82. GENOME MINING, PATHOGENECITY AND SECONDARY METABOLISM OF THREE STRAINS OF *FUSARIUM FUJIKUROI*, THE CAUSAL AGENT OF BAKANE DISEASE ON RICE. E. Piombo[1], H. Banani[1,2], I. Siciliano[2], P. Abbruscato[3], A. Acquadro[1], M.L. Gullino[1,2], D. Spadaro[1,2]. *[1]Università degli Studi di Torino, Centro AGROINNOVA, Largo Braccini 2 - 10095 Grugliasco (TO), Italy. [2]Universita degli Studi di Torino, DISAFA, Largo Braccini 2 - 10095 Grugliasco (TO), Italy. [3]Bioeconomy Unit, Parco Tecnologico Padano, via Einstein, 26900 Lodi, Italy. E-mail: davide.spadaro@unito.it*

Bakanae is an important seedborne disease of rice, caused by *Fusarium fujikuroi.* This pathogen can produce a wide range of secondary metabolites, including fumonisins, gibberellins and fusaric acid. In order to gain insight into secondary metabolites (SM) synthesis in *Fusarium fujikuroi,* we sequenced the genome of three strains named Augusto2, CSV1 and I1.3, identified the allelic variants in the genes responsible for SM production, and compared the virulence on rice and the SM production *in vitro* and on rice. Sequence analysis was conducted by *de novo* genome assembly. Three genomes of 42.8 Mb on average were obtained. The gene clusters responsible for fumonisin, gibberellin and fusaric acid production, formed by 15, 7 and 12 genes, respectively, were analyzed and aminoacidic differences were identified for *fum1, fum13* and *fum21. In vitro* colony diameters significantly increased with time and the three *F. fujikuroi* strains exhibited distinct differences in colony morphology and growth kinetics. We further compared the virulence and fumonisin production of the three strains on rice 'Galileo'. At 3 weeks post germination, *F. fujikuroi* strain I1.3 showed statistically higher virulence compared to Augusto2 and CSV1. Augusto2 was the major producer of fumonisins both *in vitro* and *in vivo*, followed by CSV1and I1.3. CSV1 was unable to produce gibberellins *in vivo* and *in vitro* on Petri dish, confirming the different symptomatology of CSV1 on rice, characterized by dwarfing and chlorosis, but lack of stem elongation.

This work permits to add a new tile to the complex puzzle of rice-F. fujikuroi interactions.

83. A REVISED AND EFFECTIVE PIPELINE BASED ON RELATIVE COVERAGE FOR THE GENOME RECONSTRUCTION OF PHYTOPLASMA AND OTHER FASTIDIOUS PROKARYOTES. C. Polano, P. Ermacora, M. Martini, R. Musetti, N. Loi, G. Firrao. *Università degli Studi di Udine, DI4A, Via delle Scienze 206 - 33100 Udine, Italy. E-mail: paolo.ermacora@uniud.it*

An alternative to the difficult, inefficient and time consuming methods that require purification of the pathogen DNA for its genomic analysis is to sequence a large library of DNA extracted from diseased plants and then select pathogen specific sequences. However, pathogen sequence selection is not trivial and many genome drafts published so far are incomplete.

The procedure developed here exploits the differential coverage of sequences originating from pathogen and those from host, due to the size difference between the prokaryote and the plant genomes and the relative abundance of pathogen even in samples with less than 10% pathogen DNA.

In brief, the procedure requires a reference genome for the uninfected plant, and the sequence reads obtained from an Illumina MiSeq. The procedure starts assembling the mixed genome reads and the reference genome reads if necessary. A perl script then calculates the coverages and then groups the contigs accordingly. The following steps are looped to find the optimum cutoff. The reads of the contigs with coverage higher than the cutoff are aligned against the healthy assembly. The non-mapping sequences are assembled with the A5 pipeline, to obtain the non-mapping-on-healthy contigs list. Another perl script queries a database from the reference genome contigs file to check the effectiveness of the cutoff and filtering parameter used.

Using this pipeline we have recently obtained very high quality draft assemblies of the phytoplasma strains associated with Lime Witches' Broom in Brazil, with the Cassava Frogskin Disease, with Chicory Phyllody and of a *Spiroplasma citri* strain.

84. SURVEY FOR THE PRESENCE OF *XYLELLA FASTIDIOSA* subsp. *PAUCA* STRAIN CoDiRO IN THE NATIVE FLORA OF THE SALENTO PENINSULA. O. Potere[1], L. Susca[1], F. Civita[1], S. Marullo[1], G. Loconsole[1], M. Saponari[2], D. Boscia[2], V.N. Savino[1,2], P. La Notte[2]. *[1]Dipartimento di Scienze del Suolo, della Pianta e degli Alimenti, Università degli Studi di Bari Aldo Moro, Via Amendola 165/A - 70126 Bari, Italy. [2]Istituto per la Protezione Sostenibile delle Piante, CNR, SS di Bari, Via Amendola 165/A - 70126 Bari, Italy. E- mail: oriana.potere@uniba.it*

Xylella fastidiosa subsp. *pauca* strain CoDiRO was identified as associated with the "Olive Quick Decline Syndrome", a devastating disease first observed in October 2013 in the southeastern Apulia. At least 350 plant species belonging to 75 families are reported as hosts of *X. fastidiosa.* These provide a source of inoculum for the vectors (xylem sap-feeding leafhoppers), thus playing a major epidemiological role and facilitating the entrenchment of the pathogen in the affected area. To investigate the CoDiRO strain host range in Salento, monthly samplings of the native flora of two heavily infected olive groves and of the side of adjacent roads were conducted from January 2014 onwards. One of the groves was grass-covered, whereas periodic tillage was performed in the other. Overall, more than 200 species of 50 families were sampled, observed for the presence of symptoms, photographed and identified. In the spring, *Philaenus spumarius* the main vector of the Salentinian *X. fastidiosa* strain was abundantly present on the herbaceous flora and shrubs at all sites. All samples, in pools of no less that 3 to 5 plants, were tested by DAS-ELISA and uncertain/positive results were verified by conventional and real time PCR. Bacterial isolates were obtained in axenic culture from some positive species. In a two-year survey, only *Euphorbia terracina* proved to be *Xylella*-positive among the herbaceous hosts, whereas some shrubs and subshrubs i.e. *Asparagus acutifolius*, *Cistus creticus*, *Myrtus communis*, *Phillyrea latifolia*, *Rhamnus alaternus* and *Rosmarinus officinalis* were infected. These results provide as strong indication that, rather than weeds, are the perennial shrubs that play a major role in the epidemiology of the *X. fastidiosa* in this area.

85. GENETIC DIVERSITY AND VIRULENCE OF STRAINS OF *PSEUDOMONAS SYRINGAE* pv. *ACTINIDIAE* ISOLATED FROM *ACTINIDIA DELICIOSA* IN PIEDMONT. S. Prencipe[1], A. Garibaldi[2], D. Spadaro[1]. *[1]Università di Torino, DISAFA, Grugliasco, Italy; - [2]Università di Torino, AGROINNOVA, Grugliasco, Italy. E-mail: angelo.garibaldi@unito.it*

Pseudomonas syringae pv. *actinidiae* (*Psa*), the causal agent of bacterial canker of kiwifruit, is responsible for significant economic losses, both in yield and quality. During the first severe outbreak (2008-2010) and few years afterwards 40 strains of *Psa* were isolated. To analyse the genetic diversity of the pathogen REP, RAPD-PCR, and MLST of six housekeeping and effector genes were performed. RAPD technique, compared to REP-PCR showed an increased level of resolution in evaluating the genetic diversity within the pathovar *actinidiae* of *P. syringae*, with the sets of primers used in this study. The molecular fingerprinting, *Na*, *Ne*, *H*, *I*, polymorphic loci, and AMOVA showed a high level of variability and genetic