparently, GT can represent either a beneficial or a deleterious fungal metabolite depending on the situation. More information about GT production by BCAs and its environmental role should be gained in order to minimize the GT 'load' in the environment and for the safe use of *Trichoderma* spp. as a biofungicide. Three different *Trichoderma* species have been analyzed and the presence of the gliotoxin biosynthetic genes cluster has been ascertained in the isolates *T. virens* Gv29-8, known to be a GT producing strain, *T. reesei* RUT-C30 and QM6a, known to be GT non-producing strains and *T. afroharzianum* T6776 and B97, for which no information about GT production was available. A time course of their GT production *in vitro* has been produced and GT quantified by HPLC analysis. Moreover, the induction of GT biosynthesis by plants or other fungi has been evaluated either by HPLC and *gli* cluster gene expressions analysis.

DEVELOPMENT OF REAL TIME RT-qPCR FOR THE DETECTION OF OLIVE LEAF YELLOWING ASSOCIATED VIRUS. A. Fontana, A. Tiberini, G. Albanese. *Università degli Studi Mediterranea di Reggio Calabria, Dipartimento di AGRARIA, Località Feo di Vito, 89122 Reggio Calabria (RC) Italy. Email: anna.fontana@unirc.it*

Olive leaf yellowing associated virus (OLYaV, an unassigned virus in the family *Closteroviridae*) after the first detection in Italy was found in many orchards of southern Italy as well as in other Mediterranean countries. Detection protocol of olive viruses needs repeated analyses for several subsequent spring and autumn seasons due to the erratic distribution in olive plants. In the frame of PON3PE00090_2 OLIVO research project, a Real-Time RT-qPCR assay specific for OLYaV was developed. Primer sets were designed on published nucleotide sequences of HSP70 by Primer Express software (Applied Biosystems) and applied in a Real-Time RT-qPCR assay using a Power SYBR® Green RNA-to-CT™ 1-Step Kit. Reactions were performed in a StepOnePlus™ Real-Time PCR System with a standard amplification cycle including melting curves analysis to exclude aspecific amplicons. Analytical specificity was assessed *versus* other viruses phylogenetically related to OLYaV (*i.e. Citrus tristeza virus*, genus *Closterovirus*) and/or other viruses reported on olive, as *Strawberry latent ringspot virus* (genus *Nepovirus*). No amplification products were detected for the non-target viruses analyzed. Moreover, analytical sensitivity was determined using 5-fold dilution series of OLYaV-infected olive leaf extracts. The analytical sensitivity has been directly compared with end point RT-PCR. Moreover, to assess a relative quantification method based on a ΔΔCt method, different reference genes, commonly used and reported in literature, were compared and validated to be successfully applied to normalize the relative quantification data allowing to investigate the modulation of virus titer in the pathosystem OLYaV-olive over the project research.

NEW FUNGI ATTACKING POMEGRANATE (*PUNICA GRANATUM*) IN ITALY. S. Frisullo[1], S.M. Mang[2], H.S. Elshafie[2], I. Camele[2]. *[1]Department of Agricultural Sciences, Food and Environment, University of Foggia, Via Napoli 25, 71121 Foggia, Italy. [2]School of Agricultural, Forestry, Food and Environmental Sciences, University of Basilicata, Viale dell'Ateneo Lucano 10, 85100 Potenza, Italy. E-mail: ippolito.camele@unibas.it*

Symptoms of chlorosis, decay, leaves falling and plant death were observed in autumn 2015/16 in commercial orchards on pomegranate (*Punica granatum* L.) in Apulia and Basilicata regions (southern Italy). In some cases the disease incidence reached 20%. Sections of the trunks taken from symptomatic plants revealed vessels browning. In order to isolate the likely pathogens, sterilized pieces of symptomatic tissues were plated in Petri dishes containing potato dextrose agar (PDA). Three fungal species were isolated consistently on PDA. Total DNA of three isolates for each species was extracted and the ITS1-5.8S ITS2 region was amplified with ITS5/ITS4 primers and sequenced. On the basis of morphological features and ITS sequence, isolates were identified as *Xenoacremonium falcatus*, *Sporotrix variecibatus* and *S. mexicana*. The ITS sequences of one isolate for each species were deposited in GenBank under the accession Nos. LT799728-LT799729-LT799730. Thirty healthy twigs of pomegranate (10 cm length) for each fungal species, previously surface sterilized, were wound-inoculated with a single fungal pure culture on agar (0.3 × 0.3 cm) and then placed in a moist chamber at 25°C. Ten controls were agar-inoculated using the same procedure. After 15 days the pathogen inoculated twigs showed vessels browning until 3-4 cm length from the inoculation zone. *X. falcatus*, *S. variecibatus* and *S. mexicana* were always reisolated only from twigs individually inoculated with the above fungi. The occurrence of *X. falcatus*, *S. variecibatus* and *S. mexicana* is reported for the first time on *P. granatum* plants in Italy.

OCCURRENCE OF *COLLETOTRICHUM ACUTATUM* ON *ACCA SELLOWIANA* IN ITALY. S. Frisullo[1], S.M. Mang[2], H.S. Elshafie[2], I. Camele[2]. *[1]Department of Agricultural Sciences, Food and Environment, University of Foggia, Via Napoli 25, 71121 Foggia, Italy. [2]School of Agricultural, Forestry, Food and Environmental Sciences, University of Basilicata, Viale dell'Ateneo Lucano 10, 85100 Potenza, Italy. E-mail: ippolito.camele@unibas.it*

In summer-autumn 2015/16, a severe typical symptoms of anthracnose with an incidence of 10 to 20% were observed on feijoa [*Acca sellowiana* (*O. Berg*) Burret] mature fruits and leaves in gardens and commercial orchards (about 35 hectares) located in Bari and Matera Provinces (southern Italy). The symptoms were dark sunken necrotic lesions on mature fruits which may become covered by pink conidial masses in the center with successively fruit rot. On the leaves, chlorotic lesions that evolved to brown spots and sunken necrotic zones were observed. In order to isolate the likely pathogen, small pieces of symptomatic plant tissues were plated in Petri dishes containing potato dextrose agar (PDA). A *Colletotrichum* species was isolated consistently on PDA. Conidia produced by the pure isolates were hyaline, smooth-walled, aseptate and cylindrical to fusiform. On the basis of colony and conidia morphology, isolates were identified as *Colletotrichum acutatum* J.H. Simmonds. Ten *C. acutatum* isolates were identified through both morphological and molecular analysis based on a multilocus investigation of four genes: the rDNA-ITS region (ITS), partial actin (ACT), β-tubulin (TUB-2) and glyceraldehyde-3-phosphate dehydrogenase (GAPDH). Phylogeny inferred from combined datasets of ACT, ITS, β-tubulin and GAPDH revealed its belonging to *C. acutatum*. Pathogenicity tests were carried out twice. Symptoms identical to those observed in the field appeared within 10 to 15 days. *C. acutatum* was always isolated only from inoculated leaves and fruits. The occurrence of

C. acutatum is reported for the first time on *A. sellowiana* plants in Italy.

PRELIMINARY INVESTIGATIONS ON PRESENCE OF FUNGI ON OLIVE TREES INFECTED AND APPARENTLY UNINFECTED BY *XYLELLA FASTIDIOSA* STRAIN CoDiRO IN LECCE PROVINCE (SOUTHERN ITALY). **S. Frisullo[1], S.M. Mang[2], H.S. Elshafie[2], L. Prudente[1], I. Camele[2].** *[1]Department of Agricultural Sciences, Food and Environment, University of Foggia, Via Napoli 25, 71121 Foggia, Italy. [2]School of Agricultural, Forestry, Food and Environmental Sciences, University of Basilicata, Viale dell'Ateneo Lucano 10, 85100 Potenza, Italy. E-mail: ippolito.camele@unibas.it*

In order to verify the presence and frequency of fungi on olive trees infected and apparently not infected by *Xylella fastidiosa* strain CoDiRO, a survey was carried out in 20 farms located in Lecce province (southern Italy) in spring/summer 2015/2016. For this scope, 20 two-year-old twigs were collected from apparently uninfected and *X. fastidiosa* infected olive trees, respectively. Three sections were performed on each twig taken from different height lengths. Both under bark and woody cylinder parts were used for fungal isolation on Potato Dextrose Agar (PDA). Other 10 branches, three- to five-year old, from infected and from apparently non infected olive trees, respectively, were used for fungal isolation only from woody cylinder part (3 sections/branch) on PDA. Outcomes of this investigation showed that a higher number of genera and fungal isolates were present on apparently uninfected olive trees, from both twigs and branches, compared with the *X. fastidiosa* infected samples. The fungal genera more frequently isolated from apparently healthy olive trees were *Botryosphaeria*, *Colletotrichum*, *Alternaria*, *Phaeoacremonium*, *Phaeomoniella* and *Phialophora*. The *X. fastidiosa* role in the bacteria/fungi pathosystem will require further investigations.

This study was supported by Apulia Region under the project "EZIOCONTROL".

INVESTIGATION OF THE ROLE OF HISTIDINE KINASE 1 IN *STEMPHYLIUM VESICARIUM*. **K. Gazzetti, A. Ciriani, M. Collina.** *University of Bologna, Department of Agricultural Science, Viale G. Fanin 46, 40127 Bologna (Italy). E-mail: marina.collina@unibo.it*

Brown spot of pear (BSP), a fungal disease caused by *Stemphylium vesicarium* (Wallr.) Simmons, is the most important pear pathogen in Italy and in other European countries. Preventive applications of fungicides are needed from petal fall to fruit ripening to control the disease. A previous molecular analysis conducted by our group on field isolates of *S. vesicarum*, established a correlation between dicarboximide and phenylpirroles fungicide resistance and single amino acid substitutions, corresponding to single nucleotide polymorphisms (SNPs) in the nucleotidic sequence of *SvHK1*. This gene is predicted to encode a 1,329 amino acid protein, putatively belonging to the class III of two-component histidine kinases, homologous to Nik1/OS1 from *Neurospora crassa*. *SvHK1* orthologues are involved in fungicides resistance, osmotic and oxidative stress response, hyphal growth, development and differentiation, nutrient-sensing pathways, pathogenicity mechanisms. Gene deletion mutants were generated by gene replacement from protoplasts obtained from a reference sensitive strain (WT), and monoconidial mutant strains were characterized in order to collect insights on the role of HK1 in *S. vesicarium*. The whole genome of WT strain has been *de novo* sequenced and is currently available on our private Online Personal BLAST Server, together with an *ab initio* gene prediction. The database is very helpful to identify the components of signaling pathway to which SvHK1 belongs, and his relationship with other signal cascades.

PRODUCTION OF ACTIVE PROPAGULES OF BENEFICIAL MICROBES USING AGRO-FOOD WASTES. **C. Gigliotti[1], M. Consales[1], G. Murolo[1], F.L. Fedele[1], M. Pascale[2], S. Lanzuise[2], E. Comite[2], M. Napolitano[2], A. Sicari[1], S. Bolletti Censi[1], M. Lorito[2].** *[1]Linfa S.C.A.R.L., Zona Industriale Snc-89900 Porto Salvo Vibo Valentia (VV), Italy. [2]Università degli Studi di Napoli Federico II, Dipartimento di Agraria, Via Università 100, 80055 Portici (NA), Italy. E-mail: info@laboratoriolinfa.it*

Annually, the agro-food industry produces considerable amounts of by-products and waste materials that can cause serious environmental problems, with storage and disposal having an elevated cost. Bioactive molecules, such as antioxidants, vitamins, pigments, etc., are often present in high concentrations in industrial wastes, and they can be recovered for use in the formulation of functional foods. Extraction of valuable nutrients or recycling of the industrial by-products represent a valid alternative to simply discarding the waste. This work proposes an innovative application for undesirable material from industrial processing. Tomato or olive by-products were used as a growth medium for the cultivation of beneficial microbes, such as fungi belonging to the genus *Trichoderma*, that are used as the active ingredient of biofertilizers and biopesticides marketed worldwide for agricultural applications. Tomato or olive by-products, obtained from the food processing companies involved in the "Linfa" project, were treated in four different manners (sterile or nonsterile, with or without the addition of 1% sucrose) then inoculated with *Trichoderma* spp. Solid state fermentations were performed in air strip bags, incubated at 25°C for one month in the dark. Fungal growth and spore production were monitored every seven days. Generally, the fungus developed and sporulated abundantly on both tomato and olive residues, with the highest spore concentrations obtained when the substrate was sterilized and amended with sucrose. Further investigations using different microbes or growth conditions are in progress.

Work was supported by LINFA project (PON03PE_00026).

BEHAVIOR OF *FUSARIUM EQUISETI* ON LETTUCE AND WILD ROCKET: RESULTS FROM EPIDEMIOLOGY STUDIES UNDER CONTROLLED CONDITIONS. **G. Gilardi[1], A. Garibaldi[1], M.L. Gullino[1,2].** *[1]Università degli Studi di Torino, Centro AGROINNOVA, Largo P. Braccini 2, 10095 Grugliasco (TO), Italy. [2]Università degli Studi di Torino, DISAFA, Largo P. Braccini 2, 10095 Grugliasco (TO), Italy. E-mail: giovanna.gilardi@unito.it*

Fusarium equiseti is spreading quickly in Italy on different leafy vegetables, such as lettuce, wild and cultivated rocket, which are grown intensively in monoculture. In order to have a better understanding of the effect of temperature and relative humidity (RH) exposure on the severity of *F. equiseti*, trials have been carried out in growth chambers at 10, 15, 20, 25, 30 and 35°C at 100% RH. Twenty-five-day-old plants of lettuce and wild rocket were sprayed with 1×10^6 conidia/ml of the pathogen isolated from affected plants in field. Plants were kept at the different temperatures and covered with a transparent polyethylene film for 0, 1, 3, 6, 12, 24 and 48 hours. Disease incidence (DI) and severity (DS) were recorded 8 days after inoculation counting the number of affected leaves out of 100 and estimating the percentage of leaf area affected (DS). Results showed that *F. equiseti* severely affected lettuce quality

at 25 and 30°C. One hour at 30°C under high RH caused on lettuce a DI of 54.0 and DS of 23.9, while at 15°C at least 12 hours under high RH were necessary to cause a DI of 32.4 and DS of 10.1. For wild rocket DI and DS was higher at 30 and 35°C. Although only one hour at high RH was sufficient to cause DI 29.8 and DS 16.1 at 35°C, 24 hours were necessary to cause significant losses at 10°C. The present results provide evidence that *F. equiseti* could represent a serious threat for crops grown in the Mediterranean area.

ASSESSING THE EFFECTS OF HAIL WOUNDS ON THE RESURGENCE OF CHESTNUT BLIGHT IN A SITE IN NORTH-WESTERN ITALY. L. Giordano[1,2], G. Lione[1], P. Gonthier[1]. *[1]University of Torino, Department of Agricultural, Forest and Food Sciences (DISAFA), Largo Paolo Braccini 2, I-10095 Grugliasco (TO), Italy. [2]University of Torino, Centre of Competence for the Innovation in the Agro-Environmental Field (AGROINNOVA), Largo Paolo Braccini 2, I-10095 Grugliasco (TO), Italy. E-mail: paolo.gonthier@unito.it*

Recently, extensive diebacks of *Castanea sativa* Mill. in northwestern Italy have been observed in association with a resurgence of chestnut blight caused by the fungal pathogen *Cryphonectria parasitica* (Murrill) M.E. Barr. *C. parasitica* is a wound pathogen infecting its host through grafting and pruning wounds or mechanical injuries. Although hail wounds could potentially trigger new infections, very little is known on the effects of hailstorms on the severity of chestnut blight. In order to improve our understanding on this epidemiological aspect, symptomatic and asymptomatic branches of chestnut were sampled in 6 different plots in Peveragno (CN, north-western Italy). Isolations were performed and *C. parasitica* isolates were identified based on their culture morphology. Infected and uninfected branches were contrasted by comparing morphological and pathological features, including the number and extension of hail wounds. The same comparisons were performed between symptomatic and asymptomatic portions of infected branches. Although infected branches displayed an average number of hail wounds significantly lower than uninfected branches (31 *vs* 39; $p < 0.05$), the extension of such wounds was on average 3 times larger (22.78 mm^2 *vs* 6.92 mm^2; $p < 0.05$). Moreover, while symptomatic and asymptomatic portions of infected branches were comparable considering the average number of hail wounds (16 *vs* 15, $p > 0.05$), the average extension of wounds was significantly larger in the former than the latter (25.13 mm^2 *vs* 14.61 mm^2; $p < 0.05$). These results suggest that the extension of hail wounds, rather than their abundance, plays a key role in the epidemiology of *C. parasitica*.

FUNGAL INTERACTION IN MAIZE EARS IN FIELD DURING THE GROWING SEASON. P. Giorni[1], T. Bertuzzi[2], M. Camardo Leggieri[1], P. Battilani[1]. *[1]Dipartimento di Scienze delle Produzioni Vegetali Sostenibili, Area Protezione Sostenibile delle Piante e degli Alimenti. [2]Istituto di Scienze degli Alimenti e della Nutrizione, Università Cattolica del Sacro Cuore, Via Emilia Parmense 84, 29122 Piacenza, Italy. E-mail: paola.battilani@unicatt.it*

Maize is an important crop cultivated worldwide that may be infected by several mycotoxigenic fungi. They interact and the dominant fungus depends mainly on meteorological conditions during the growing season; other factors linked to the plant and the cropping system can contribute. In Italy, the major mycotoxigenic fungi found on ripening maize are *Aspergillus flavus*, *Fusarium verticillioides* and, to a lower extend, *F. graminearum*. These fungi produce different mycotoxins, in particular, aflatoxins (AFs), fumonisins (FBs) and deossinivalenol (DON), respectively. Many studies have been done on fungi in field, but poor information is available regarding co-occurrence of fungi and, consequently, on the effect of their interaction on mycotoxins production. In 2016, maize ears in a field crop were artificially inoculated after silk emergence with the three mentioned fungi, alone or in co-presence. Ear samples were collected every 14 days up to harvest time. When fungi were single-inoculated, the highest incidence resulted after 42 days for *F. verticillioides* and after 28 days for *A. flavus* and *F. graminearum*. When co-inoculated, fungi showed a different behaviour; *F. verticilliodes* showed the highest incidence after 28 days and *A. flavus* and *F. graminearum* only after 42 days, irrespective of the co-inoculated species. Regarding mycotoxins, *A. flavus* always produced more AFs when competitors were present while FBs and DON production was faster or increased only when *Fusaria* co-occurred. Data obtained will be useful to support/validate predictive models for fungal growth and mycotoxin production in field with co-occurring fungi, more frequent in recent years due to climate change.

SURVEY OF MYCOTOXIGENIC FUNGI AND MYCOTOXINS ON ITALIAN RICE USED FOR BABY FOODS DURING STORAGE. P. Giorni[1], T. Bertuzzi[2], M. Romani[3]. *[1]Dipartimento di Scienze delle Produzioni Vegetali Sostenibili, Area Protezione Sostenibile delle Piante e degli Alimenti. [2]Istituto di Scienze degli Alimenti e della Nutrizione, Università Cattolica del Sacro Cuore, Via Emilia Parmense 84, 29122 Piacenza, Italy. [3]Ente Nazionale Risi Research Centre, Strada per Ceretto 4, 27030 Castello d'Agogna (PV), Italy. E-mail: paola.giorni@unicatt.it*

Italy is the main rice producer in Europe covering about 50% of the European production. Depending on climate registered during the growing season and on environmental conditions during storage, mycotoxins contamination of rice may result higher than levels fixed by the European Commission (Regulaments CE 1881/2006, UE 165/2010, UE 488/2014 and UE 1006/2015). In particular, for aflatoxin B_1 (AFB_1) and deossinivalenol (DON), maximum amounts admitted are very low in rice (0.1 µg/kg and 200 µg/kg, respectively) becoming even more severe for rice destined to babyfoods. Since few data on contamination of Italian rice are available, a study was planned taking into account two different storage methods (traditional warehouse and refrigerated silos) and six different rice varieties. Samples were collected at the beginning and at the end of the storage period and, when possible, every three weeks for a maximum of four months. Samples were analysed for fungal incidence, with particular attention to *Fusarium* spp. and *Aspergillus* spp. and for AFB_1, DON and sterigmatocystin (STC), a mycotoxin recently signalled in rice, using HPLC-FLD, GC-MS and LC-MS/MS, respectively. *Fusarium* spp. were the most represented fungal species found in rice (incidence from 2 to 43%) but also *Aspergillus* spp. were found in several samples, in particular *A.* section *Flavi* (found in 1 variety), *A.* section *Nigri* (found in 2 varieties) and *A. versicolor* (found in all sampled varieties). Mycotoxins tested resulted always below legal limits, but a direct correlation occurs between DON and *Fusarium* spp. incidence and between STC and *A. versicolor* incidence.

BIOCONTROL OF THE BACTERIAL BLIGHT OF WALNUT: IS THERE A CHANCE TO REDUCE COPPER INPUTS INTO WALNUT GROVES? D. Giovanardi[1], L. Fagioli[2], L. Gilli[1], E. Stefani[1]. *[1]Università di Modena e Reggio Emilia, Dipartimento di Scienze della Vita, via Amendola 2, Pad. Besta, 42122 Reggio Emilia (Italy). [2]Consorzio Agrario di Ravenna, via Madonna di Genova 39, 48010 Cotignola (Italy). E-mail: davide.giovanardi@unimore.it*

The bacterial blight is a re-emerging disease, severely affecting the productivity of walnut groves. Disease symptoms are observed

on all aerial parts of the host plant, but the most damaging are the necrotic spots developing on fruits, leading to massive fruit drops, mainly before endocarp hardening. The causal agent is *Xanthomonas arboricola* pv. *juglandis* (*Xaj*), a gram-negative rod with a worldwide distribution. The population structure of *Xaj* includes several morphotypes, at least five sequence types, and other features confirming its genomic heterogeneity. Copper resistance is widespread and very effective among *Xaj* populations: therefore, disease management based on copper sprays is not particularly effective, even in cases of 10-14 treatments during the growing season. The recent, severe outbreaks reported in Italy are due to two concomitant events: i) the rapid increase of walnut acreage, especially in the northern part of the Country and ii) the development of *Xaj* populations showing high resistance to copper. Since most of the cultivated walnut varieties are either highly susceptible or susceptible to the disease, we tried to implement control strategies based on the use of a single antagonist or the field application of microbial consortia. Additionally, innovative agrochemicals, with a reduced copper content, have also been used. Results showed that: i) microbial biocontrol agents were able to significantly reduce the disease in the field; ii) innovative agrochemicals may reduce the bacterial blight, but might enhance phytotoxicity; iii) a significant reduction of copper inputs is possible, coupled with an effective disease control in walnut groves.

DISTIBUTION OF GRAPEVINE PINOT GRIS VIRUS IN HERBACEOUS HOST IN VINEYARD. V. Gualandri, P. Bianchedi, E. Asquini, A. Si-Ammour. *Fondazione Edmund Mach, Via E. Mach 1, 38010 S. Michele all'Adige, Trento. E-mail: valeria.gualandri@fmach.it*

Grapevine Pinot gris virus (GPGV, genus *Trichovirus*, family *Betaflexiviridae*) was reported for the first time in 2012 in Trentino region and is currently an emerging virus in *Vitis vinifera*. Symptoms of deformations, chlorosis and mottling on leaf and stunting of shoots ('grapevine leaf mottling and deformations disease' – GLMD) are always associated with the presence of GPGV, but sometimes the virus is detected in symptomless vines, too. The existence of different strains of the virus was demonstrated to be the probable responsible for eliciting the symptoms. Recently, the virus was also detected in wild herbaceous hosts collected in vineyards and showing chlorosis and mottling such as *Chenopodium album* and *Silene latifolia* subsp. *alba*. The full-length GPGV RNA genome was amplified from these infected hosts. With the aim to investigate and clarify the importance of weed species as a possible reservoir of GPGV, and their role in the epidemiology of the associated disease in vineyards, a screening on different weed species was carried out from 2012 to 2017 in GPGV infected vineyards at different locations in Italy. The survey included species composing the weed association along and between the vines rows, where GPGV affected plants were present. Symptomatic herbaceous plants were picked up and total RNA was extracted. The presence of GPGV was assessed in RT-PCR with specific primer pairs amplifying movement protein (MP) and coat protein (CP) genes and in RT-qPCR on MP gene.

SEVERITY OF *FUSARIUM EQUISETI* ON BRASSICA CROPS UNDER A CLIMATE CHANGE SCENARIO: RESULTS FROM SIMULATION IN PHYTOTRON. M.L. Gullino[1,2], G. Gilardi[1], A. Garibaldi[1]. *[1]Università degli Studi di Torino, Centro AGROINNOVA, Largo P. Braccini 2, 10095 Grugliasco (TO), Italy. [2]Università degli Studi di Torino, DISAFA, Largo P. Braccini 2, 10095 Grugliasco (TO), Italy. E-mail: giovanna.gilardi@unito.it*

Climate change can have an important effect on the phytosanitary situation of many hosts, and can lead to the quick spread of new pathogens, such as *Fusarium equiseti* on crops grown in succession. A study was undertaken under phytotron to evaluate the effects of temperature (from 14 to 30°C) and CO_2 (450 and 850 ppm) values and their interaction on the incidence and severity of *F. equiseti* on wild rocket and radish, that have been found as new hosts for this pathogen. A suspension containing 1×10^7 conidia/ml of the pathogen was sprayed onto 20 to 25-day-old plants, 7 days after their transfer to the phytotrons. *F. equiseti* on this hosts caused severe symptoms at the tested temperature regimes of 18-22°C, 22-26°C and 26-30°C, with significantly lower attacks at 14-18°C. At all the temperature regimes tested, the highest CO_2 value caused an increase in the percentage of affected leaves (DI) from 27 to 37 and percentage of affected leaf area (DS) from 11 to 18 on rocket, which resulted significant at 26-30°C. Symptoms on radish significantly increased from 14-18°C (DI 30.6 and DS 8.5) to 26-30°C (DI 47.0 and DS 18.5) at 850 ppm of CO_2, while DS resulted affected by the increase in CO_2 at 26-30°C DS (from 8.4 to 18.5). Results showed that the combination of the CO_2 and temperature factors significantly influence *F. equiseti* leaf spot on both hosts and add more concern to the possible negative effects on the spread of new pathogens.

DIFFERENTIAL GENE EXPRESSION PROFILES BETWEEN *FUSARIUM OXYSPORUM* f. sp. *MELONIS* RACE 1 AND 1,2 IN INFECTED GRAFTED MELON PLANTS. A. Haegi[1], M. Scotton[2], S. Vitale[1], L. Luongo[1], A. Belisario[1]. *[1]Consiglio per la Ricerca in Agricoltura e l'Analisi Economica (CREA) - Centro di Ricerca Difesa e Certificazione (CREA-DC), Via C.G. Bertero 22, 00156. [2]Dipartimento di Agronomia Animali Alimenti Risorse Naturali e Ambiente, Università di Padova, Viale dell'Università 16, 35020 Legnaro, Padova, Italy. E-mail: anita.haegi@crea.gov.it*

Fusarium oxysporum f. sp. *melonis* (FOM) is the causal agent of a devastating wilting disease of melon worldwide. Four races are known of this pathogen, 0, 1, 2, and 1,2, the latter the most widespread and virulent, being controlled by partial resistant melon cultivars or grafting on multigenic resistant rootstocks. To understand FOM-melon interaction and molecular mechanisms of the pathogen in grafted plants, expression of genes selected from a previous transcriptome analysis were analyzed by quantitative real-time PCR (qPCR) for their temporal transcription profiles. Infections were performed in a grafted combinatorial system with a resistant and susceptible melon genotype used in both combinations and gene profiles were investigated both in scion and rootstock at 2 and 18 days post inoculation (dpi). Eleven fungal genes related to pathogenicity were considered such as Zn-Cys transcription factor FOW2, xylanase, ROS degrading catalase/peroxidase enzyme and actin-binding protein. The expression of the selected genes varied according to the fungal race, susceptible or resistant interaction, and time, clustering into six profiles. Temporal transcription profiles of genes related to infection were consistent with the growth of FOM race 1,2 in the resistant combination and confirmed that plant colonization by race 1,2 is independent from virulence.

PORT.NOC PROJECT - STRATEGIES FOR THE MANAGEMENT OF PHYTOPHTHORA WALNUT DECLINE BY SOURCES OF RESISTANCE AND A ROBUST DETECTION METHOD. A. Haegi[1], S. Vitale[1], L. Ferretti[1], M. Scarpari[1], L. Luongo[1], L. Tizzani[1], M. Galli[1], M. Gras[2], G. Mughini[2], A. Belisario[1]. *[1]Consiglio per la ricerca in agricoltura e l'analisi economica (CREA) - Centro di ricerca Difesa e Certificazione (CREA-DC). [2]CREA - Centro di ricerca Foreste e Legno (CREA-FL). E-mail: alessandra.belisario@crea.gov.it*

Phytophthora spp. are serious soilborne pathogens of Persian (English) walnut, causing crown and root rot and quite dramatic production losses worldwide. Presently, eight species of *Phytophthora* have been implicated in common walnut decline and death in Italy, and among them *P. cinnamomi* is especially aggressive and difficult to manage. Several commercial walnut orchards have been decimated because of *Phytophthora* attacks incited by *P. cinnamomi* either in northern or in central Italy. As a response to *Phytophthora* attacks, different *Juglans* species other than *J. regia* are taken into account as rootstock. The Italian Ministry of Agriculture financially supported a project named PORT.NOC addressed to respond to this issue. For this purpose, *J. microcarpa* and *J. major* as well as their hybrids have been investigated as sources of resistance. The implementation of detection methods for *Phytophthora* diagnosis and quantification are also in progress. The presence of Cherry leaf roll virus in the breeding material is detected and the susceptibility to blackline of the eventual rootstocks will be assessed. Advances in micropropagation and breeding technologies facilitate the utilization of diverse *Juglans* species and hybrids for the improvement of walnut rootstocks.

TEST PERFORMANCE STUDY OF METHODS FOR THE DIAGNOSIS OF '*CANDIDATUS* LIBERIBACTER SOLANACEARUM' IN CARROT SEEDS. V. Ilardi, V. Lumia, E. Di Nicola. *CREA, Centro di ricerca Difesa e Certificazione, sede di Roma, Via C.G. Bertero 22, 00156 Rome, Italy. E-mail: vincenza.ilardi@crea.gov.it*

'*Candidatus* Liberibacter solanacearum' (CaLsol), a fastidious phloem-limited bacterium, is associated with several important diseases in crops belonging to Solanaceae and Apiaceae families. It has been shown to be seed-transmitted in carrot, and emergency measures for exportation in notable extra-European markets require that the carrot seed is PCR tested and found CaLsol free. Thus, the identification and harmonization of a protocol for CaLsol diagnosis in carrot seed are becoming of socio-economic priority. We set up a DNA extraction method for Apiaceae seeds and identified, among the widely used PCR tests to detect and identify CaLsol, one real-time PCR and one end-point PCR as the most sensitive ones. The two PCR methods were first intralaboratory validated followed by a "test performance study," involving 11 Italian laboratories of the Italian regional plant protection services. The DNA extraction method was effective for the molecular detection of CaLsol. Between the PCR-based methods, the real-time PCR was the most sensitive and specific showing the higher percentage of accordance and concordance when evaluated in interlaboratory comparison assay. The obtained results could be used for the appropriate application of phytosanitary measures for CaLsol detection and identification in Apiaceae seeds.

CHEMOTROPIC EFFECT ON SOIL MICROBES BY THE ROOTS OF PLANTS SUBJECTED TO BIOTIC AND ABIOTIC STRESSES. N. Lombardi[1], R. Marra[2], G. d'Errico[2], D. Turrà[3], M. Reverberi[4], M. Ruocco[1], B. Voltura[5], A. D'Angelo[1], S.L. Woo[2,5], M. Lorito[1,2]. *[1]Consiglio Nazionale delle Ricerche, Istituto per la Protezione Sostenibile delle Piante (CNR-IPSP), via Università 133, 80055 Portici (NA), Italy. [2]Università degli Studi di Napoli Federico II, Dipartimento di Agraria, Via Università 100, 80055 Portici (NA), Italy. [3]Departamento de Genetica, Facultad de Ciencias, Campus Rabanales 14071, Córdoba, Spain. [4]Dipartimento di Biologia Ambientale, Università la Sapienza, Via De Lollis 21, 00185 Roma, Italy. [5]Università degli Studi di Napoli Federico II, Dipartimento di Farmacia, Via Montesano 49, 80131 Napoli, Italy. E-mail: nadia.lombardi@ipsp.cnr.it*

Plant root systems release different bioactive compounds acting as chemical signals that may influence the activity, and subsequently affect the composition of the rhizosphere microbiome. In this work, we investigated the initial phase of the molecular cross-talk between tomato plants and a biocontrol strain of *Trichoderma harzianum* (T22). Root exudates (RE) were obtained from tomato grown in a split-root system, whereby the roots were exposed to different biotic or abiotic stresses. The collected RE were tested to determine their effect on germinating T22 spores, as observed in a microscope-guided assay. The RE produced by plants subjected to pathogen attack, salinity or wounding were found to act as chemoattractants to the developing *Trichoderma* spores. Conversely, most of the same RE treatments did not enhance the chemotropism of the fungal pathogen *Fusarium oxysporum* f. sp. *lycopersici*, suggesting a differential mechanism that may be selective for non-pathogenic versus pathogenic microbes interacting with the plant. Furthermore, specific molecules were identified as being systemically released by tomato roots during the initial phase of the stress response. Their involvement, as part of the chemical signal, was examined by applying the pure compounds alone or in presence of a specific inhibitor, to test the effect on the attraction of *Trichoderma* germ tubes. Moreover, we characterized the pattern of these metabolites secreted by the roots in the diverse stress conditions. This new information can contribute to a better understanding of how plants influence their microbiome and of microbe-plant interactions that produce positive effects on crops.

BIOLOGICAL CONTROL OF *PHELIPANCHE RAMOSA* IN TOMATO CROP IN SOUTHERN ITALY. F. Lops, M.L. Raimondo, G. Disciglio, E. Tarantino, F. Cibelli, A. Carlucci. *Università degli Studi di Foggia, SAFE, Via Napoli 25, 71122 Foggia, Italy. E-mail: antonia.carlucci@unifg.it.*

In Italy, *Phelipanche ramosa*, a root holoparasitic weed plant of many cultivations, is one of the most important problem of tomato cultivation, causing severe quality damage and reduced yields. The biological control of this parasitic weed includes the use of fungi and bacteria able to reduce the density of the parasite and crop damage by direct infections to parasitic weeds, and by improving the crop growth and deterring the parasitic infections. In the present work, arbuscular mycorrhizal fungi (*Rhizoglomus intraradices*), fungal pathogens as *Fusarium oxysporum*, and a trade microbial formulate were used to control and/or limit the infections of *Phelipanche ramosa* on tomato crop in open field. The experimental trial was carried out in Foggia province (Apulia region, southern Italy), during the spring-summer season 2016, in order to assess the effect of the three biological treatments alone and in combination, and included eight treatments. The setup of trial was performed with eight parcels of 500 m^2 (50 × 10 m), where tomato seedlings of cultivar 'Dres' were transplanted. At the end of tomato cultivation, the number of broomrape shoots was determined for each treatment. The data were subjected to statistical analyses. The results obtained showed that the use of mycorrihzas and *Fusarium* inoculum alone and combined to control/limit the weed parasitic were ineffective, while the microbial formulate alone and combined with mycorrhizas and/or *Fusarium* inoculum resulted to be more effective because it reduced significantly the broomrape shoot emergence.

POLYGLUCOSAMINE TREATMENT CAUSES DEEP TRANSCRIPTOME CHANGES IN *PSEUDOMONAS SYRINGAE* pv. *ACTINIDIAE,* AFFECTING IMPORTANT ASPECTS OF BACTERIAL PATHOGENICITY. A. Lovato[1], M. Petriccione[2], T. Colombo[3], N. Vitulo[1], A. Regaiolo[1], E. Vandelle[1], M. Scortichini[2], A. Polverari[1]. *[1]Dipartimento di Biotecnologie, Università degli Studi*

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Pseudomonas syringae pv. *actinidiae* (*Psa*), the agent of bacterial canker of kiwifruit, causes severe economic losses in all kiwifruit-growing areas of the world, the highly virulent biovar 3 in particular. Preventive measures rely on integrated management of orchards, use of protective plastic nets and sprays with copper or alternative treatments with biostimulants or biocontrol agents, for coexistence with the disease. Among the tested compounds, polyglucosamine (chitosan) showed a good effectiveness in reducing exudate emission and pathogen dispersal. These preparations show both antimicrobial and resistance inductions activities, but their mechanisms of action are not yet completely unravelled. In this work we analysed the effect of a polyglucosamine preparation (Hendophyt®) on *Psa* transcriptional profile, following treatment of bacteria at sublethal concentrations. *Psa* gene expression changes were analysed on a *Psa*-specific microarray resource, previously produced by our group, carrying a non-redundant collection of *Psa* whole genome sequences, representing all 3 main biovars. Results showed that down-regulated genes are strongly enriched in genes related to cell motility and flagella biosynthesis, while up-regulated genes are enriched in different functional categories, such as lipid metabolism or RNA processing and gene expression, accordingly with the huge number of up-regulated genes following treatment. These data suggest that polyglucosamine not only act by physically interacting with bacterial components but can deeply affect gene expression in crucial aspects of bacterial pathogenicity.

GENETIC VARIABILITY AND POPULATION STRUCTURE OF *PLASMOPARA VITICOLA* IN ITALY. G. Maddalena[1], P. Campia[1], G. De Lorenzis[1], O. Failla[1], P.A. Bianco[1], F. Delmotte[2], S.L. Toffolatti[1]. *[1]Università degli Studi di Milano, Dipartimento di Scienze Agrarie e Ambientali - Produzione, Territorio, Agroenergia (DISAA), via Celoria 2, 20133 Milano, Italy. [2]INRA, ISVV, UMR Santé et Agroécologie du Vignoble, Villenave d'Ornon, France. E-mail: silvia.toffolatti@unimi.it*

Plasmopara viticola (Berk. *et* Curt.) Berl. and De Toni is an obligate biotrophic oomycete which causes grapevine downy mildew, one of the most important grapevine disease, affecting particularly *Vitis vinifera*. The pathogen originated in Northern America and was for the first time reported in Europe in 1878 undergoing a bottleneck event, resulting in reduced genetic diversity across Europe. The genetic variability of *P. viticola* in Italy was investigated by genotyping, at 32 microsatellite markers, 106 *P. viticola* strains collected from 13 different regions. Each strain analysed presented a different multilocus genotype, suggesting that sexual reproduction is important. The lack of significant deviation from the Hardy–Weinberg equilibrium indicates that *P. viticola* populations are panmictic. The study also revealed a low genetic diversity and the lack of a genetic structure according to geographic origin, *V. vinifera* cultivars or disease management (organic *vs* IPM). The mixed reproductive system of *P. viticola* results in a great evolutionary potential and adaptability that could have a negative impact on the durability of resistance over a long term period. Therefore, all disease control methods available should be integrated in order to reduce the selection pressure towards pathogen strains able to overcome the plant defense system.

NATURAL BIOPOLYMERS AND *TRICHODERMA* CO-FORMULATIONS TO IMPROVE PLANT PRODUCTION. G. Manganiello[1], M. Napolitano[2], V. Guastaferro[2], D. Padrevita[2], S. Lanzuise[2], M. Gallo[2], D. Stellitano[1], M. Malinconico[3], P. Mormile[4], S. Bolletti Censi[5], M. Lorito[1,2], F. Vinale[1,2]. *[1]Consiglio Nazionale delle Ricerche- Istituto per la Protezione Sostenibile delle Piante (CNR-IPSP), Via Università 133, 80055 Portici (NA). [2]Università degli studi di Napoli Federico II - Dipartimento di Agraria, Via Università 100, 80055 Portici (NA). [3]Consiglio Nazionale delle Ricerche - Istituto dei Polimeri, Compositi e Biomateriali (IPCB), Via Campi Flegrei 34, 80078 Pozzuoli (NA). [4]Consiglio Nazionale delle Ricerche - Istituto di Scienze Applicate e Sistemi Intelligenti (ISASI), Via Campi Flegrei 34, 80078 Pozzuoli (NA). [5]Marea Scarl, Via Vittoria Colonna 14, 80121 Napoli. E-mail: francesco.vinale@ipsp.cnr.it.*

Trichoderma spp. have been widely studied and are presently marketed as biopesticides and biofertilizers. New formulations obtained by combining these microbes and innovative natural biopolymers are proposed in this work. Polysaccharide-based biopolymers are a class of interesting molecules because of their bioactivity, biodegradability and possible use in formulations as a carrier to immobilize, protect, retain and release inoculum of beneficial microbes such as *Trichoderma*. Further, the mixture of high and low molecular weight carbohydrates forms a biofilm that can be useful for agricultural purposes. We evaluated the effect of polysaccharide mixtures and selected *Trichoderma* strains on the growth and induction of resistance to disease in tomato plants. Applications of a 0.1% polysaccharide mixture and *Trichoderma* strains (10^6 spore/ml) were found to promote plant development. In particular, the co-formulation with the biopolymer plus *T. harzianum* strain M10 incremented root fresh weight by 44%, 9% and 37% in comparison to plants treated respectively with water, the polysaccharide mixture or M10 alone. Furthermore, stem length was greater with the combined treatment of the polysaccharide mixture plus *T. atroviride* strain P1, demonstrating a 12%, 16% and 27% increased growth in comparison to untreated plants, the polysaccharide mixture or P1 alone, respectively. Induction of disease resistance is currently being confirmed, with preliminary results indicating a significant effect of symptoms caused by *Botrytis cinerea* and *Rhizoctonia solani*. The co-formulation of biopolymers and plant beneficial microbes may permit the development of more effective and reliable biostimulants and plant protection products.

Work supported by MAREA project (MIUR PON03PE_00106_1).

HERBARIA AND REFERENCED FIELD STUDIES AS USEFUL TOOLS TO INTERPRET TRENDS OF PLANT DISEASES UNDER CLIMATE CHANGE. G. Marchi, C. Aglietti, P. Capretti, L. Ghelardini. *Università degli Studi di Firenze, DISPAA, Piazzale delle Cascine 28, 50144 Firenze, Italy. E-mail: paolo.capretti@unifi.it*

Epidemiology and phytopathological modeling generally use real data to predict the establishment, re-emergence, or disappearance of infectious diseases. Predictive models may be more or less reliable depending on available information and input data. In the case of Forest pathology, the data on the pathogen, the host and the environmental components are often scarce, partial or extremely variable. Only recently, data on genetic variability of pathogens and host populations and on climatic variables have become available, reliable and continuous. Considering the length of tree life cycles, the use of bibliographic data and herbarium samples, their analysis, implemented with molecular methods, might enable the reconstruction of epidemiological trends of serious infectious diseases, especially those due to introduced pathogens, and might help to evaluate the impact of climate on the spread of forest tree pathogens. This contribution reports about a data analysis of over 2230 samples of the Phytopathological Herbarium of the Royal Forest

Institute of Vallombrosa (Firenze). Samples from the Herbarium, assembled during the period 1880 to the 1940s, include many rusts (belonging to genera: *Chrysomyxa*, *Gymnosporangium*, *Melampsorella*, *Peridermium Puccinia*), powdery mildew (genera: *Microsphaera Sphaerotheca Uncinula*), agents foliage and shoot disease (*Gnomoniaceae*, *Rhytismaceae*, *Taphrinaceae*), decay and root fungi (*Agaricaceae*, *Polyporaceae*, *Xylariaceae*). To quantify the effect of plant diseases in a certain territory, Herbaria information needs to be supported by Bibliographic data. A combined analysis will help to reconstruct and interpret the phytopathological situation of northern Italy in the past hundred years and to attempt relating it to the changes in climatic conditions.

OUTBREAK OF *DOTHISTROMA SEPTOSPORUM* ON CORSICAN PINE IN SOUTHERN ITALY. G. Marchi, L. Ghelardini. *Università degli Studi di FIRENZE, DISPAA, Piazzale delle Cascine 28, 50144 Firenze. E-mail: luisa.ghelardini@unifi.it*

In spring 2017, serious and widespread crown damage and defoliation were observed in forests of Corsican pine (*Pinus nigra laricio*) in La Sila Massif, Italy. The needles of affected individuals showed yellowing and progressive dessication from the tip accompanied by typical red spots and bands surrounding spore-containing brown-black fruiting bodies. A fungus with morphological traits compatible with those of *Dothistroma* species was isolated by plating conidial suspensions onto MEA. Sequencing of the ITS gene region and of the elongation factor 1α gene confirmed the identification of isolates with *Dothistroma septosporum*, a listed quarantine pathogen in Europe, which is the causal agent of Red Band Needle Blight. Previously, the pathogen had been reported in southern Italy only once in 1970 on the exotic species *Pinus radiata* at a location about 100 km apart from the site of the current outbreak. Currently, the pathogen is spreading in natural woods of the native Corsican pine, which is the most common conifer species in the region and an extremely susceptible host to this pathogen. The concomitant presence of an abundant and especially susceptible host species and of conducive local climatic conditions, *i.e.* relatively frequent rainfall and high relative humidity also during late spring and summer months, and mild temperatures, may favor the occurrence of a severe disease outbreak on a larger area.

MORE THAN SCIENCE: A MULTI-TOOL APPROACH FOR RESEARCH COMMUNICATION IN PLANT PATHOLOGY AND PLANT BIOSECURITY. A. Masino[1], A. Bertin[2], M.L. Gullino[1]. *[1]Centro AGROINNOVA, Università degli Studi di Torino, Torino, Italy. [2]SPIN-TO srl, Via Roma 366, Torino (TO), Italy. E-mail: andrea.masino@unito.it*

The first instruments of communication for researchers are the scientific papers. Particularly young scientists need to publish on peer reviewed, international papers. This is the first form of communication, within peers. Moreover, in applied fields such as plant pathology, researchers need to communicate with technicians and extension services. The industrial partners could facilitate the translation of research findings into marketable products or services. Dissemination, communication, and life-long learning must be a task, mostly in the frame of European projects. This task could bring together enterprises, policy advisors, regional officers, farmers and associations, natural and social sciences and the general public to discuss the results obtained by researchers as well as cross-cutting issues related to plant health. Moreover, it could involve children and young students from the schools, with the aim to teach how scientists live and work. Researchers must learn to communicate also through innovative technologies, attending training courses, provided by specialists, on special topics related to the current need of new technologies (websites, newsletters and social media). Because research communication involves the whole society, it is fundamental that citizens better understand how research works to raise visibility of the global importance of plant health, sustainable agriculture, food security and environmental protection. Through new formats and technologies, phytopathologists can communicate the results of research, using a multi-tool approach, and learn more about stakeholders' perceptions of how to protect the EU agrifood sector from both alien and native pests and pathogens.

SPECIFIC DETECTION AND QUANTIFICATION OF *FUSARIUM FUJIKUROI* IN RICE PLANTS AND SEEDS BY DIFFERENT PCR ASSAYS. S. Matić[1], D. Spadaro[1,2], A. Garibaldi[1], M.L. Gullino[1,2]. *[1]Università degli Studi di Torino, Centro AGROINNOVA, Largo Braccini 2, 10095 Grugliasco (TO), Italy. [2]Università degli Studi di Torino, DISAFA, Largo Braccini 2, 10095 Grugliasco (TO), Italy. E-mail: slavica.matic@unito.it*

Bakanae disease, caused by *Fusarium fujikuroi*, is the seedborne disease diffused in the main rice cultivation areas throughout the world. The limited availability of suitable chemical seed-dressing products brought attention of the rice seed companies to Bakanae management. Several PCR techniques have been developed for a specific detection of *F. fujikuroi* in different rice tissues. Conventional PCR allowed the discrimination of *F. fujikuroi* from 20 other *Fusarium* species encountered on the rice. Optimized SYBR green real-time PCR assay kept the specificity of the conventional PCR, and was able to detect up to 10 pg μl^{-1} of DNA, but it was not sensitive enough to detect *F. fujikuroi* from rice seeds. Sensitivity of a real-time PCR was further ameliorated by development of a TaqMan real-time PCR assay, allowing the detection of about 28 fg of DNA, which corresponds approximately to one haploid genome of *F. fujikuroi*. This assay was sufficiently sensitive to detect around 10 *F. fujikuroi* cells per gram of the rice seeds, and it was able to reveal the pathogen in asymptomatic rice tissues in early phases of Bakanae development.

ANTIFUNGAL ACTIVITY OF AQUEOUS AND ETHANOL EXTRACTS OF TROPICAL PLANTS AGAINST TOMATO FUNGAL PATHOGENES. P.N. Mekam[1,2], S. Martini[2], D. Tagliazucchi[2], J. Nguefack[1], E. Stefani[2]. *[1]University of Yaoundé 1, Faculty of Science, Department of Biochemistry, PO. BOX 812, Yaoundé, Cameroon. [2]University of Modena and Reggio Emilia, Department of Life Sciences, Via Amendola 2, 42122-Reggio Emilia, Italy. E-mail: pascalnoel.mekam@unimore.it*

Several plant extracts may have a strong antifungal activity that can be exploited in the management of fungal diseases as an alternative to synthetic fungicides, the abuse of which may have consequences on the environment and health. Ethanol extracts (EE) and water extracts (WE) of three tropical plants, *Stachytarpheta cayennensis* (*Verbenaceae*), *Oxalis barrelieri* (*Oxalidaceae*) and *Euphorbia hirta* (*Euphorbiaceae*) were obtained and screened for their antifungal activity against three major phytopathogenic fungi of tomato: *Fusarium* sp., *Alternaria* sp. and *Colletotrichum* sp. The phytopathogenic fungi used across the experiments were isolated in Cameroon on severely affected tomatoes and their molecular identification and characterization is ongoing. The antifungal activity was checked *in vitro* on different media supplemented with different concentrations of the extracts. *O. barrelieri* EE, at the concentration of 25 mg/ml, inhibited the mycelium growth of *Fusarium* sp. by 26.8% and remarkably modified its morphology. WE and EE of the same *O. barrelieri* inhibited the mycelium growth of *Colletotrichum*

sp. by 43.7% and 50.3% respectively, but showed no inhibition on *Alternaria* sp. The phytochemical analysis of these plant extracts revealed that EE of *S. cayennensis* was the richest in polyphenols and flavonoids. EE from *O. barrelieri* was particularly rich in alkaloids. The inhibitory effects on the phytopathogenic fungi were possibly related to the amount of polyphenols and alkaloids obtained through the extraction. Field experiments are being conducted on tomato to confirm the action of such extracts *in vivo*. These findings may contribute to develop new biofungicides to protect tomato from some fungal pathogens.

EVALUATION OF AN ENDOPHYTIC SYMBIONT AS A PUTATIVE BIOCONTROL AGENT OF THE CoDiRO STRAIN OF *XYLELLA FASTIDIOSA*. **M. Morelli[1], G. D'Attoma[1,2], M. Saponari[1], P. Saldarelli[1].** *[1]CNR-Istituto per la Protezione Sostenibile delle Piante (IPSP), 70126 Bari, Italy. [2]Università degli Studi di Bari Aldo Moro, Dipartimento di Scienze del Suolo, della Pianta e degli Alimenti, 70126 Bari, Italy. E-mail: massimiliano.morelli@ipsp.cnr.it*

The quarantine bacterium *Xylella fastidiosa* (*Xf*) is responsible for diseases of a wide range of cultivated and wild plants. Few efforts have been made to investigate the potential use of endophytic symbionts on the disease phenotype of *Xf*-infected plants. The aim of our study was to evaluate if *Paraburkholderia phytofirmans* PsJN strain, a plant growth-promoting rhizobacterium, whose beneficial effects in the reduction of symptom severity caused by *Xf* in grapevine affected by Pierce's Disease have recently been proven, may play a role as biocontrol agent against *Xf* CoDiRO strain, the agent of a severe disease of olives in Apulia (southern Italy). Greenhouse trials are being conducted to test the ability of *P. phytofirmans* to colonise xylem vessels of olive, *Nicotiana benthamiana* and oleander, following inoculation of bacterial suspensions by needle puncture and root dipping. A conventional PCR assay for detection of *P. phytofirmans* movement in plants has been developed to be used in combination with plate isolation and a qPCR specific assay. Preliminary results showed that needle-inoculated bacterial cells were detectable in the leaf petioles of the three hosts, away from the inoculation site. Root dipping proved successful in infecting *in vitro*-cultured olive plantlets. Double-infection assays, currently underway, will prove if *P. phytofirmans* PsJN shows a beneficial interaction with *Xf* CoDiRO.

DRAFT GENOME OF A HIGH VIRULENT ITALIAN STRAIN OF *PSEUDOMONAS SYRINGAE* pv. *SYRINGAE* ISOLATED FROM EGGPLANT. **C. Moretti[1], P. Rallo[1], E. Caballo-Ponce[2], A. Pintado[2], C. Ramos[2], G. Carannante[3], V. Stravato[3], R. Buonaurio[1].** *[1]Università degli Studi di Perugia, Dipartimento di Scienze Agrarie, Alimentari e Ambientali, Borgo XX Giugno 74, 06121 Perugia (PG), Italy. [2]Área de Genética, Facultad de Ciencias, Universidad de Málaga, Instituto de Hortofruticultura Subtropical y Mediterránea "La Mayora" (IHSM-UMA-CSIC), Málaga, Spain. [3]GENISTA s.r.l., via San Vincenzo 13, 04022 Fondi (LT), Italy. E-mail: chiaraluce.moretti@unipg.it*

Two identical strains of *Pseudomonas syringae* pv. *syringae* (*Psy*) were isolated from the stem of eggplants (*Solanum melongena* L.) showing severe wilting symptoms. Since both strains showed a high virulence when inoculated in eggplant seedlings, several molecular and phenotypic analyses were carried out for a better characterization. A phylogenetic analysis of one of these isolates, *Psy* DAPP-PG 773, in comparison with 22 other *Psy* strains was carried out based on three housekeeping genes (*gapA*, *rpoA* and *recA*). The results revealed that *Psy* DAPP-PG 773 clustered with other *Psy* strains infecting herbaceous plants. To better understand the molecular basis of the virulence of *Psy* DAPP-PG773, its genome was sequenced on an Illumina MiSeq platform using indexed paired-end 250-nucleotide v2 chemistry. A total of 7.907.342 pairs of reads were obtained, representing approximately 182-fold coverage of the genome and comprising 320 contigs. The assumed genome size was 5.95 Mb and the G/C content was 59.4%. Annotation of the *Psy* DAPP-PG 773 draft genome sequence assigned a total of 4872 candidate protein coding genes. Furthermore, several phenotypic traits relevant for the epiphytic and pathogenic lifestyle of *Psy* were investigated, *i.e.* biofilm formation, indol-3-acetic acid and exopolysaccharides production, motility and production of quorum sensing signal molecules. In order to verify the colonization of this bacterium in the host plant, *Psy* DAPP-PG773 was transformed with the green fluorescent protein and inoculated in eggplants. Observations are in progress to monitor the distribution of the bacterium in the host.

IDENTIFICATION AND CHARACTERIZATION OF A NEGATIVE STRAND RNA VIRUS CLOSELY ASSOCIATED WITH CITRUS CONCAVE GUM DISEASE. **B. Navarro[1], M. Minutolo[2], A. De Stradis[1], F. Palmisano[3], D. Alioto[2], F. Di Serio[1].** *[1]Istituto per la Protezione Sostenibile delle Piante, Consiglio Nazionale delle Ricerche, Bari, Italy. [2]Dipartimento di Agraria, Università degli Studi di Napoli Federico II, Portici (NA), Italy. [3]Centro di Ricerca, Sperimentazione e Formazione in Agricoltura Basile Caramia, Locorotondo (BA), Italy. E-mail: beatriz.navarro@ipsp.cnr.it*

Concave gum (CG) is a virus-like disease of citrus first described in the early 1930s, whose aetiology has remained elusive. Here we report a novel negative strand RNA (nsRNA) virus identified in CG-affected citrus trees. A field survey confirmed the close association between the disease and the novel virus denoted Citrus concave gum-associated virus (CCGaV). CCGaV has a bipartipe genome composed of a negative-sense RNA-1, coding for the RNA-dependent RNA polymerase (RdRp), and an ambisense RNA-2 encoding the nucleocapside (N) and a putative movement protein (MP). RNA-2 contains an intergenic region involved in the regulation of the transcription of CCGaV mRNAs in both polarity strands. Electron microscopy coupled with immunolabelling of the viral CP showed that CCGaV has flexuous not enveloped particles. The need of creating a new genus for classifying this bipartite negative-stranded RNA virus will be discussed. Interestingly, phylogenetic links were detected between CCGaV and members of the genus *Phlebovirus*, which are arthropod-transmitted viruses infecting mammals, and some phlebo-like viruses exclusively infecting arthropods. Phylogenetic reconstructions also showed that, as for other nsRNA viruses, the candidate ancestor of CCGaV was an invertebrate-restricted virus, although the adaptation of the CCGaV ancestor to plant hosts may have taken place independently of that of the other known plant-infecting nsRNA viruses. The impact of specific detection methods developed in the course of this study on sanitation and certification programs of citrus will also be discussed.

ISSR MARKERS DETECT LOW GENETIC VARIATION AMONG *FUSARIUM CULMORUM* ISOLATES FROM TUNISIA. **S. Oufensou[1,2], V. Balmas[1], B. Scherm[1], D. Rau[1], M. Ben Attia[2], S. Gargouri[3], M. Pasquali[4], Q. Migheli[1].** *[1]Dipartimento di Agraria, Università degli Studi di Sassari, Via E. De Nicola 9, I - 07100 Sassari, Italy. [2]Laboratoire de Bio-surveillance de l'environnement, Faculté des Sciences de Bizerte, Route de Tunis, 7021 Zarzouna, Université de Carthage, Tunisia. [3]Laboratoire de Protection des Végétaux, Institut National de Recherche Agronomique de Tunis, Rue Hédi Karray, 2049 Ariana Tunisia. [4]DeFENS-Department of Food*

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Inter-simple sequence repeat technique using three primers (ISSR4, ISSR5 and ISSR6) was performed to investigate the genetic variability and population structure of *Fusarium culmorum* isolated from diseased durum wheat basal stems. A total of 114 isolates, representing six geographically distinct populations (Bizerte, Mateur, Beja, Bousalem, Kef and Siliana), was collected from four climatic regions in Northern Tunisia (Humid, Sub-humid, middle semi-arid and high semi-arid) representing more than 90% of the national wheat production areas. A total of 27 fragments were obtained with 8 polymorphic bands. Cluster analysis with UPGMA using genetic distances showed a high level of similarity between isolates. Analysis of molecular variance (AMOVA) did not reveal any trend with regard to geographic origin and confirmed that the most of the genetic variability was within populations (97.5% of the total variance) when the small proportion of variability (2.49%) distributed among populations was not even statistically significant ($P = 0.08$). To our knowledge, this study represents the first report on the use of ISSR markers to assess the genetic variability of *F. culmorum* populations in Tunisia. Based on these results, we can conclude that the *F. culmorum* isolates recovered from different regions in Tunisia might be part of a single population pool and that spore dispersal probably occurs over a wide geographic area. Quite obviously, these hypotheses need to be further explored with a far larger number of additional markers.

TRANSCRIPTIONAL ANALYSIS OF BARBERA AND NEBBIOLO GRAPEVINES RECOVERED FROM FLAVESCENCE DORÉE. D. Pacifico, M. Legovich, L. Galetto, C. Marzachì, S. Palmano. *Istituto per la Protezione Sostenibile delle Piante, CNR, (IPSP) Strada delle Cacce 73, 10135 Torino, Italy. E-mail: sabrina.palmano@ipsp.cnr.it*

Grapevine (*Vitis vinifera*) can be severely affected by phytoplasmas, phloem sieve-restricted *Mollicutes*, transmitted by sap-feeding insect vectors. Flavescence dorèe (FD) is a serious threat to vineyard survival in several grape-growing areas, and is subjected to quarantine restrictions in Europe. Following initial infection and symptom expression, the plants may recover from the disease in the following years. Recovery is therefore an interesting but still largely unexplored aspect of the grapevine-phytoplasma interaction. The biological bases of recovery were investigated in two Italian cultivars, Nebbiolo and Barbera, showing different sensitivity to phytoplasma infection and different recovery abilities. For each cultivar, leaves of healthy and FD–recovered plants were collected at two time-points during the vegetative season (July and August) in a productive vineyard located in Piemonte (northern Italy). The vineyard was monitored for several years to estimate FD incidence and recovery rate. A list of 18 grapevine genes related to hormone signaling, ossidative response and defence pathways, possibly involved in the recovery phenomenon, was identified on the basis of the available literature and preliminary studies. Variation in mRNA level of the target genes, was evaluated by quantitative Real Time-PCR and correlated to the season and to the phytosanitary status. Although the season had a deep impact on gene expression variation, few genes related with hydrogen peroxide production and jasmonic acid pathway could discriminate healthy from recovered samples of both cultivars, confirming a probable role for these metabolisms during FD-recovery.

INFLUENCE OF EXTRACELLULAR ENVIRONMENT AND MICROBIAL COMPETITION ON FUSARIC ACID PRODUCTION BY *FUSARIUM OXYSPORUM*. D. Palmieri, F. De Curtis, D. Vitullo, G. Lima. *Università degli studi del Molise, Dipartimento Agricoltura, Ambiente e Alimenti, Via De Sanctis snc, 86100 Campobasso, Italy. E-mail: davide.palmieri@studenti.unimol.it*

Fusaric acid is one of the most important secondary metabolites produced by pathogenic fungi belonging to the genus *Fusarium*. This molecule plays an important role in pathogenesis and its concentration in plant tissues is positively correlated with both fungal virulence and symptoms severity. It is known that fusaric acid can exert a toxic action on plants, rhizobacteria as well as on other fungal species. In this study, the effect of both some environmental variables and microbial competition on fusaric acid production by *F. oxysporum* f. sp. *lycopersici* (*Fol*) strain 4287 (race 2) were evaluated *in vitro*. In particular, the effect of i) nitrogen source, ii) iron content, iii) pH and iv) siderophores on fusaric acid production were evaluated. The production of the phytotoxin during the interaction of *Fol* with the biocontrol agent *Rahnella aquatilis* strain 36 (*Ra*) and its knockout mutant derivate Δ*gcd* was also investigated. Our study evidenced as fusaric acid production by *Fol* is consistently associated with the extracellular alkalinization, which in turn depends on the nitrogen source. The presence of both a chelating compound (EDTA or pyoverdine) as well as the pH7 buffered, limits the iron solubility and consequently increases the fusaric acid production. The interaction between *Fol* and *Ra* drastically reduces the fusaric acid production; conversely the interaction of *Fol* with the *Ra* Δ*gcd* slightly increases the fusaric acid production. The wild type bacterial strain actively counteracts the extracellular alkalinisation produced by fungal proliferation and consequently the fusaric acid production, while the Δ*gcd* mutant does not affect the pH and also the production of this metabolite. These results suggest that fusaric acid can act as a siderophore playing a critical role in the cation competition with microbiome and host plant.

MODELLING AND PREDICTING RISKS FROM MYCOTOXIN MIXTURES, FROM FIELD PRODUCTION TO TOXIC EFFECTS. R. Palumbo[1], C. Brera[2], K. Campbell[3], C. Dall'Asta[4], A. Gkrillas[4], I. Oswald[5], P. Toscano[6], A. Venancio[7], P. Battilani[1]. *[1] Università Cattolica del Sacro Cuore, Piacenza, Italy. [2] Istituto Superiore di Sanità, Rome, Italy. [3] The Queens University of Belfast, Belfast, Northern Ireland. [4] Università degli Studi di Parma, Parma, Italy. [5] Toxalim, (Research Centre in Food Toxicology), Université de Toulouse, INRA, ENVT, INP-Purpan, UPS, Toulouse, France. [6] CNR-IBIMET, Florence, Italy. [7] CEB, Universidade do Minho, Braga, Portugal. E-mail: paola.battilani@unicatt.it*

Mycotoxins are secondary metabolites of fungi with toxic effects on humans and animals that result in illnesses and economic losses. Their occurrence, often as a mixture, is common in commodities including cereals, nuts, fruits and products of animal origin. In vegetables, mycotoxins are commonly produced during crop growing, with possible post-harvest accumulation when the environment stays suitable for fungal activity. Thus, mycotoxins represent a challenge for human and animal health. Many efforts have been done to reduce consumer exposure, mainly developing rational guidelines to mitigate mycotoxin content in all the exposed products and setting legal limits for maximum admitted content in food and feed products. Nevertheless, methods for carrying out risk assessment for combined exposure to multiple mycotoxins have not been adequately implemented. The main objective of MYCHIF project is to develop an integrated innovative modelling methodology for the risk assessment of mycotoxin mixtures in food and feed. A flexible holistic approach will be applied to guarantee effective possible enlargement to other chemical compounds. Planned activities are organized in three work packages (WP): WP1 focuses on fungi and mycotoxin production in the crop production chain. All relevant

topics regarding fungi, host crops, environment and their interaction will be considered. Toxicity data for single and co-occurring mycotoxins will be covered in WP2. WP3 will use all available data (stored in databases) to generate the MYCHIF risk assessment model applied to 5 case studies including the whole maize chain (cropping system, food and feed, animal products, non-compliance, and future scenarios).

Work supported by EFSA - GA/EFSA/AFSCO/2016/01-02

EVOLUTIONARY ANALYSIS OF *TOMATO LEAF CURL NEW DELHI VIRUS* IN ITALY: GENE FLOW AND EPIDEMIOLOGY. **S. Panno[1], M. Luigi[2], E. Troiano[3], A.G. Caruso[1], L. Tomassoli[2], G. Parrella[3], S. Davino[1].** *[1]Università degli Studi di Palermo, Dipartimento Scienze Agrarie, Alimentari e Forestali, Viale delle Scienze Ed. 5, 90128 Palermo, Italy. [2]Consiglio per la ricerca in agricoltura e l'analisi dell'economia agraria, centro di ricerca difesa e certificazione, Via C.G. Bertero 22, 00156 Roma, Italy. [3]Istituto per la Protezione Sostenibile delle Piante (IPSP-CNR), S.S. di Portici, Via Università 133, 80055 Portici (Napoli), Italy. E-mail: salvatore.davino@unipa.it*

Tomato leaf curl New Delhi virus (ToLCNDV) is a bipartite begomovirus, that causes damage in cultivated plant species such as *Solanaceae* and *Cucurbitaceae*. In zucchini squash it causes severe leaf curling, yellow mosaic, swelling of veins of young leaves, shortening of internodes, interveinal yellowing, roughness of the skin of fruit. In October 2015 the first outbreak occurred on zucchini squashes in Sicily. During the years 2015-2016 monitoring activities were carried out in southern Italy. A total of 50 symptomatic samples were collected and total DNA was purified. PCR analysis to amplify full coat protein gene and Sanger sequencing of amplicons were performed in order to evaluate the genetic variability of the isolates. Phylogenetic analysis showed that the Italian ToLCNDV isolates split out into two groups. One cluster grouped all isolates from different areas of Sicily together with the reference isolates from Tunisia and Spain, and included a subgroup represented by Sardinian and some Lazio isolates. A second cluster included samples from Campania and Lazio. These results suggest that in Sicily at least two independent introductions of ToLCNDV occurred, the first in 2015 from southern Spain and the second in 2016 from Tunisia, Sardinia and Campania only one accession occurred, while in Lazio at least two accession occurred. Results also suggest the rapid spread of the virus in southern Italy probably due to the moving of nursery plants infested by the vector *Bemisia tabaci* from one area to another. The presence of ToLCNDV in the Mediterranean basin represents a developing threat for economically important cucurbit crops.

MULTITOXIN DETECTION IN "I.G.P." HORTICULTURAL CROPS. **A. Parroni[1], M. Scarpari[1], M. Reverberi[1], L. Mannina[2], C. Ingallina[2], S. Circi[2], A. Sobolev[2], C. Fanelli[1].** *[1]Department of Environmental Biology, University of Rome "Sapienza", Rome, Italy. [2]Dipartimento di Chimica e Tecnologie del Farmaco, Sapienza Università di Roma, P.le Aldo Moro 5, 00185 Rome, Italy. E-mail: alessia.parroni@uniroma1.it*

Fungi can contaminate plants, fruits and seeds in field and during the post-harvest process, causing several losses. Some pathogenic fungi (*Aspergillus*, *Penicillium* and *Fusarium* species) in suitable conditions can produce mycotoxins, secondary metabolites hazardous for human and animal health. It is to underline that mycotoxins are resistant to extreme environmental conditions, *e.g.* high temperature, for this reason it is possible to find them even in absence of living mycelia. In addition, the same plant can be contaminated by different mycotoxigenic fungi, therefore different mycotoxins can be found simultaneously in the same crop. Because of their dangerousness, the European Community stricktly controls mycotoxins level in foods and feeds to protect human and animal health. The search of mycotoxins in vegetables seems to be quite neglected and often, in some of them, is carried out with obsolete techniques. Taking all together, it is useful to have a rapid, precise, unequivocal and simultaneous method for the detection of mycotoxin panel starting from the same sample. Here we propose a multitoxin LC MS/MS technique starting from a single extraction using few sample grams. We analysed red pepper, celery, "favetta" strawberry and "torpedino" tomato, grown in Lazio under the indication of geographical tipicity "I.G.P.". The results showed that mycotoxins belong mainly to *Fusarium* toxins (e.g. deoxynivalenol, fumonisins and enniatins), however in strawberry and tomato also ochratoxin A and alternariol were detected. Nevertheless, in all these cases the levels of mycotoxins were under the limits imposed by the EC for these products.

TOXIGENIC AND NONTOXIGENIC *ASPERGILLUS FLAVUS* ISOLATES IN MAIZE FIELDS TREATED WITH THE BIOLOGICAL PRODUCT AF-X1: SOIL AND GRAIN COLONIZATION. **S. Pecchia[1], L. De Martino[1], R. Bosco[1], G. Puntoni[1], G. Ragaglini[2], C. Tozzini[2], G. Vannacci[1].** *[1]Università di Pisa, DISAAA-a, Via del Borghetto 80, 56124 Pisa, Italy. [2]Institute of Life Sciences, Scuola Superiore S. Anna, Via Santa Cecilia 3, 56127 Pisa, Italy. E-mail: susanna.pecchia@unipi.it*

Aflatoxins contamination of maize, used for both human and animal consumption, is a serious constraint for economical crop production. Different strategies have been developed to manage aflatoxins in crops and among them biological control has shown great promise. This strategy is based on the application of nontoxigenic strains in maize fields to competitively exclude naturally toxigenic strains in the same niche and compete for crop substrates. Field trials were conducted in two different Tuscany locations using maize hybrids of maturity class FAO 400 and 600. Geo-referenced plots (1 ha) were treated or not treated with the biological product AF-X1 (25 kg ha^{-1}). Data were collected from five sites of each plot along two corner-to-corner diagonals (X shaped). *Aspergillus* section *Flavi* populations were enumerated and isolated from soils using a modified AFPA medium and were plated on YES medium to assess aflatoxin production by the ammonia vapour method. One-hundred maize kernels from each experimental plot were surface sterilized and plated on PDA amended with streptomycin. All the *Aspergillus* spp. developed colonies were transferred on AFPA, CZ and YES media in order to determine toxigenic and nontoxigenic *A. flavus* isolates. Soil propagule density of toxigenic isolates was higher in untreated than in AF-X1 treated plots. *A. flavus* was isolated from about 80% of all kernels tested, regardless of treatment. Greater than 99% of *A. flavus* isolates recovered from treated plots were nontoxigenic and significant values of toxigenic isolates were observed in untreated plots.

***ASPERGILLUS* SECTION *FLAVI* FROM CHESTNUTS: BIOLOGICAL, MOLECULAR AND CHEMICAL CHARACTERIZATION.** **S. Prencipe[1,2], I. Siciliano[2], A. Garibaldi[2], M.L. Gullino[1,2], D. Spadaro[1,2].** *[1]DISAFA - Dipartimento di Scienze Agrarie, Forestali ed Alimentari, Università degli Studi di Torino, Grugliasco (TO), Italy. [2]Centro di competenza per l'Innovazione in campo agro-ambientale (AGROINNOVA), Università degli Studi di Torino, Grugliasco (TO), Italy. E-mail: davide.spadaro@unito.it*

Aspergillus section *Flavi*, and particularly *Aspergillus flavus*, are considered the main responsible for aflatoxin contamination along

the chestnut production chain. During 2015, an extensive sampling of *Aspergillus* section *Flavi* from the field and throughout the processing phases of the chestnut flour production chain was carried out. Biological, molecular, and chemical assays were performed in order to characterize fifty-eight isolates. β-tubulin and calmodulin gene sequences were used to identify the isolates. The main species was *A. flavus*, followed by *A. oryzae* var. *effusus*, *A. tamarii*, *A. parasiticus* and *A. toxicarius*. By considering aflatoxins (AFs) production, 19% of the strains produced AFs *in vitro* and 40% *in vivo*. 56 out of 58 strains resulted virulent based on results from the pathogenicity assay. The *A. flavus* strains showed an intraspecific variability by molecular, morphological, chemical and biological analyses, confirming that a polyphasic approach is necessary to discriminate the species inside the section *Flavi*. The ability of the strains to produce AFs *in vivo* and the pathogenicity tests showed the potential contamination along the chestnut production chain. Guidelines to manage the aflatoxin risk should be developed for the chestnut chain, as common practices are not able to efficiently reduce the risk of contamination by aflatoxigenic fungi. This research represents the first characterization of aflatoxigenic fungi from fresh chestnut and the chestnut flour process.

CRITICAL FEATURES OF THE DIAGNOSIS OF *CLAVIBACTER MICHIGANENSIS* subsp. *MICHIGANENSIS* FROM TOMATO SEED. N. Pucci[1], V. Catara[2], M. Scortichini[3], E. Stefani[4], G. Perez[1], S. Loreti[1]. *[1]Consiglio per la Ricerca in Agricoltura e l'Analisi dell'Economia Agraria-Centro di Ricerca Difesa e Certificazione. [2]Dipartimento di Scienze delle produzioni Agrarie e Alimentari-Università degli Studi di Catania, Catania. [3]Consiglio per la Ricerca in Agricoltura e l'Analisi dell'Economia Agraria-Centro di Ricerca Olivicoltura, Frutticoltura e Agrumicoltura. [4]Dipartimento di Scienze della Vita-Università di Modena e Reggio Emilia, Modena. E-mail: stefania.loreti@crea.gov.it*

Bacterial canker, caused by *Clavibacter michiganensis* subsp. *michiganensis* (*Cmm*) is one of the most important bacterial disease of tomato (*Solanum lycopersicum*) worldwide. Seeds are the main pathway for the transmission of the bacterium, listed as an A2 quarantine pest, consequently the reliability of seed detection tests represents a critical point to prevent the introduction and spread of the pathogen. In this study some conventional (isolation and immunofluorescence) and molecular (end-point PCR) methods for the detection of *Cmm* from tomato seed samples were compared by an inter-laboratory comparison (ITC). Several Italian laboratories, belonging to different institutions, analysed the same panel consisting of 11 *Cmm*-spiked tomato seed samples for the evaluated methods. The obtained results showed that end-point PCR gave acceptable performance values, even if the choice of enzyme in the PCR reaction was crucial. Currently the EPPO protocol (Standard PM7/42 2) does not contemplate a preliminary screening phase based on molecular methods, but suggests two parallel flow diagrams that use, as first step, the isolation and immunofluorescence, respectively. Since these latter techniques showed low values of performance criteria, the study highlighted the necessity to integrate the EPPO protocol with a preliminary molecular screening test. It is advisable to validate new molecular methods more specific and sensitive than end-point PCR, based on systems such as real time PCR, LAMP and digital PCR.

EFFICACY OF COMPOST IN THE CONTROL OF *PSEUDOMONAS SYRINGAE* pv. *ACTINIDIAE* ON POTTED KIWIFRUIT PLANTS. M. Pugliese[1,2,3], M. Monchiero[4], M.L. Gullino[1,2,3], A. Garibaldi[1,3]. *[1]Università degli Studi di Torino, Centro AGROINNOVA, Largo Braccini 2, 10095 Grugliasco (TO), Italy. [2]Università degli Studi di Torino,* DISAFA*, Largo Braccini 2, 10095 Grugliasco (TO), Italy. [3]AgriNewTech srl, Via G. Quarello 15/A, 10135 Torino, Italy. [4]ANT-NET srl, Via G. Quarello 15/A, 10135 Torino, Italy. E-mail: massimo.pugliese@unito.it*

The recent outbreak of bacterial canker on kiwifruit, caused by *Pseudomonas syringae* pv. *actinidiae*, caused considerable damage to the international kiwifruit industry. The objective of the present work was to evaluate a preventive strategy, using disease suppressive compost to control this pathogen. Four trials were performed in glasshouse in two subsequent years, 2012 and 2013. Each treatment was carried out on 10 potted female plants of *Actinidia deliciosa* cv. *Hayward*, 30-40 cm high, and inoculated with strains of *Pseudomonas syringae* pv. *actinidiae* by directly spraying cell suspensions (10^8 cells ml^{-1}) onto the leaves. A set of plants were grown in black plastic pots containing 5 l of a sterilized peat mixture substrate, and part of them were weekly treated with copper hydroxide at 30 g/hl and used as controls. In the case of compost treatments, kiwifruit plants were planted in a mixture of steamed peat mixture substrate (80%, v/v), and compost (20%), but no other treatment was performed. A green compost (6 month maturation) and an urban biowaste compost (4 month maturation) where used. Surveys were carried out every 14 days. A partial but significant symptom reduction occurred when the plants were grown in the peat-compost mixture substrate, showing significantly fewer leaf spots compared with untreated controls and a disease control level like copper hydroxide. In the framework of integrated control strategies, compost could be used starting from nursery and in combination with copper, to develop new strategies to reduce the disease development and spread.

***FUSARIUM GRAMINEARUM* HYDROPHOBINS: ROLE IN FUNGAL GROWTH AND PLANT INFECTION. A. Quarantin[1,2], B. Hadeler[2], E.B. Monaghan[2], L. Sella[1], W. Schäfer[2], A.L. Martínez-Rocha[2].** *[1]University of Padova, Department of Land, Environment, Agriculture and Forestry, Viale dell'Università 16, 35020, Legnaro (PD), Italy. [2]Biocenter Klein Flottbek, Molecular Phytopathology and Genetics, University of Hamburg, Hamburg (Germany). E-mail: alessandra.quarantin@gmail.com.*

Hydrophobins are small fungal-specific proteins which are present at the surface of aerial hyphae and spores. They may play a role in several processes such as formation of fungal aerial structures, attachment to hydrophobic surfaces, interaction with the environment and protection against the host defense system by masking the fungal cell wall. The plant pathogen *Fusarium graminearum*, a necrotrophic fungus which causes *Fusarium* head blight (FHB) of wheat, barley and other cereal grains, contains five hydrophobin genes. To determine their role in *F. graminearum*, single and triple mutants of the five genes were produced and characterized. Interestingly, *Hyd1* is dramatically upregulated during initial epiphytical growth on wheat paleas and glumes. A reduction in hydrophobicity was observed for the single mutants *Δhyd1* and *Dhyd3* as well as the triple mutants including *Δhyd1* or *Dhyd3*. Single disruptants *Dhyd2*, *Dhyd4* or *Dhyd5* behaved wild type-like. The single mutant *Δhydr3* and the triple mutants including *Δhydr3* showed a decrease in growth compared to the wild type in a complete or minimal growth medium. Surprisingly, a reduced growth was registered when these mutants were grown under osmotic stress conditions or in the presence of H_2O_2. In order to verify possible defects in their cell wall, the growth of the mutants is currently under scrutiny in the presence of β-1,3-glucanase, chitinase and two fungicides. Point inoculation of wheat spikes with all mutants showed no role of the hydrophobins during infection. Preliminary data suggest a reduction in virulence of the *Hyd1* mutant after spray inoculation.

LAMP-BASED DETECTION OF *XANTHOMONAS CAMPESTRIS* pv. *CAMPESTRIS* IN BRASSICA PLANTS AND SEEDS. **G.R. Quintero Macías[1], G. Stampone[1], S. Panno[2], S. Davino[2], S. Drago[1], V. Catara[3], P. Bella[2].** *[1]Enbiotech S.r.l., Via Aquileia 34, 90144 Palermo, Italy. [2]Dipartimento di Scienze Agrarie, Alimentari e Forestali, Università degli Studi di Palermo, Viale delle Scienze Ed. 5, 90128 Palermo, Italy. [3]Dipartimento di Agricoltura, Alimentazione e Ambiente, Università degli Studi di Catania, Via S. Sofia 100, 95123 Catania, Italy. E-mail: patrizia.bella@unipa.it*

Xanthomonas campestris pv. *campestris* (*Xcc*) is the causal agent of black rot, a severe seed-borne disease of *Brassicaceae*. Other two closely related pathovars, *X. campestris* pv. *raphani* and *X. campestris* pv. *incanae* are responsible for a leaf spot and bacterial blight disease on brassica species. In this study, a new LAMP protocol was developed for the identification and detection of *X. campestris* pv. *campestris* and related pathovars based on the ICGENE technology (Enbiotech, Palermo) characterized by ready to use and temperature stable reagents and the portable ICGENE mini device. A set of six primers were designed on the *ndvB* gene which encodes a glycosyltransferase required for cyclic glucan synthesis and involved in the virulence of *Xcc*. Positive amplifications were obtained from a collection of *Xanthomonas campestris* pathovars isolated from different brassica species in different geographical location and including strains of different *Xcc* races. No target DNA amplification was obtained from other xanthomonads. A rapid DNA extraction protocol at 65°C for 10 min coupled with the LAMP assay in the ICGENE mini device allowed to detect the bacterium from artificially inoculated plant tissues and spiked seed extracts in about 40 min. The detection limit of the LAMP assay was the same both for pure bacteria culture and spiked seed extracts indicating that the assay is not affected by plant inhibitors. The ICGENE technology represents a rapid, sensitive and cost-effective tool for detecting *Xcc* by LAMP from cultures and from plants. In addition, results suggest its application in the seed testing protocol.

PATHOGENICITY ASSESSMENT OF DIFFERENT *PLECTOSPHAERELLA* SPECIES ON BASIL, PEPPER AND TOMATO CROPS. **M.L. Raimondo, A. Carlucci.** *Università degli Studi di Foggia SAFE, Via Napoli 25, 71122 Foggia, Italy. E-mail: antonia.carlucci@unifg.it*

Plectosphaerella species have been isolated in many countries from different hosts such as tomato, sunflower, soybean, melon, pumpkin and other cucurbits, endive and rocket, and Lamb lettuce. The most common and known species of *Plectosphaerella* is *Pa. cucumerina*, which was reported as a pathogen and endophyte from different horticultural crops as well as a biological agent to control of *Galium spurium*, *Sagittaria trifolia* and nematodes of potato. To date *Plectosphaerella* genus consists of 11 species such as *Pa. alismatis*, *Pa. citrulli*, *Pa. cucumerina*, *Pa. delsorboi*, *Pa. melonis*, *Pa. oligotrophica*, *Pa. oratosquillae*, *Pa. pauciseptata*, *Pa. plurivora*, *Pa. populi* and *Pa. ramiseptata*. To ascertain the role that these fungi play in diseases of horticultural crops, nine *Plectosphaerella* species were artificially inoculated on three different hosts (basil, pepper and tomato) to perform pathogenicity tests *in vitro* (detached lives) and *in vivo* (young 30-day-old plants). These tests were carried out in a greenhouse with an experimental design consisting of two independent batches. Each host per isolate combination was replicated five times. The individual disease severity was assessed 15-30 days post inoculation on leaves, roots and collars showing symptoms. Pathogenicity tests demonstrated that except for *Pa. oratosquillae*, all *Plectosphaerella* species tested are able to cause symptoms on all hosts essayed with different levels of disease severity. *Pa. paucisepatata* and *Pa. plurivora* showed a vascular behaviour while the others seven species had a parenchymatous behaviour. *Pa. ramiseptata* proved to be the most pathogenic species to the three hosts.

ACTIVITY OF POLYSACCHARIDES EXTRACTED FROM *ECKLONIA* sp. AND *JANIA* sp. AGAINST *BOTRYTIS CINEREA*. **H. Righini[1], E. Baraldi[1], A. Martel-Quintana[2], Y. García-Fernández[2], C. Pérez-Reyes[2], R. Roberti[1].** *[1]Dipartimento di Scienze Agrarie, Alma Mater Studiorum, Università di Bologna. [2]BEA-Banco Español de Algas, Universidad de Las Palmas de Gran Canaria (ULPGC), Las Palmas, Canary Islands, Spain. E-mail: roberta.roberti@unibo.it*

Algae extracts contain several bioactive compounds, such as polysaccharides, well known to be elicitors of plant defence responses, but few information are available on their antifungal activity. The aim of this research was to investigate the effect of cationic polysaccharides extracted from two macroalgae, *Ecklonia* sp. (*Ochrophyta*) and *Jania* sp. (*Rhodophyta*) against *Botrytis cinerea in vitro* and *in vivo* on strawberry. For the *in vitro* assay, fungal colony portions were treated by immersion in polysaccharides aqueous concentrations, 1.65, 0.82 and 0.41 mg/ml for *Ecklonia* sp. and 0.18, 0.09 and 0.045 mg/ml for *Jania* sp. After 6 h of incubation, the colony growth was measured daily. *Ecklonia* sp. polysaccharides significantly inhibited *B. cinerea* growth by 21% (1.65 mg/ml) and 23% (0.82 mg/ml) two days after treatment. *Jania* sp. polysaccharides did not inhibit fungal growth. For the biological assay, strawberry ripe fruits, cv. Cristal, were treated before or after harvesting, by immersion in polysaccharide aqueous solutions of the two algae (0.82 and 0.41 mg/ml for *Ecklonia* sp. and 0.09 and 0.045 mg/ml for *Jania* sp.). *Botrytis cinerea* was inoculated by spraying fruits with spore suspension (1×10^5 spore/ml) 24 h after treatment. Disease symptoms were scored as percentage of infected area. Pre-harvest treatment with *Jania* sp. reduced disease symptoms by 100% at 0.09 mg/ml and of 50% at 0.045 mg/ml and with *Ecklonia* sp. by 17% (0.82 mg/ml) and 11% (0.41 mg/ml). No inhibition of disease symptoms was obtained in post-harvest treatment.

ELUCIDATION OF THE MECHANISM OF ACTION OF ESSENTIAL OILS TO CONTROL POSTHARVEST DISEASES OF APPLES AND PEACHES. **K. Santoro[1,2], D. Spadaro[1,2], A. Garibaldi[2], M.L. Gullino[1,2].** *[1]DISAFA - Dipartimento di Scienze Agrarie, Forestali ed Alimentari, Università degli Studi di Torino, Grugliasco (TO), Italy. [2]Centro di competenza per l'Innovazione in campo agro-ambientale (AGROINNOVA), Università degli Studi di Torino, Grugliasco (TO), Italy. E-mail: davide.spadaro@unito.it*

Essential oils are considered a powerful and natural resource to control postharvest pathogens of pome and stone fruits. The efficacy of these natural products has been deeply investigated *in vitro* but only few of them are applied *in vivo*. Essential oils can be applied in different ways, by dipping or spraying the fruit surface. Their high volatility permits the application also by fumigation, which is preferable because of the lack of contact with the fruit. Thyme and savory essential oils were successfully applied through biofumigation at 0.5% and 0.1% against brown rots on nectarines and peaches. The most effective components of thyme and savory essential oils are thymol and carvacrol, respectively. The antimicrobial activity of essential oils, useful to control fungal pathogens, could be due to a synergy of chemical components. In addition to direct inhibition of pathogen growth, essential oils can induce resistance in the fruit host. Thyme essential oil can promote the expression of the pathogenesis related gene PR-8 in apple, which is involved in host defense response. Moreover, essential oils showed a positive role in slowing down senescence processes reducing

weight loss and preserving vitamin C and carotenoid content during storage.

MICROCANTILEVER RESONATORS FOR OCHRATOXIN A DETECTION IN FOOD SAMPLES. **K. Santoro[1,3], D. Spadaro[1,2], M.L. Gullino[1,2], C. Ricciardi[3].** *[1]Università degli Studi di Torino, DISAFA, Largo Braccini 2, 10095 Grugliasco (TO), Italy. [2]Università degli Studi di Torino, Centro AGROINNOVA, Largo Braccini 2, 10095 Grugliasco (TO), Italy. [3]Politecnico di Torino, DISAT, Corso Duca degli Abruzzi 24, 10129 Torino (TO), Italy. E-mail: davide.spadaro@unito.it*

An innovative and rapid detection method based on microcantilever resonators for ochratoxin A (OTA) detection in food matrix was developed. The harmful effects of OTA on human and animal health lead to develop and optimize highly sensitive, fast and accurate methods for OTA detection. Ochratoxin A can contaminate a wide number of foodstuffs during postharvest representing a serious threat to human health. Microcantilever resonator arrays could effectively identify OTA at low concentrations (less than 6 ng/ml), with relatively low uncertainty (about 10%) and good reproducibility for the same target concentration. Furthermore, the developed immunosensing method showed limited cross-reactivity to different mycotoxins, paving the way to a highly specific technique, able to identify different mycotoxins in the sample. The microcantilever technology was tested in different food matrices, to detect OTA in grape juice, green coffee and red wine with high sensitivity and reproducibility. This work demonstrates the possibility to apply microcantilever technology in food safety field, developing an innovative biosensing platform able to detect OTA with high sensitivity and reproducibility.

IMPROVEMENT OF QUADRUPLEX TAQMAN REAL TIME METHOD TO SCREEN THE PRESENCE OF *XANTHOMONAS VESICATORIA*, *X. EUVESICATORIA*, *X. PERFORANS*, *X. GARDNERI* IN TOMATO SEEDS. **A. L'Aurora, V. Scala, N. Pucci, S. Loreti.** *Council for Agricultural Research and Economics, Research Center for Plant Protection and Certification, Roma, Italy. E-mail: stefania.loreti@crea.gov.it*

Bacterial spot of tomato, a major problem in many tomato production areas, is caused by *Xanthomonas vesicatoria*, *X. euvesicatoria*, *X. perforans* and *X. gardneri*. In the frame of the project ASPROPI financed by the Italian Ministry of Agricolture and Forestry, we investigated the possibility to validate a new protocol for the preliminary screening of *Xanthomonas vesicatoria*, *X. euvesicatoria*, *X. perforans* and *X. gardneri* in tomato seeds. In order to detect the bacterial spot pathogens, the region of *hrpB* operon was evaluated as target for a quadruplex real-time polymerase chain reaction (PCR). The PCR products are highly conserved within each species, with a single-nucleotide polymorphism (SNP) among bacterial spot of tomato agents. Four probes and two primers were employed to detect the four bacterial spot pathogens simultaneously. The optimized quadruplex assay was assessed for analytical specificity and sensitivity showing good performance criteria. The new protocol was validated within a test performance study (TPS) and compared with the already available diagnostic methods. Seven different laboratories of the Plant Protection Services participated to the TPS to verify the reproducibility of the tested method. The obtained results showed that this method holds great potential as a diagnostic tool for the detection of each bacterial spot pathogen from seed tomato matrix, and also for the identification of *Xanthomonas*-like pure cultures.

NEW CLASS OF LIPID COMPOUNDS IN *XYLELLA FASTIDIOSA* STRAIN CoDiRO. **V. Scala[1], N. Pucci[1], S. Lucchesi[1], A. L'Aurora[1], M. Ludovici[2], M. Reverberi[3], S. Loreti[1].** *[1]Council for Agricultural Research and Economics, Research Center for Plant Protection and Certification, Roma, Italy. [2]Laboratory of Cutaneous Physiopathology and Integrated Center of Metabolomics, San Gallicano Dermatologic Institute (IRCCS), Rome, Italy. [3]Department of Environmental biology, Sapienza University of Rome, Italy. E-mail: valeria.scala@crea.gov.it*

Modulating signals involved in plant-pathogen interaction represent a powerful mean to develop innovative and sustainable approaches to control plant pathogens. We investigated some of these signals, *i.e.* oxylipins in *Xylella fastidiosa* CoDiRO strain (variant "sequence type 53"), associated with the olive quick decline syndrome. During plant-pathogen interactions, lipids have different roles, as pathogen perception, signal transduction and downstream defence responses. The composition of the bacterial membrane is not constant but depends on the environmental conditions to which the cells are exposed. Oxidized fatty acids are an important class of signalling molecule especially related to stress responses. Recently, other authors reported that oxylipins have a regulation activity in motility, biofilm formation and virulence of *Pseudomonas aeruginosa*. In the frame of the Xf-actors project we explored the oxylipin signals of *X. fastidiosa* subsp. *pauca* CoDiRO strain CFBP8402 in pure culture and during the interaction with the model plant *Nicotiana tabacum* "Petite Havana SR1". The analyses were performed by LC-MS/MS in dynamic MRM modality allowing quantification of oleic, linoleic and linolenic acid-derived oxylipins. The results showed the presence of oxidized fatty acids in *X. fastidiosa* CoDiRO strain in pure culture and in inoculated tobacco plants.

DEEP SEQUENCING OF TWO *CITRUS TRISTEZA VIRUS* ISOLATES CROSS PROTECTIVE AGAINST HOMOLOGOUS SEEDLING YELLOWS-VT STRAIN. **G. Scuderi[1], M. Russo[1], R. Ferraro[2], M.C. Bazzano[1], O.F. Giarrusso[1], A. Catara[2], G. Licciardello[1].** *[1]Agrobiotech, ZI Blocco Palma I, Via V. Lancia 57, 95121 Catania, Italy. [2]Parco Scientifico e Tecnologico della Sicilia, ZI Blocco Palma I, Via V. Lancia 57, 95121 Catania, Italy. E-mail: glicciardello@agrobiotech.it*

A wide indexing and genotyping of VT-like *Citrus tristeza virus* (CTV) in Sicily has identified some isolates that are potentially cross-protective (CP) on sour orange against the prevalent seedling yellows (SY) isolate SG29, but retain the stem pitting (SP) phenotype on grapefruit. As a result of bioindexing they have a biotype 10 instead of severe SY associated to biotype 4. Deep sequencing of two candidate CP isolates and alignment of their full genomes against the challenger SG29 (KC748392) showed 13 and 14 point mutations, respectively, including 5 and 6 silent mutations. No evidence of recombination and/or additional strains was present. Interestingly, eight changes were shared in the same position and three of them were located within *p33* (positions 11490, 11585, 11756 nt), and one in *p23* (18508 nt). The two isolates differed by nine nucleotides, three within *orf1A* (position 486, 3208, 4780 nt), one in *p33* (positions 11721 nt), three in the intergenic region *p33-p6* (11790, 11791, 11792 nt), one in *p25* (position 16316 nt) and one in *p18* (position 16970). While the precise genomic events that have led to the mutation changes remain to be established, the results show that: (i) the "superinfection exclusion" conditions for cross protection predicted for T36 strains attain also to VT strain; (ii) it may occur "naturally" in the field, eventually from recovered plants and aphid spread; (iii) the genetic determinants of SP phenotype are different from those of SY; (iv) the search for cross protective mutants needs a mandatory genotyping and a phenotype analysis on specific indicators.

CHARACTERIZATION OF THE ROLE PLAYED BY *MAGNAPORTHE ORYZAE* POLYSACCHARIDE MONOOXYGENASES AND RELATED ENZYMES DURING INFECTION OF RICE. L. Sella[1], A. Quarantin[1], F. Favaron[1], L.T. Đô[2], V.V. Van[3], N.M. Hung[2]. *[1]University of Padova, Department of Land, Environment, Agriculture and Forestry, Viale dell'Università 16, 35020, Legnaro (PD), Italy. [2]Duy Tan University, K7/25 Quang Trung, Da Nang, Vietnam. [3]Nguyen Tat Thanh University, 298-300A Nguyen Tat Thanh Street, District 4, Ho Chi Minh City, Vietnam. E-mail: luca.sella@unipd.it; lethdo@hotmail.com*

In 2030, the global rice production is expected to increase to meet the demand of the growing world population. However, rice is severely affected by the blast disease caused by the fungus *Magnaporthe oryzae*, which can reduce by 10-30% the total annual rice production. In the early stages of the infection process, *M. oryzae* forms an appressorium to assist its penetration into plant tissue and expresses many polysaccharide and lignin-degrading enzymes. Among these, polysaccharide monooxygenases (PMOs) degrade their substrates by an oxidative mechanism and could be important virulence factors for the fungus. The first objective of the project, which is part of the Scientific and Technological Cooperation Agreement between the Italian Ministry of Foreign Affairs and International Cooperation and the Department of International Cooperation of the Ministry of Science and Technology of Vietnam, is to identify the role played by *M. oryzae* PMOs and related enzymes during pathogenesis, with the final aim to develop new methods to control rice blast disease. Candidate *M. oryzae* genes encoding PMOs and related enzymes have been identified by an *in silico* analysis of the fungal genome and their expression during the infection process, and particularly during appressorium formation, has been characterized by transcriptomic analysis. Knock-out mutants of the most expressed genes will be generated and their virulence evaluated on rice plants. The role played by the target enzymes on appressorium formation will also be evaluated.

EVALUATION OF BARLEY ENTRIES DEVELOPED FOR ORGANIC OR LOW INPUT AGRICULTURE FOR THEIR SUSCEPTIBILITY TO *FUSARIUM* HEAD BLIGHT INFECTIONS AND MYCOTOXIN ACCUMULATION. M.T. Senatore[1], G. Beccari[1], F. Coccia[1], L. Raggi[1], U. Bonciarelli[1], F. Tini[1], M. Sulyok[2], M. Guiducci[1], V. Negri[1], L. Covarelli[1]. *[1]Department of Agricultural, Food and Environmental Sciences, University of Perugia, Borgo XX Giugno 74, 06121 Perugia, Italy. [2]Department of Agrobiotechnology (IFA-Tulln), University of Natural Resources and Life Sciences, Vienna, Konrad Lorenzstr. 20, 3430 Tulln, Austria. E-mail: lorenzo.covarelli@unipg.it*

The aim of the present study was to evaluate the response to *Fusarium* Head Blight (FHB) infections and mycotoxin accumulation of nine different barley entries: i) five pure lines (namely SOL 1, SOL 2, SOL 7, SOL 30 and SOL 57, respectively), ii) one Composite Cross Population (CCP) (namely AUT DBA) and iii) one line mixture (namely *mix48*) all developed for organic or low input agriculture using an Evolutionary Breeding approach, and iv) two commercial varieties used as controls (Quench, Rattan). The evaluation was conducted in an experimental field plot trial under both artificial inoculation, with a mixture of four *Fusarium* species (*F. graminearum*, *F. culmorum*, *F. avenaceum* and *F. poae*), and natural inoculum conditions. FHB was evaluated by isolation and identification of the different *Fusarium* species infecting the grains, fungal biomass quantification of the four artificially inoculated *Fusarium* species by q-PCR and fungal secondary metabolites quantification in the grains by LC-MS/MS. Under natural infection conditions the fungal complex associated with FHB was predominantly composed of *F. tricinctum*, while *F. culmorum* was the most isolated species under artificial inoculation conditions. Quantitative PCR and LC-MS/MS analyses highlighted differences between the tested barley entries. For example, the lowest levels were detected in the commercial varieties Quench and Rattan, probably because of their late cycle with respect to the others. However, some lines such as SOL 57, SOL 1 and the mixture *mix48* proved to be in some instances not significantly different from Quench variety.

EFFICACY OF *BACILLUS* spp. IN THE CONTROL OF *ASPERGILLUS PARASITICUS* AND AFLATOXINS ON PISTACHIO. F. Siahmoshteh[1], I. Siciliano[2], M. Razzaghi-Abyaneh[3], M.L. Gullino[2,4], D. Spadaro[2,4]. *[1]Department of Food Science and Technology, Faculty of Agriculture, Tarbiat Modares University, Tehran, Iran. [2]DISAFA - Dipartimento di Scienze Agrarie, Forestali ed Alimentari, Università degli Studi di Torino, Grugliasco (TO), Italy. [3]Department of Mycology, Pasteur Institute of Iran, Tehran 13164, Iran. [4]Centro di competenza per l'Innovazione in campo agro-ambientale (AGROINNOVA), Università degli Studi di Torino, Grugliasco (TO), Italy. E-mail: davide.spadaro@unito.it*

Pistachio (*Pistacia vera*) has important economic, nutritional and health aspects but it can be contaminated by aflatoxigenic fungi in the field and during storage. Biological control could be considered as an alternative to chemical treatment. Two *Bacillus* spp. were tested *in vitro*, and both strains were able to reduce the mycelial growth and they were able to degrade aflatoxins (AFB1, AFb2, AFG1, and AFG2) during the first three days after inoculation. The cultivar of pistachio 'Ahmad-Aghaei' was the most susceptible to fungal colonization among the four main Iranian cultivars, and was used in this study. *Aspergillus parasiticus* was able to grow and produce aflatoxins on pistachios, but at longer inoculation periods a natural decrease of aflatoxins was registered. The highest reduction for AFB_1 was recorded at eight days after inoculation for both strains (54.9% and 52.5%), anyway both antagonists were able to reduce the fungal incidence and the number of spores on pistachio with a stronger effect during the first five days after inoculation. Both bacterial strains showed good antifungal activity and aflatoxin reduction on pistachio kernels. Altogether, these results indicate that *Bacillus* species could be considered as potential biocontrol agents to reduce the growth of mycotoxigenic fungi and the subsequent aflatoxin contamination of nuts in practice.

ROASTING AND COLD ATMOSPHERIC PLASMA ARE EFFICIENT METHODS FOR AFLATOXIN DECONTAMINATION ON HAZELNUTS. I. Siciliano[1], D. Spadaro[1,2], M.L. Gullino[1,2]. *[1]Università degli Studi di Torino, Centro AGROINNOVA, Largo Braccini 2, 10095 Grugliasco (TO), Italy. [2]Università degli Studi di Torino, DISAFA, Largo Braccini 2, 10095 Grugliasco (TO), Italy. E-mail: ilenia.siciliano@unito.it*

Aflatoxins, produced by *Aspergillus flavus* and *A. parasiticus* are a group of secondary metabolites dangerous to humans and animals that can contaminate different foodstuffs, such as nuts. Food processes, including roasting, may have different effects on mycotoxins, and high temperatures have proven to be very effective in the reduction of mycotoxins. Traditional static hot air roasting and infra-red rays roasting were applied and compared for the detoxification of hazelnuts from aflatoxins. At the temperature of 140°C for 40 min of exposure, detoxification was effective for both roasting techniques, residual aflatoxins were lower than 5%. After roasting, the perisperm was detached from the nuts, residual aflatoxins in the perisperm ranged from 80 up to 100%. Cold atmospheric pressure plasma also has the potential to be a promising method for aflatoxin

detoxification on food. On hazelnuts, with a 1000 W power and 12 min exposure, a reduction in the concentration of total aflatoxins and aflatoxin B_1 of over 70% was obtained. Aflatoxins B_1 and G_1 were more sensitive to plasma treatments compared to aflatoxins B_2 and G_2, respectively. Under plasma treatment, aflatoxin B_1 was more sensitive compared to aflatoxin G_1. The synergistic use of these two treatments along the hazelnut production chain could reduce the health risks associated with the presence of aflatoxins.

DIFFUSION OF BAKANAE DISEASE WITHIN THE RICE FIELD. D. Spadaro[1,2], S. Matić[1], A. Garibaldi[1], M.L. Gullino[1,2]. *[1]Università degli Studi di Torino, Centro AGROINNOVA, Largo Braccini 2, 10095 Grugliasco (TO), Italy. [2]Università degli Studi di Torino, DISAFA, Largo Braccini 2, 10095 Grugliasco (TO), Italy. E-mail: davide.spadaro@unito.it*

Bakanae is a monocyclic disease caused by *Fusarium fujikuroi*. The fungus is easily spread by conidia from the infected plants by wind and water. This study has been carried out to confirm the involvement of the wind in the conidial spread and in Bakanae diffusion in the rice field. A seven-day spore trap was located in the center of a rice field (Vercelli, Northern Italy), sown with local rice lines in order to capture air-borne particles. The rice lines were surrounded by a susceptible rice cultivar, 'Galileo', highly infected with *F. fujikuroi*. Spore monitoring was performed from flowering until harvest on a daily basis. There was no uniform trend in conidial transmission of *F. fujikuroi* during the monitored period by the microscopic observations of the tape, but there was an increase during the flowering and late maturation stage. A slight increase in diffusion of conidia was found at the milky stage of grain maturation, too. A higher occurrence of winds and rains was also registered at flowering and at the end of maturation, compared to the other periods of the monitoring and the previous cultivation seasons, which suggests that wind and rain might participate in conidial transmission of *F. fujikuroi*. In conclusion, the results obtained show that aerial conidial diffusion of *F. fujikuroi* happens, as a consequence of the spread of conidia from the severely infected rice cultivar.

MYCOTOXIGENIC FUNGI AND MYCOTOXINS IN CHESTNUTS AND DERIVATIVES. D. Spadaro[1,2], S. Prencipe[1,2], I. Siciliano[2], A. Garibaldi[1,2], M.L. Gullino[1,2]. *[1]AGROINNOVA, Università di Torino, Largo P. Braccini 2, 10095 Grugliasco, Torino, Italy. [2]DISAFA, Università di Torino, Largo P. Braccini 2, 10095 Grugliasco, Torino, Italy. E-mail: davide.spadaro@unito.it*

Italy is the first chestnut producing country in Europe. Almost 20% of the total production is devoted to industrial processing, including chestnut flour, dried chestnuts and marrons glacés. In postharvest, chestnuts and derivate products can be affected by parasitic fungi, including species of *Penicillium*, agents of green mould, and some species of *Aspergillus*, able to produce mycotoxins, among the others aflatoxins and ochratoxin A. European Commission Regulation 165/2010 establishes the maximum thresholds for aflatoxins in nuts, including chestnuts. Nowadays, the levels of other mycotoxins are not regulated in chestnuts. Aflatoxins are produced by *A. parasiticus* and *A. flavus*. Among the *Penicillium* spp., *P. crustosum* is able to produce ochratoxin A, penitrem A and roquefortine C, *P. expansum* can produce roquefortine C and patulin, while *P. bialowiezense* is able to produce mycophenolic acid. Prevention of contamination by mycotoxigenic fungi represents the most rational and economic strategy to reduce the mycotoxin risk. When prevention is not effective, mycotoxin detoxification can be an alternative to be developed for the chestnut chain.

***FcRav2*, A GENE WITH ROGDI DOMAIN INVOLVED IN FUSARIUM HEAD BLIGHT AND CROWN ROT ON DURUM WHEAT CAUSED BY *FUSARIUM CULMORUM*. F. Spanu[1], B. Scherm[1], I. Camboni[1], V. Balmas[1], G. Pani[1], S. Oufensou[1,2], N. Macciotta[1], M. Pasquali[3], Q. Migheli[1].** *[1]Dipartimento di Agraria, Università degli Studi di Sassari, Via E. De Nicola 9, I - 07100 Sassari, Italy. [2]Laboratoire de Bio-surveillance de l'environnement, Faculté des Sciences de Bizerte, Route de Tunis, 7021 Zarzouna, Université de Carthage. [3]DeFENS-Department of Food Environmental and Nutritional Sciences, Università degli Studi di Milano, Via Celoria 2, 20133 Milano, Italy. E-mail: soufensou@uniss.it*

Fusarium culmorum is a soil-borne fungal pathogen able to cause foot and root rot and Fusarium head blight on small grain cereals, particularly on wheat and barley. It causes significant yield and quality loss and results in the contamination of kernels with type B trichothecene mycotoxins. Knowledge on pathogenicity factors of this fungus is still limited. A transposon tagging approach based on the *mimp1/impala* double component system has allowed us to select a mutant altered in multiple metabolic and morphological processes, trichothecene production and virulence. The flanking regions of *mimp1* were used to seek homologies in the *F. culmorum* genome and revealed that *mimp1* had reinserted within the last exon of a gene encoding a hypothetical protein of 318 amino acids which contains a ROGDI like leucine zipper domain, supposedly playing a protein-protein interaction or a regulatory role. By functional complementation and bioinformatic analysis we characterized the gene as yeast *Rav2* homologue, acknowledging the high level of divergence in multicellular fungi. Deletion of *FcRav2* or its orthologous gene in *F. graminearum* highlighted its ability to influence a number of functions including virulence, trichothecene type B biosynthesis, resistance to azoles and resistance to osmotic and oxidative stress. Our results indicate that the FcRav2 protein (and possibly the RAVE complex on the whole) may become a suitable target for new antifungal drug development or plant-mediated resistance response also in filamentous fungi of agricultural interest.

EU-COST ACTION CA16107 - EUROXANTH: INTEGRATING SCIENCE ON *XANTHOMONADACEAE* FOR INTEGRATED PLANT DISEASE MANAGEMENT IN EUROPE. E. Stefani[1], V. Catara[2], E. Emeriau[3], R. Koebnik[4]. *[1]Università di Modena e Reggio Emilia, Dipartimento di Scienze della Vita, via Amendola 2, 42122 Reggio Emilia (Italy). [2]Università di Catania, Dipartimento di Agricoltura, Alimentazione e Ambiente, via S. Sofia 100, 95123 Catania, Italy. [3]COST Association, Avenue Louise 149, 1050 Bruxelles, Belgium. [4]IRD, Cirad, Université Montpellier, UMR IPME, 911 Avenue Agropolis, 34394 Montpellier, France. E-mail: emilio.stefani@unimore.it*

Bacteria of the family *Xanthomonadaceae*, including species of *Xanthomonas* and *Xylella fastidiosa*, are devastating plant pathogens. Many are quarantine organisms in the EU and their study is of uttermost importance. These pathogens infect all kinds of crop plants. The COST Action CA16107 "EuroXanth" aims at creating an interdisciplinary network in order to develop strategies for sustainably protecting plants and prevent yield losses. COST (European Cooperation in Science and Technology) is a funding agency for research and innovation networks. COST Actions help connect research initiatives and enable scientists to grow and share ideas with their peers. Specifically, this COST Action addresses key aspects of the pathogen-vector-host interactions, from the cellular to the population level. A better insight into population structures and virulence mechanisms of the pathogens, together with the exploration of the molecular mechanisms underlying disease resistance, will enable development of durably resistant plant cultivars and exploitation of bio-control schemes tailored to population

and pathogen. This COST Action has duration of 4 years (March 2017-March 2021) and will generate a platform that gathers experts from different disciplines, such as molecular diagnostics, molecular host-microbe interactions, plant resistance breeding, etc. The network includes 43 working groups from 21 different countries. Joining their efforts will help to develop and implement effective plant protection schemes, be it via resistant crop cultivars or via other control mechanisms. This goal will be achieved by mobilizing and training scientists from major European institutions, regulatory bodies and commercial companies working on the various aspects of this complex of problems.

NEXT GENERATION ECOFRIENDLY CONTROL OF GRAM NEGATIVE PLANT PATHOGENIC BACTERIA: VIRULENCE INHIBITING PEPTIDES AND POLYPHENOLS FROM NO-FOOD PLANT BIOMASS. S. Tegli, M. Cerboneschi, C. Biancalani, S. Calamai, L. Bini. *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. E-mail: stefania.tegli @unifi.it*

Plant diseases caused by bacteria may be highly destructive under adverse environmental conditions or in the case of quarantine pathogens. Bacterial diseases of plants still remain a challenging issue, basically relying on the use of copper and antibiotics, the latter not allowed in EU for plant protection. Given the negative ecotoxicological profile of copper, alternatives to its use are urgently needed, to meet the demands concerning agro-industry productivity and environmental protection. Here we propose an innovative strategy, based on the use of newly designed peptides and of plant polyphenols extracted from no-food biomass by a "green chemistry" process, targeting bacterial pathogenicity and virulence mechanisms, but not viability, thus avoiding the risk to develop any resistance. Both virulence inhibiting peptides (VIPs) and plant polyphenol extracts affect the TTSS and QS functionality, both *in vitro* and *in planta*, at concentrations of 30-60 µM, using *Pseudomonas savastanoi*, *P. syringae* pv. *tabaci* and *P. syringae* pv. *actinidiae* as model systems. Their effectiveness was demonstrated by pathogenicity trials and by bacterial gene expression studies, through real time PCR and several promoter-reporter systems. VIPs effectiveness was also demonstrated in *Nicotiana tabacum* and *Actinidia chinensis* stably transformed for VIPs expression, when challenged by *P. syringae* pv. *tabaci* and pv. *actinidiae*, respectively. No negative side-effects and no toxicity have been found on soil microflora, on model organisms and microorganisms, on biomimetic cellular membranes, as well as on Ca-ATPase pumps.

We gratefully acknowledge EU Commission (project LIFE12 ENV/IT/336 "AFTER Cu" and LIFE13 ENV/IT/461 EVERGREEN) for the financial support.

SURVEYS ON SWEET PEPPER (*CAPSICUM* spp.) IN SICILY (ITALY). A. Tiberini[1], A. Fontana[1], G. Leo[1], L. Tomassoli[2], S. Davino[3]. *[1]Università degli Studi "Mediterranea" di Reggio Calabria, Feo di Vito, 89122 Reggio Calabria (RC), Italy. [2]Consiglio per la ricerca in agricoltura e l'analisi dell'economia agraria - Centro di ricerca difesa e certificazione, Via C.G. Bertero 22, 00156 Roma (RM), Italy. [3]Università degli Studi di Palermo, Dipartimento Scienze Agrarie e Forestali, Viale delle Scienze ED. 5, 90128 Palermo, Italy. E-mail: antonio.tiberini@unirc.it*

Surveys to determine the incidence, diversity and distribution of viruses infecting sweet pepper (*Capsicum annuum*) in controlled conditions were conducted in several farms located in the two main production districts (Mazzara del Vallo and Ragusa) in Sicily, in 2017. Symptomatic plants were collected showing mosaic, mottling, puckering, reduction in leaf size, vein yellowing, interveinal yellowing, fruit deformation and stunting. Preliminary, symptomatic leaf samples were examined by ELISA for the most common viruses reported on pepper as: Potato virus Y (PVY), Pepper mild mottle virus (PMMoV), Tobacco mosaic virus (TMV), Cucumber mosaic virus (CMV), Tomato mosaic virus (ToMV), Tomato spotted wilt virus (TSWV), Alfalfa mosaic virus (AMV). Further, after nucleic acid extraction (Real Kit, Durviz), RT-PCR assays were performed using species-specific and genus-specific primer sets to investigate the presence of Tomato chlorosis virus (ToCV), Tomato infectious chlorosis virus (TICV), Pepper yellow leaf curl virus (PYLCV) and Pepper vein yellows virus (PeVYV). Results showed that the highest viral incidence in both surveyed areas is related to TSWV and poleroviruses (PYLCV and PeVYV), thereof few representative isolates were included in a phylogenetic analysis to better investigate the evolutionary status. Data confirmed how the *Polerovirus* genus can be regarded as a new phytosanitary threat in Sicily, especially in view of their high recombination capability.

SOURCES OF RESISTANCE TO THE DOWNY MILDEW AGENT IN THE EUROPEAN GRAPEVINE GERMPLASM. S.L. Toffolatti[1]*, G. De Lorenzis[1]*, G. Maddalena[1], A. Costa[2], C. Bonza[2], P. Casati[1], G. Venturini[1], M. Pindo[3], A. Cestaro[3], O. Failla[1], P.A. Bianco[1], F. Quaglino[1]. *[1]Università degli Studi di Milano, Dipartimento di Scienze Agrarie e Ambientali - Produzione, Territorio, Agroenergia (DISAA), via Celoria 2, 20133 Milano, Italy. [2]Università degli Studi di Milano, Dipartimento di Bioscienze, via Celoria 26, 20133 Milano, Italy. [3]Fondazione E. Mach, Centro Ricerca e Innovazione, Via E. Mach 1, 38010 San Michele all'Adige (TN). E-mail: silvia.toffolatti@unimi.it*

Exploiting the natural defense mechanism of a plant is one of the most specific, environmentally safe and innovative ways to protect agricultural crops from pathogen infections. Grapevine is affected by *Plasmopara viticola* (Berk. *et* Curt.) Berl. and De Toni, a pathogen of North American origin that causes downy mildew. Since the most common cultivars of *Vitis vinifera* L., the European grapevine, are highly susceptible to *P. viticola*, sources of resistance have been traditionally searched in the American germplasm that co-evolved with the pathogen. The availability of a huge collection of less common *V. vinifera* germplasm coming from Eastern Europe, recently led to the discovery of a cultivar, 'Mgaloblishvili N', showing low susceptibility to *P. viticola*. The interaction between the pathogen and 'Mgaloblishvili N' was characterized by analyzing phenotypic traits and genes differentially expressed in inoculated and non-inoculated leaves, to identify putative pathways of the plant response to the pathogen. The same analysis was carried out on two reference cultivars: 'Pinot noir N', susceptible to the pathogen, and 'Bianca B', a *Vitis* interspecific cross holding resistance traits. The results showed a significantly reduced disease severity and sporulation in 'Mgaloblishvili N', compared to 'Pinot noir N', associated with alterations of the pathogen structures, and significant differences in the expression levels of genes encoding defense proteins. 'Mgaloblishvili N' transcriptome proved to be different from that of the reference cultivars, highlighting specific molecular mechanisms of plant-pathogen interaction that should be more deeply investigated to exploit the resistant traits in *V. vinifera* breeding programs.

**These authors contributed equally to the work*

MOLECULAR IDENTIFICATION OF *VENTURIA ASPERATA* FROM ATYPICAL SCAB-LIKE SYMPTOMS ON APPLES IN ITALY. C. Turan[1], M. Menghini[1], G. Ceredi[2], M. Mari[1],

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Similar apple scab symptoms were first observed at the end of July in 2012 in northern Italy (Cesena) on fruits of apple cultivar Modì carrying the *Rvi6* major resistance gene to *Venturia inaequalis*. The aim of this work was to identify the causal agent of the atypical scab-like symptoms by molecular techniques. Symptomatic fruits were collected during May in one orchard in 2015. Ten monoconidial isolates were obtained through recovering the conidia from *ca*. 10 fruits. Conidial suspension was then streaked on Petri dishes of water agar amended with streptomycin sulfate. After 24 h of incubation at 20°C, single germinated spores were selected under stereomicroscope, then picked up and placed on PDA amended with three antibiotics. The isolates were cultivated at 20°C until molecular characterization together with the reference strain of *Venturia asperata*. Amplification of ITS fragments was carried out to specifically amplify rDNA of *V. asperata*, *V. inaequalis* and *Venturia pirina*. Approximately 4-5 hyphae were removed from each isolate and transferred without DNA extraction to the PCR tube with the addition of BSA. DNA amplification was obtained for all isolates by primers specific for *V. asperata*, while no amplification was observed using primers specific for *V. inaequalis* and *V. pirina*. These results point out the presence of *V. asperata* from the atypical scab-like symptoms but further studies are in progress to obtain a more precise identification of the pathogen.

CRISPR-CAS FOR THE GENOME EDITING OF TWO *TRICHODERMA* spp. BENEFICIAL ISOLATES. I. Vicente Muñoz, S. Sarrocco, G. Vannacci. *Università di Pisa, Dipartimento di Scienze Agrarie, Alimentari e Agro-ambientali, Via del Borghetto 80, 56124 Pisa. E-mail: i.vicente@studenti.unipi.it*

The genus *Trichoderma* includes different species with high value as biocontrol and plant growth promoter agents, such as *Trichoderma gamsii* T6085 and *Trichoderma afroharzianum* T6776, well known for the ability to control Fusarium Head Blight causal agents and for the beneficial effect on tomato plant, respectively. In this work, these isolates have been chosen to be genome-edited by the CRISPR-Cas9 technology, in order to develop a proof of concept of the feasibility of this new technique in our strains. A gene encoding a polyketide-synthase (*PKST*) was chosen as a target gene, since its disruption can be easily detected phenotypically. Thanks to the availability of the genomes of both the isolates, the *PKST* nucleotide sequence was used to design the RNA-guide to be included in the RGR-cassette via Gibson Assembly. The cassette was then assembled in a Cas9 expressing plasmid and the final vector used for fungal transformation by protoplasts. The presence of a truncated AMA1 sequence enables to remove the plasmid from the edited strains simply by reducing the selective pressure. Resulting transformants will be phenotypically and molecularly checked to verify the knockout of the selected gene. The presence of foreign DNA into the mutants will be also analyzed to contribute to the debate about the inclusion of this type of genetically modified microorganisms within GMO. The ability to genetically manipulate beneficial isolates with the CRISPR-Cas9 technique represents a tool to deepen our knowledge about how these fungi interact with their hosts and how to exploit these beneficial interplays.

***PYTHIUM MYRIOTYLUM*, CAUSAL AGENT OF CROWN AND ROOT ROT ON GREEN BEAN SOILLESS CULTURE IN ITALY. S. Vitale[1], L. Luongo[1], E. Marinelli[1], M. Galli[1], R. Bellardini[2], A. Belisario[1].** *[1]Consiglio per la Ricerca in Agricoltura e l'analisi dell'economia agraria, Centro di ricerca per la Difesa e la Certificazione (CREA-DC), Via C.G. Bertero 22, 00156 Roma (Italy). [2]Consulente del Centro Agroalimentare di Roma ScpA, Via della Tenuta del Cavaliere 1, 00012 Guidonia (RM). E-mail: salvatore.vitale@crea.gov.it*

During the summer 2016 green bean plants (*Phaseolus vulgaris* cv. "Valdarno") soilless cultivated in pots with sterile perlite and coconut fiber in greenhouses in central Italy (Latium Region) showed symptoms of crown and root rot followed by wilting. The infected crowns and roots appeared water-soaked with a cinnamon-brown color. In greenhouse we estimated 20-30% of dead plants occurring in distinct and well delimited areas. *Pythium* spp. colonies were consistently isolated from diseased plants by using a semi selective medium (P_5ARP) and three pure cultures were obtained by single-hyphal transfers on potato dextrose agar (PDA). On the basis of morphological features, the isolates were identified as *P. myriotylum* Drechsler. The identity was confirmed by internal transcribed spacer (ITS) sequence comparison with 99% homology with sequences available in GenBank (e.g. KY019272), and by cytochrome oxidase subunit 1 (COI) gene with 100% homology with HQ708745. The sequences of the three isolates AB290, AB291 and AB292 were deposited in European Nucleotide Archive (ENA). Pathogenicity tests confirmed *P. myriotylum* isolates pathogenic onto artificially inoculated *P. vulgaris* cv."Valdarno" plants. *P. myriotylum* is reported as a causal agent of root rot on several economically important crops including peanut, tomato, rye, wheat, oat, cucumber, soya bean, sorghum, tobacco, cabbage and maize. However, within Europe *P. myriotylum* on green bean was just reported in Spain. To our knowledge, this is the first report of *P. myriotylum* as the causal agent of root and stem rot on green bean plants in Italy.

GWAS-BASED IDENTIFICATION OF LOCI FOR BAKANAE DISEASE RESISTANCE IN RICE. A. Volante[1], A. Tondelli[2], M. Aragona[3], M.T. Valente[3], C. Biselli[2], F. Desiderio[2], P. Bagnaresi[2], S. Matic[4], M.L. Gullino[4,5], A. Infantino[3], D. Spadaro[4,5], G. Valè[1,2]. *[1]Council for Agricultural Research and Economics (CREA), Rice Research Unit, S.S. 11 to Torino, Km 2.5, 13100, Vercelli, Italy. [2]Council for Agricultural Research and Economics (CREA), Genomics Research Centre, Via S. Protaso 302, 29017, Fiorenzuola d'Arda, Piacenza, Italy. [3]Council for Agricultural Research and Economics (CREA), Plant Pathology Research Centre, Via C.G. Bertero 22, 00156, Roma, Italy. [4]AGROINNOVA, Università di Torino, Largo P. Braccini 2, 10095 Grugliasco, Torino, Italy. [5]DISAFA, Università di Torino, Largo P. Braccini 2, 10095 Grugliasco, Torino, Italy. E-mail: davide.spadaro@unito.it*

Bakanae disease is one of the most serious and oldest problems of rice production, caused by one or more seed-borne *Fusarium* species, mainly *F. fujikuroi*. The disease may affect rice plants from the pre-emergence to mature stage. Identification and cultivation of resistant rice cultivars represent efficient procedures for bakanae disease control. However, only few rice accessions were reported to have high resistance to the infection. Similarly, the knowledge of mapped genes conferring resistance to bakanae is very restricted. In the present study, a japonica rice collection comprising 142 accessions was screened for bakanae resistance after artificial inoculation with *F. fujikuroi*. A large variability was observed, with few lines showing mild disease symptoms, indicating that genetic determinants for resistance are segregating in the collection under analysis. Molecular variation between the same rice accessions was evaluated by a Genotyping-by-Sequencing approach, which yielded a total of about 31,000 informative single nucleotide polymorphisms (SNPs) with a Minor Allele Frequency above 10%. The genome-wide association study approach (GWAS) uncovered two genomic

regions highly associated with the observed phenotypic variation for response to bakanae infection on the short arm of chromosome 1 and on the long arm of chromosome 4. High levels of phenotypic resistance to bakanae were associated with the cumulated presence of the resistant alleles at the two resistance loci, suggesting that they can provide useful levels of disease protection in breeding for resistance.

SYMBIOTIC AGRICULTURE: PLANT GROWTH PROMOTION AND BIOCONTROL ACTIVITY OF BENEFICIAL MICROORGANISMS. S.S.K.P. Vurukonda, D. Giovanardi, E. Stefani. *Università di Modena e Reggio Emilia, Dipartimento di Scienze della Vita, via Amendola 2, 42122 Reggio Emilia, Italy. E-mail: saishivakrishnaprasad.vurukonda@unimore.it*

Symbiotic microbial inoculation is emerging as a potential technology for sustainable agriculture. Towards a sustainable agricultural vision, crops need to be equipped with disease resistance to various biotic and abiotic stresses, together with better nutritional value. To fulfil the above desired crop properties, one possibility is to use soil microorganisms (bacteria, fungi, algae, etc.) in order to enhance microbial biodiversity. Among these potential soil microorganisms, bacteria known as plant growth promoting rhizobacteria (PGPR) are the most promising. In the present study three *Streptomyces* sp. strains SB14, SA51 and SL81, two *Pseudomonas* sp. strains PT65 & PN53 and one *Agrobacterium* sp. strain AR39 were evaluated *in vitro* for different plant growth promoting and biocontrol activities. Our aim is to identify possible antagonists able to inhibit different plant bacterial pathogens like *Xanthomonas vesicatoria*, *Clavibacter michiganensis* subsp. *michiganensis*, *Clavibacter michiganensis subsp. sepedonicus*, *Acidovorax citrulli* and *Ralstonia solanacearum*. All the strains were screened for biocontrol activity on three different media ISP – 2 (International streptomyces project), PDA (Potato Dextrose Agar) and HPDA (Half Strength PDA) and AIA (average inhibition area) was calculated. Among the isolates, each strain showed different ability to inhibit the pathogens: *Streptomyces* sp. strain SA51 was found to be most active. The most prospective strains were further evaluated in the field, as possible biocontrol agents for the tomato spot disease (*X. vesicatoria*), singularly and as a consortium. Results will improve our understanding on the use of such microbial biocontrol agents and will implement innovative biocontrol strategies to bacterial diseases.

TWIG WILTING AND LEAF SPOTTING CAUSED BY COPPER-RESISTANT *PSEUDOMONAS SYRINGAE* pv. *SYRINGAE* STRAINS ON *ACTINIDIA DELICIOSA*. L. Zampella, F. Mastrobuoni, M. Petriccione, M. Scortichini. *Consiglio per la ricerca in agricoltura e l'analisi dell'economia agraria, Centro di ricerca per l'Olivicoltura, Frutticoltura e Agrumicoltura - sede di Caserta, Via Torrino 3, 81100 Caserta (CE). E-mail: luigi.zampella@crea.gov.it*

During field monitoring performed in Campania region (southern Italy) in 2016 to ascertain the incidence and severity of kiwifruit bacterial canker, caused by *Pseudomonas syringae* pv. *actinidiae* (*Psa*), extensive leaf spotting and twig dieback were noticed in one adult *Actinidia deliciosa* cv. Hayward orchard. These symptoms were mainly observed in spring and autumn on about 25% of the plants. Isolations were carried out onto King's medium B (KB) by taking small pieces of tissue (1-2 mm) from the edge of the necrotic lesions. The LOPAT tests were performed with some colonies fluorescent on KB. In addition, pathogenicity tests were carried out on leaves of pot-cultivated, two-years-old *A. deliciosa* cv. Hayward and pear cv. Conference plants, as well as on leaves of tomato and pepper seedlings and lemon fruits, by using a bacterial concentration of $1\text{-}2 \times 10^7$ cfu/ml. Control plants were inoculated solely with sterile water. The isolates were also assessed for the presence of the *syrB* gene and for resistance to copper by detecting the *copABCDRS* operon as well as for growth onto the NBY medium added with serial doses of copper sulphate. After 10 days of incubation, the leaves and fruits started to show tiny yellowish and necrotic lesions. The isolates possessed *syrB* gene, the entire *cop* operon and grew on NBY supplemented with 2.1 mM of copper sulphate. *Psa* was not found in the orchard. This is the first report of copper-resistant *P. s.* pv. *syringae* strains causing severe symptoms on *A. deliciosa* in Italy.

ANTAGONISTIC PROPERTIES OF EPIPHYTIC STRAINS OF *ASPERGILLUS CRISTATUS* AND *TALAROMYCES PINOPHILUS* AS RELATED TO PRODUCTION OF SECONDARY METABOLITES. B. Zimowska[1], R. Nicoletti[2], E. Krol[1], R. Marra[3], A. Furmanczyk[1], L. Gioia[3], S.L. Woo[4,5], M. Lorito[3,4], F. Vinale[4]. *[1]Department of Plant Pathology and Mycology, University of Life Sciences, Leszczyńskiego 7, 20069 Lublin, Poland. [2]Consiglio per la Ricerca in Agricoltura e l'Analisi dell'Economia Agraria. Current address: Dipartimento di Agraria, Universita degli Studi di Napoli Federico II, Via Università 100, 80055 Portici (NA), Italy. [3]Dipartimento di Agraria, Università degli Studi di Napoli Federico II, Via Università 100, 80055 Portici (NA), Italy. [4]Consiglio Nazionale delle Ricerche - Istituto per la Protezione Sostenibile delle Piante (CNR-IPSP), Via Università 133, 80055 Portici (NA), Italy. [5]Dipartimento di Farmacia, Università degli Studi di Napoli Federico II,Via D. Montesano 49, 80131 Napoli (NA). E-mail: francesco.vinale@ipsp.cnr.it*

The fungal biocenosis harboured in galls produced by midges of the genus *Asphondylia* (*Diptera*: *Cecidomyiidae*) in flowers of medicinal plants belonging to the *Lamiaceae*, is dominated by *Botryosphaeria dothidea* and by species in the *Cladosporium cladosporioides* and *Alternaria alternata* species complexes. Normal flowers, without galls, host a more varied assemblage of fungi, including species known for their antagonistic properties, which may play a role in impairing the formation of galls by inhibiting the fungal symbiont necessary for the development of the midges. Among the many putative antagonistic fungi recovered from the ovaries of flowers of *Ballota nigra* (Konopnica, Poland) and *Micromeria graeca* (Astroni, Italy), two fungi, *Aspergillus cristatus* and *Talaromyces pinophilus*, were selected to evaluate their effects towards *B. dothidea* and other plant pathogens. The inhibitory and/or mycoparasitic ability of these fungi was assessed in dual cultures on PDA, according to the biotic series method. The strongest pathogen inhibition was observed with *T. pinophilus*, with a red halo noted at the edge of the inhibition zone as a sign of changes in the array of fungal compounds released into the medium. Moreover, coiling around pathogen hyphae was observed in the interaction zone, together with deplasmolysis and lysis of the mycelium. Metabolomic analysis of substances extracted from the growth substrate indicated that a complex combination of antifungal metabolites were synthesized by the interacting fungi. In particular, *T. pinophilus* produced funicone-like compounds, while *A. cristatus* released penitricins, neoechinulins, and related products.

XXIII Annual Meeting of the Italian Society for Plant Pathology (SIPaV)

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Finito di stampare nel mese di settembre 2017
in Pisa dalle
EDIZIONI ETS
Piazza Carrara, 16-19, I - 56126 Pisa
info@edizioniets.com - www.edizioniets.com