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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.

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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.

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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.

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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 1-2 × 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.