

DISEASE NOTE

**A NEW BEGOMOVIRUS ASSOCIATED
WITH LEAF CURL DISEASE
OF *EUPHORBIA PULCHERRIMA***

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A virus isolate (G35-1) was obtained from *Euphorbia pulcherrima* with leaf curl and vein thickening symptoms from Tianyang in Guangxi province of China. PCR of total DNA extracts from the G35-1-infected plant was performed using the degenerate primer pair PA and PB, which were designed to amplify part of the intergenic region and the AV2 gene of DNA-A of begomoviruses (Zhou *et al.*, 2003). The resulting amplicon of approximately 500 bp was cloned and sequenced. Sequence comparisons showed that it is a fragment of a begomovirus genome. A full-length copy of G35-1 DNA-A was obtained by PCR using overlapping primers G35-1F (5'-GTCCTCGTCACAAACAAAAGG-3') and G35-1R (5'-CGATTGACCCATGATTCCT-3') designed based on the determined sequences. The complete DNA-A sequence of G35-1 was 2747 nucleotides in length (AM411424) and had a genetic organization typical of a begomovirus. Comparisons with DNA-A of other begomoviruses showed that G35-1 is most closely related to Tomato leaf curl Guangxi virus (AM236786) with an 86.3% nucleotide sequence identity. Based on the molecular data and sequence analysis, G35-1 is considered to be a new begomovirus species, for which the name *Euphorbia leaf curl Guangxi virus* (ELCGXV) is proposed. *Euphorbia pulcherrima* has previously been reported to be infected by *Euphorbia leaf curl virus* (ELCV) (Ma *et al.*, 2004). This is another distinct begomovirus infecting *Euphorbia pulcherrima*.

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DISEASE NOTE

**FIRST REPORT OF LEAF BLIGHT
OF *MEDICAGO SATIVA* CAUSED
BY *ALTERNARIA LONGIPES* IN INDIA**

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A leaf blight disease of alfalfa (*Medicago sativa*) was observed in January to March 2004-06 in different areas of South Bengal, India. The symptoms first appeared as pin-head like, small, ferruginous, brownish spots (2-8 mm) scattered at the tip of the leaflets. With time, the spots gradually enlarged in size (up to 20 mm), became irregular in shape, coalesced and became surrounded by a yellow halo. Ultimately, infected leaves were shed. Isolations on PDA at 27°C from diseased leaves yielded colonies of a fungus with abundantly branched, septate, brownish mycelium. The cultures produced effuse, amphigenous, erect, septate (41.5-68.5×2.5-3.3 µm) conidiphores bearing chains of obclavate, mostly 5-6 septate, muriform conidia 40.5-46×16-18 µm in size. Pathogenicity tests were carried out spraying ten healthy 2-month-old potted alfalfa plants with a conidial suspension (10⁵ conidia ml⁻¹). Controls were sprayed with sterile water and all plants were covered with plastic bags for the first 7 days. After 11±2 days, symptoms resembling those exhibited by naturally infected field plants developed on inoculated plants, whereas no symptoms were shown by controls. A pathogen identical to that used as inoculum was re-isolated from infected tissues of inoculated plants which was identified as *Alternaria longipes* (Ellis et Everhart.) Mason (Subramaniam, 1971) by the Agharkar Research Institute, Pune, India. This appears to be the first report of *A. longipes* on *M. sativa* in India.

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DISEASE NOTE

**FIRST REPORT OF DOWNY MILDEW
CAUSED BY *PERONOSPORA* SP.
ON BASIL IN NORTHERN IRAN**

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Common sweet basil (*Ocimum basilicum*), a world-wide-grown, annual, economically important aromatic plant, is cultivated on a commercial scale also in Iran. In the past couple of years, downy mildew was reported as a serious threat to sweet basil crops in several European countries (Switzerland, France, Belgium, Italy) and South Africa. In 2006, a severe outbreak of downy mildew was observed in sweet basil fields in Iran, which resulted in important crop losses. Initial symptoms were chlorotic leaves that soon developed a brown sporulation on the abaxial surface. Microscopic observations of the fungal growth were consistent with the characteristics of a *Peronospora* sp. The 130-290 µm (average 194 µm) long sporangiophores branched two to five times and ended with dichotomously branched denticels bearing single detachable sporangia. These were brown, elliptical, and measured 26-34×20-28 µm (average 30×24 µm). Sporangia were similar in shape, colour, and size range to those previously reported for the *Peronospora* sp. infecting sweet basil (Belbahri *et al.* 2005; Garibaldi *et al.* 2004). Molecular analyses were conducted on five leaves from three independent disease events. DNA extracted from spores was subjected to PCR amplification and sequencing of the internal transcribed spacer region (ITS1, 5.8S rDNA gene, ITS2) according to Belbahri *et al.* (2005). Resulting sequences were identical to one another (GenBank Accession No. EF153670, EF153669, EF153668, EF153667, EF153666). BLAST analyses revealed a 99% similarity to the *Peronospora* sp. that was isolated from sweet basil in Switzerland, Italy and South Africa. To our knowledge, this is the first report of this *Peronospora* sp. on sweet basil in Iran.

Belbahri L, Calmin G., Pawlowski J., Lefort F. 2005. Phylogenetic analysis and real time PCR detection of a presumably undescribed *Peronospora* species on sweet basil and sage. *Mycological research* **109**: 1276-1387.

Garibaldi A., Minuto A, Minuto G., Gullino M.L. 2004. First report of downy mildew on basil (*Ocimum basilicum*) in Italy. *Plant Disease* **88**: 312, 2004.

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DISEASE NOTE

**FIRST REPORT OF STEM AND ROOT ROT
OF *SCHLUMBERGERA TRUNCATA* CAUSED
BY *FUSARIUM OXYSPORUM* IN ARGENTINA**

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Schlumbergera truncata (Haw.) Moran (syn. *Zygocactus truncatus* (Haw.) K. Schum) is a vegetatively propagated, succulent species of the family *Cactaceae* (Dimitri, 1972), cultivated as potted ornamental under the name of "Santa Teresita" in Argentina and "Thanksgiving cactus" in English-speaking countries. In October 2005, a root and crown rot condition was observed in recently propagated plants, that caused loss of turgidity of the stem and, eventually, death of the plants. The disease was observed in a commercial greenhouse located in Escobar (Buenos Aires province, Argentina). An isolate of *Fusarium* sp. was obtained from stem and root fragments of diseased plants, that were plated on potato dextrose agar (PDA) after disinfection with 70% ethanol for 1.5 min and 0.2% NaOCl for 2 min. Pathogenicity tests were conducted by watering 5 potted *S. truncata* plants with a monosporic single conidial inoculum adjusted to 4.7×10⁷ spore/ml of sterilised distilled water. Each plant was irrigated with 1 ml of inoculum, enclosed in a polyethylene bag, and incubated at 22-24°C and a 12 h photoperiod. Five control plants were irrigated with sterilised distilled water. Branches of two inoculated plants decayed and dropped on the substrate 12 days after inoculation, while the rest of the plants showed crown rot. One of them was completely rotten after 19 days. Root rot developed on all inoculated plants, whereas the controls remained healthy. The fungus that incited the disease of artificially inoculated *S. truncata* was reisolated from symptomatic plants and identified as *Fusarium oxysporum* Schlecht.:Fr based on morphological and cultural characteristics (Booth, 1971; Nelson *et al.*, 1983). This is the first report of basal rot of *S. truncata* caused by *F. oxysporum* in Buenos Aires, and the first record of this disease in Argentina.

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Dimitri M.J., 1972. Enciclopedia Argentina de Agricultura y Jardinería. Vol. I. Descripción de las Plantas Cultivadas. 2nd Ed. ACME, Buenos Aires, Argentina.

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DISEASE NOTE

FIRST REPORT OF *PSEUDOCERCOSPORA NERIELLA* ON *NERIUM OLEANDER* IN TURKEY

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Nerium oleander is an important ornamental plant widely grown in gardens and parks of southern Turkey. In late spring 2007, a disease of oleander affecting 15 to 20% of the plants was observed in several parks of the Şanlıurfa district (south eastern Anatolia). Symptoms consisted in the presence of irregular to circular spots 1-9 mm in size, localized primarily on the upper leaf surface. Initially, spots were pale green but, with time, they turned yellowish, then grey and brown. Conspicuous yellowish areas became visible on the leaves when the spots merged. On the spots, a hypophyllous and barely visible mycelium was present. Secondary mycelium was external, extensive and largely intermingled with leaf trichomes. Hyphae were sub-hyaline, 1-2 µm wide, septate and branched, and bore conidiophores as lateral branches. Conidiophores were sub-hyaline to grayish-brown, erect and unbranched. They were borne terminally or laterally on the external mycelial hyphae, were 0-2 geniculate and 0-2 septate, conically shaped at the top, and 4-22×2-3 µm in size. The 20-95 µm long conidia had from 4 to 11 septa, were obclavate to filiform, undulate or bent, pointed to some extent at the top and obconically truncate at the base. Based on morphology, the fungus was identified as *Pseudocercospora neriella* (Sacc.) Deighton (Hyde *et al.*, 1997). Pathogenicity tests were successful in that inoculation of a conidial suspension to wounded oleander leaves reproduced the field symptoms, i.e. yellowish spots from which the fungus was reisolated. To my knowledge this appears to be the first report of *P. neriella* on oleander in Turkey.

Hyde K.D., Leung H.Y.M., Goh T.K., 1997. *Pseudocercospora neriella*. IMI Descriptions of Fungi and Bacteria, No. 132, Sheet 1316.

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DISEASE NOTE

FIRST REPORT OF *PEAR BLISTER CANKER VIROID* IN MALTAD. Attard¹, M. Afechtal², M. Agius¹, S. Matic³,
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In 2006, a tissue-printing hybridisation (TPH) method was used for investigating the presence and spread in Malta of *Pear blister canker viroid* (PBCVd) (Hernández *et al.*, 1992). The survey was mainly addressed to test PBCVd infection in the local pear cv. Babinella, which is of special interest for the country. A total of 113 trees grown near Rabat (Malta) were assayed. Although none of them showed any visible symptom, 14 (ca.12%) tested positive for PBCVd. Seven of these trees were used as source material for graft-inoculating 14 seedlings of the pear indicator LA62 (two seedlings for each infected accession). Six months later, PBCVd was detected in all inoculated seedlings by molecular hybridization and by RT-PCR assays, using the specific primers designed by Malfitano *et al.* (2003), and the expected 315 bp cDNA amplicon was sequenced. Negative results were obtained when the same analyses were done on LA62 seedlings which, in parallel experiments, had been graft-inoculated with material from TPH-negative Babinella trees grown in the same area. Like naturally PBCVd-infected trees, seedlings successfully infected by graft-inoculation did not show any symptom. Altogether, these data show that PBCVd occurs in symptomless cv. Babinella pears in Malta and further validate the TPH method for large scale surveys of PBCVd infection. To our knowledge this is the first report of PBCVd in Malta.

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DISEASE NOTE

**MOLECULAR EVIDENCE FOR APPLE
CHLOROTIC LEAF SPOT VIRUS
IN WILD AND CULTIVATED APRICOT
IN HIMACHAL PRADESH, INDIA**

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Wild apricot (*Prunus armeniaca*) is known as “chuli” or “zardalu” in Himachal Pradesh (HP, India) where it grows in the hills at altitudes of 1000-1700 meters. The fruits have medicinal properties and seedlings are used as rootstocks for commercial apricot, peach and plum orchards (Parmar and Kaushal, 1982). *Apple chlorotic leaf spot virus* (ACLSV), genus *Trichovirus*, has been reported to cause “viruela” and “butteratura” diseases in apricot and is cause of apricot decline (Sutic *et al.*, 1999). Infections are normally symptomless but can result in severe graft incompatibility with some *Prunus* combinations. During surveys for ACLSV infection in *P. armeniaca*, leaf samples (30 from cultivated and 23 from wild plants) were collected and subjected to ELISA in triplicate. Positive results were obtained from 14 samples. Inoculation of leaf extracts of the positive samples to *Chenopodium amaranticolor* resulted in small chlorotic spots on inoculated leaves. RT/PCR using degenerate primers (Accession Nos. AM490253 and AM490254) designed to yield amplicons corresponding to the complete coat protein gene gave a 758 bp product. Sequence analysis of amplicons from wild (AM498048) and cultivated (AM498045) apricot virus isolates showed 94% and 95% identity at the nucleotide and amino acid level among themselves, and were 92% to 96% identical at the amino acid level with two apple ACLSV isolates from HP. Indian ACLSV isolates were similar to others with the exception of a Turkish isolate [70% similarity (Ulubas and Ertunc, 2005)] (AJ586636). ACLSV from cultivated apricot was 91% similar to an Italian (AJ586635) and a Spanish (AJ586632) virus isolate. This is the first report of ACLSV infection of apricot in India, and of ACLSV infection of wild apricot.

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DISEASE NOTE

**GRAPEVINE YELLOW SPECKLE VIROID 1
AND GRAPEVINE YELLOW SPECKLE
VIROID 2 ISOLATES FROM CHINA**

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Of the five viroids recognised in grapevine (*Vitis vinifera*) by sequence analysis, *Hop stunt viroid*, *Australian grapevine viroid*, *Grapevine yellow speckle viroid 1* (GYSVd-1) and/or GYSVd-2 have been found in grapevines from China using dot blot hybridization with cRNA probes (Li *et al.*, 2006; Guo *et al.*, 2007). Since GYSVd-1 and GYSVd-2 are more than 80% similar in sequence, both are detected with riboprobes to GYSVd-1. GYSVd-1 and/or GYSVd-2 were detected in 29 of 70 samples tested. As GYSVd-1 sequences are more abundant in GenBank than those of GYSVd-2, GYSVd-1 may be more common than GYSVd-2 in cultivated grapevine. To investigate if both GYSVd-1 and GYSVd-2 exist in China, 12 of the samples previously shown to be GYSVd-positive were tested by RT-PCR, using primers 5'-TTGGATCC-CACCTCGGAAGGCC(G/T)CC-3' and 5'-TTGGATCC(T/A)AACCACAGGAACCACA-3', which can prime and amplify cDNA of both GYSVd-1 and GYSVd-2. The resulting amplicons were cloned into pGEM-T (Promega, Madison, WI, USA) and sequenced. GYSVd-1 (DQ371462-371470) was detected in cvs Zhiyuan-540, Guixiangyi, 86-11, Shafu Seedless, Jingchao, Beiquan and Thompson Seedless; GYSVd-2 (DQ377124-377132) was detected in cvs Black Olympia, Takasumi, Zaoyu, and Thompson Seedless. Thompson Seedless was the only doubly infected sample. To our knowledge, this is the first confirmation that both GYSVd-1 and GYSVd-2 are present in grapevines in China. Our results are also only the second report of a sequence for GYSVd-2.

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DISEASE NOTE

***FUSARIUM SAMBUCINUM* CAUSING
CANKER ON *BOUGAINVILLEA GLABRA***S.M. Wolcan^{1,2}, P.J. Grego^{1,2} and G.A. Lori^{1,2}¹ CIDEFI, Facultad de Ciencias Agrarias y Forestales, UNLP,
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Bougainvillea (*Bougainvillea glabra* L.) is an ornamental climbing shrub native to South America. In 2005, plants showing elongate cankers on branches and twigs were observed in La Plata city (Buenos Aires province). Cankers had brown or purple margins and were scattered or confluent. Leaves from affected branches and twigs showed minute tan spots and could desiccate and fall. Pieces (4×4 mm) of affected tissues were surface-sterilised with NaOCl and plated on potato dextrose agar (PDA). Coloured colonies of *Fusarium* sp. developed after 5 days at 25°C. Fungal structures and cultural characteristics were evaluated on PDA, carnation leaf agar, and distilled water agar (Booth, 1971; Nelson *et al.*, 1983). Based on conidia and chlamydospore development and on colony growth rate, the fungus was identified as *F. sambucinum* Fuckel *sensu stricto*. Two inoculation methods were used for pathogenicity tests carried out on 9 potted bougainvillea plants (3 wounded with a pin, 3 non-wounded and 3 uninoculated controls). The first method consisted in depositing PDA plugs colonised by *F. sambucinum* on wounded and non wounded branches and twigs. With the second method plants were sprayed with a conidial suspension (3×10⁶ spores/ml). In both cases, plants were placed in a moist chamber for 4 h at 18-24°C. Symptoms were observed between 15 and 20 days after inoculation. The largest cankers developed in plants inoculated with the first method, wilts with both procedures, the leaves close to infected nodes desiccated and fell. *F. sambucinum* was reisolated from infected tissues. *F. sambucinum* is known as a canker-causing organism (Booth, 1971) and was recently recorded from *Phytolacca dioica* in Argentina (Carranza *et al.*, 2004). To our knowledge, this is the first world report of *F. sambucinum* causing cankers on *B. glabra*.

Booth C. 1971. The Genus *Fusarium*. Commonwealth Mycological Institute, Kew, Surrey, UK.Carranza M., Lori G., Larran S., 2004. Canker on umbra tree caused by *Fusarium sambucinum*. *Plant Pathology* **53**: 532.Nelson P.E., Tousson T.A., Marasas W.F.O., 1983. *Fusarium* Species: An illustrated manual for identification. The Pennsylvania State University Press, University Park, PA, USA.Corresponding author: S.M. Wolcan
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DISEASE NOTE

**FIRST REPORT OF *OLIVE LATENT VIRUS*
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Olive (*Olea europea* L.) is one of the most economically relevant crops in Turkey. However, notwithstanding its importance, Turkey has no certification programme for olive and only a few studies of virus diseases of olive have been made using ELISA. *Olive latent virus 1* (OLV-1, genus *Necrovirus*, family *Tombusviridae*) induces symptomless infections and is transmitted by mechanical inoculation. The virus was first isolated in Turkey from citrus trees affected by chlorotic dwarf disease (Martelli *et al.*, 1996) but, to our knowledge, there are no records of OLV-1 in olive trees in Turkey. In this study, we tested 43 olive trees from Hatay province by RT-PCR. Total RNA was extracted from 0.1 g phloem tissue using RNeasy Plant Mini Kit (Qiagen, Valencia, CA, USA). Reverse transcription and PCR amplification, performed according to Faggioli *et al.* (2005), yielded amplicons of the expected size (*ca.* 299 bp) from two samples. The presence of OLV-1 was confirmed by sequencing each of the two amplicons spanning the capsid protein gene, and comparing the obtained sequences with those of OLV-1 in databases (Accession No. NC-001721, DQ083996 and AB061815). The two Turkish olive sequences were 99% identical to the citrus isolate sequence (NC-001721), 94% or 95% identical to the tulip isolate sequence (AB061815) and 93% identical to the olive isolate sequence (DQ083996). Homology was thus correlated more to the geographical origin than the host.

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DISEASE NOTE

**ENDOPHYTIC OCCURRENCE
OF A PATHOGENIC STRAIN
OF *FUSARIUM RETICULATUM*
IN ENGLISH OAK IN ITALY**

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In autumn 2002, the uppermost leaf and bud, the stem, collar, main root and a randomly chosen lateral root were excised from 20 symptomless 3-year-old *Quercus robur* L. seedlings growing in a severely declining Italian oak forest at Cessalto (northern Italy). All samples were surface-sterilized with 0.5 % sodium hypochlorite, thoroughly rinsed with sterile water and tissue fragments from each of them were plated on potato dextrose agar (PDA) according to Montecchio (2005). *Fusarium reticulatum* Mont (Nelson *et al.*, 1983) was isolated with other fungi from all parts of 6 of the 20 assayed plants. Ten symptomless 3-year-old container-grown English oaks seedlings were inoculated with a randomly chosen isolate of *F. reticulatum* (now deposited as 115133, CBS, Utrecht, The Netherlands) by gently scraping with a sterile scalpel the surface-sterilized epidermis surrounding the collar. After covering the rest of the plant with a plastic sheet, the wound was sprayed with a conidial suspension containing 10^3 macroconidia/cm³ in sterile water, and sealed with Parafilm. Ten control seedlings were wounded as above and sprayed with sterile water. All seedlings were placed in a greenhouse at 20±2°C, 80% relative humidity, and 12 h photoperiod. After 150 days, all inoculated plants showed wilted and desiccated leaves, and brownish longitudinal stripes on the outer bark, originating from the inoculation site. Stem sections 2 cm above the inoculation point showed dark brown necroses on about 70% of the area. Abundant mycelium was present in vessels and parenchyma tissue from which *F. reticulatum* was reisolated. Control plants remained symptomless, the wound healed, and isolations were unsuccessful. To our knowledge, this is the first report of *F. reticulatum* in the genus *Quercus*. Investigations on the involvement of environmental stresses in the pathogenic behaviour of this fungus and on its possible role in the expression of the oak decline syndrome, which includes seedling dieback, are in progress.

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DISEASE NOTE

**FIRST REPORT OF *STRAWBERRY
LATENT RINGSPOT VIRUS*
IN LEBANESE CHERRY ORCHARDS**

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During summer 2006, when several stone fruit tree orchards in Lebanon were surveyed to select mother plants for a certification programme, a stunted cherry tree of the local cv. Benni with deformed and mottled leaves was observed at Beskenta (Mount Lebanon). Leaf and shoot samples were collected and analysed by ELISA (Clark and Adams, 1977) using commercial kits for the detection of *Prunus necrotic ringspot virus* (PNRSV), *Prune dwarf virus* (PDV), *Apple mosaic virus*, *Apple chlorotic leaf spot virus*, *Tomato ringspot virus*, *Strawberry latent ringspot virus* (SLRSV), *Raspberry ringspot virus*, *Cherry leaf roll virus* and *Plum pox virus* (Loewe, Sauerlach, Germany). Buds from the diseased tree were grafted onto young *Prunus avium* indicators (cvs Bing and Sam) maintained in a greenhouse at 24-25°C, and leaf sap, extracted in 0.1 M phosphate buffer, pH 7 and 2.5% nicotine, was inoculated to herbaceous hosts. SLRSV and PDV were detected by ELISA in the diseased tree; PDV and PNRSV, alone or in combination, were found in other trees of the same or different cultivars from the same orchard. No SLRSV was found in more than 300 trees from other cherry-growing areas. Chlorosis of the main veins and strong deformations developed on the leaves of both cherry indicators one month after inoculation, and sap-inoculated *Chenopodium amaranticolor* and *C. quinoa* showed systemic mottling, vein clearing and leaf deformation. The presence of SLRSV in these symptomatic hosts was confirmed by ELISA and immunosorbent electron microscopy (Milne and Luisoni, 1977) tests. To our knowledge, this is the first report of infections by SLRSV in cherry and other stone fruits in Lebanon.

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DISEASE NOTE

FIRST REPORT AND CHARACTERIZATION OF PEAR DECLINE PHYTOPLASMA ON PEAR IN LEBANON

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In October 2005 and 2006, symptoms of pear decline were observed in commercial orchards of the pear cvs California and Coscia in west, central and north Bekaa valley (Lebanon). Affected plants showed premature reddening and upward rolling of the leaves which often had also down-turned petioles. In some cases, premature defoliation and reduced shoot growth were observed. DNA was extracted from 0.5g of leaf mid veins of 31 symptomatic and two symptomless pear plants according to the CTAB method (Maixner *et al.*, 1995). Nested PCR was used for phytoplasma detection with primers pairs fU5/rU3 (Seemüller and Schneider, 1994) after a first PCR with the universal phytoplasma primers pair P1/P7. Positive fU5/rU3 amplification was obtained from the majority of symptomatic samples (51%) but not from symptomless samples. The sequence of fU5/rU3 PCR products confirmed the detected phytoplasma to be a strain of *Candidatus Phytoplasma pyri*. For typing Lebanese isolates of *Ca. P. pyri*, non ribosomal specific PCR tests were made with primers pairs AcefF1/AcefR1 and the nested AcefF2/AcefR2 (Danet *et al.*, 2007) on three samples from Tanayl (central Bekaa, 2005 and 2006) and one sample from Terboul (central Bekaa, 2006). Sequence typing of the 797 pb *aceF* product amplified with AcefF2/AcefR2 showed that all Lebanese phytoplasma isolates had an identical *aceF* sequence presenting a single nucleotide polymorphism when compared with the *aceF* sequence of European strains of *Ca. P. pyri* in which no *aceF* polymorphism has been detected (Danet *et al.*, 2007). This was taken as evidence that the Lebanese *Ca. P. pyri* strain is genetically different from the European strains. To our knowledge, this is the first report of pear decline phytoplasma in Lebanon.

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DISEASE NOTE

FIRST REPORT OF LITTLE CHERRY VIRUS 1 IN CHERRY, PLUM, ALMOND AND PEACH IN ITALY

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Little cherry disease (LChD) is a widespread disorder of ornamental, sweet and sour cherries. In sensitive cultivars, it results in the production of small, pale-coloured fruits with reduced sugar content and in the premature reddening or bronzing of the leaves. Little cherry virus-1 (LChV-1) and *Little cherry virus 2* (LChV-2), both members of the family *Closteroviridae*, are associated with this disease, but often induce symptomless infection. Assays for both viruses were made during a survey in 2006 and 2007 of the sanitary status of fruit trees in Apulia (southern Italy). Samples were collected in different commercial orchards from 22 sweet cherry, 13 plum, five almond, five peach, and two apricot trees. Total nucleic acids (TNA) were extracted from the leaves as described by Foissac *et al.* (2001) and used as template for Superscript III one-step RT-PCR with Platinum *Taq* DNA polymerase (Invitrogen, Carlsbad, CA, USA) using primer sets specific for LChV-1 or LChV-2 (Rott and Jelkmann, 2001). Whereas all samples were negative for LChV-2, a 419 bp fragment corresponding to part of the 3' non-translated region of LChV-1 RNA was amplified from five cherry, four plum, one almond and one peach tree samples. These results were obtained in several independent experiments. Trees of both native and imported cultivars were infected but LChV-1 was not associated with any particular field symptoms. To our knowledge, this is the first report of LChV-1 in Italy and of its natural occurrence in plum, almond and peach.

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DISEASE NOTE

FIG MOSAIC IN CORSICA

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In Summer 2007, fig (*Ficus carica*) trees with mosaic symptoms were observed in the vicinity of Porto Vecchio (Corsica, France). Samples were collected from the quadrant of a plant showing strong chlorotic mottling of the leaves and analyzed for the presence of viruses by electron microscope observations of thin-sectioned leaf tissues and leaf dips and by RT-PCR assays using primers specific for Fig leaf mottle-associated virus 1 (FLMaV-1), or Fig leaf mottle-associated virus 2 (FLMaV-2) (Elbeaino *et al.*, 2006, 2007), or for an unnamed filamentous virus thought on molecular evidence to be a member of the family *Flexiviridae* (A. Minafra, unpublished information). Parenchyma cells of thin-sectioned tissues contained the double-membrane bodies (DMB, possibly particles of an unidentified enveloped virus) typically associated with fig mosaic disease (Martelli *et al.*, 1993), and bundles of filamentous particles. Particles of the same type in leaf dips were clearly decorated by an antiserum to the putative flexivirus. An amplicon of 380 bp was obtained in RT-PCR assays using primers specific for the coat protein gene of the putative flexivirus. No amplicon was obtained using primers specific for FLMaV-1 and FLMaV-2. The evidence suggests that the donor tree was infected by at least two viruses, which supports the idea that multiple infectious agents may be involved in the aetiology of fig mosaic. This seems to be the first record for Corsica of fig mosaic and its possible causal agents.

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DISEASE NOTE

PALEA BROWNING, A NEW DISEASE
OF RICE IN ITALY CAUSED
BY *PANTOEA ANANATIS*

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Spikelets discolouration of rice (*Oryza sativa*), observed in rice fields at Rosasco (Pavia, northern Italy), was diagnosed as palea browning. First symptoms appeared on the glumes as light-brown lesions, which, over time, darkened, coalesced and enlarged resulting in the uniform browning of the palea and, occasionally, of both lemma and palea. *Pantoea ananatis* was first reported as the causal agent of palea browning in Japan and, subsequently, in other Asian countries. In Italy, in 2005, disease incidence on early-sown rice cvs Selenio and Gladio, was 38% and 20%, respectively, whereas on the same late-sown varieties it dropped to 15% and less than 10%, respectively. Diseased samples of cv. Selenio were collected and analyzed to ascertain the presence of *P. ananatis*, using the semi-selective medium NSVC-In (Hasegawa *et al.*, 2003). All samples yielded colonies typical for *P. ananatis* after 3-day incubation at 28°C and, following purification, 40 putative strains of the bacterium were obtained. All strains, screened with API 20E (BioMérieux, Roma, Italy), proved to be *Pantoea* spp. DNA of each Italian strain and a known *P. ananatis* rice strain MAFF301720 was tested by PCR using Sn2b-As2c specific primers designed to amplify a genome fragment within the 16S-23S ITS region (R. Gitaitis, University of Georgia, Tifton, USA, personal communication) and by REP-PCR. Nineteen strains were identified as *P. ananatis*. Pathogenicity of two of these strains was tested in the field, by spraying panicles of five rice cultivars with a bacterial suspension (10^8 cfu ml⁻¹) from a 48h culture on TSBA. At harvest, the mean incidence of palea browning on inoculated panicles was ten times higher than the control. Rice varieties displayed different tolerance to the disease, cv. Carnaroli being the most tolerant. To our knowledge, this is the first report of palea browning induced by *P. ananatis* on rice in Italy. It adds to another bacterial disease of this crop elicited by *Acidovorax avenae* spp. *avenae* (Cortesi *et al.*, 2005).

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