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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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