

Disease Notes

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First Report of *Ralstonia (Pseudomonas) solanacearum* Infecting Pot Anthurium Production in Florida. D. J. Norman and J. M. F. Yuen, Department of Plant Pathology, University of Florida-IFAS, Central Florida Research and Education Center, 2807 Binion Rd., Apopka 32703, Florida Agric. Exp. Sta. Journal Series R-06699. *Plant Dis.* 83:300, 1999; published on-line as D-1998-1216-01N, 1998. Accepted for publication 15 December 1998.

Xanthomonas campestris pv. *dieffenbachiae* is a common pathogen of pot anthurium production in Florida. While *X. campestris* pv. *dieffenbachiae* was isolated from systematically infected plants with chlorotic, necrotic, and wilted leaves, a fluidal, beige bacteria was occasionally isolated on nutrient agar (Difco, Detroit, MI), as opposed to the common, yellow pigmented *Xanthomonas* sp. Distinction in the symptomology of plants systematically infected with a *Xanthomonas* sp. or this new bacterium could not be made. Three isolates were obtained of this unidentified bacterium from leaves and stems of three separate plants. With FAME (fatty acid methyl esters) analysis, using MIDI (Microbial Identification System, software version TSBA 3.90 [Newark, DE]), these isolates were classified as *Ralstonia (Pseudomonas) solanacearum* (syn. *Burkholderia solanacearum*) with a mean similarity indice of 0.895. Isolates were found to be gram negative, oxidase negative, catalase positive, motile, strictly aerobic, and metabolically classified as biovar 1; they accumulated poly- β -hydroxybutyrate and produced a hypersensitive response on tobacco within 24 h. A characteristic fluidal, white growth with a distinctive, red, swirling, egg-shaped, pigmentation pattern was observed on triphenyltetrazolium chloride medium. Further confirmation of identity as *R. solanacearum* was obtained by polymerase chain reaction amplification and electrophoretic analysis with species-specific primers (2), which in all cases produced a 148-bp product along with control strains. The three isolates were inoculated onto three plants of anthurium, tomato, triploid banana, and pothos. Inoculations were done at least twice; plants were inoculated either by stabbing the plant stems with a needle dipped in a suspension of bacteria or by applying 10 ml of a 1×10^8 CFU/ml suspension to the soil of the test plants. Chlorosis, necrosis, and wilt symptoms appeared within 2 weeks on all plant species tested. Recently, pothos (*Epipremnum aureum*) cuttings imported to Florida from Costa Rica have been implicated as a source of *R. solanacearum* (1). Imported cuttings of pothos were being grown in hanging baskets over the infected anthuriums. Although no *R. solanacearum* infections were detected in the pothos, these imported plants are the probable source of the initial inoculum for this disease outbreak on anthuriums.

References: (1) D. J. Norman and J. M. F. Yuen. *Phytopathology* 87:S70, 1997. (2) S. E. Seal et al. *Appl. Environ. Microbiol.* 58:3751, 1992.

First Report of Sweetpotato Scurf Caused by *Monilochaetes infuscans* in Italy. P. Di Lenna and N. Mascarello, Istituto di Patologia Vegetale, Università di Padova, Strada Romea 16, Legnaro (PD), Italy; E. Xodo, O.M.P., Lungargine Capuleti, 11, Verona, Italy. *Plant Dis.* 83:300, 1999; published on-line as D-1998-1217-01N, 1998. Accepted for publication 3 December 1998.

In the last few years, in the Treviso area, the incidence and severity of a sweetpotato (*Ipomoea batata*) disease has increased to become a serious economic threat to this crop. The symptoms are small, dark brown to black spots on the surface of fleshy storage roots that under high relative humidity coalesce to completely cover the root surface. Affected roots may also develop small cracks and shrink in storage. In the past, the same symptoms were observed, but the low incidence of the disease did not cause significant economic losses, and therefore it was not considered worthy of investigation. In August 1998, an investigation was initiated to determine the causal agent of the disease and to provide growers with effective control strategies. Since the symptoms were similar to those

incited by *Monilochaetes infuscans*, agent of sweetpotato scurf (1), samples of roots were collected and examined for the presence of the pathogen. Even though it is difficult to isolate, *M. infuscans* was consistently present in infected tissues. In pure culture on potato dextrose agar the pathogen formed typical, raised, domelike, dark gray to black colonies. Conidiophores were brown to black, unbranched, and support straight or curled chains of hyaline conidia. Traditional cultivation methods that ignore sanitary conditions probably caused the gradual increase in the incidence and severity of the disease. Sweetpotato scurf is common in the U.S., Japan, and some Pacific islands, but it has not been previously recorded in Italy.

Reference: (1) C. A. Clark and J. W. Moyer. 1988. *Compendium of Sweet Potato Diseases*. American Phytopathological Society, St. Paul, MN.

First Report of Black Sigatoka in Florida. R. C. Ploetz, University of Florida, IFAS, Tropical Research and Education Center, 18905 SW 280th Street, Homestead 33031-3314; and X. Mourichon, CIRAD/FLHOR, Laboratoire de Phytopathologie Vegetale, 2477, avenue du Val de Montferriand, B.P. 5035, 34032 Montpellier Cedex 1, France. *Plant Dis.* 83:300, 1999; published on-line as D-1998-1217-03N, 1998. Accepted for publication 15 December 1998.

Black Sigatoka, caused by *Mycosphaerella fijiensis*, is widely recognized as the most important disease of banana, *Musa* spp. It has spread rapidly in the Western Hemisphere since it first appeared in Honduras in 1972, and is now found in the Caribbean basin in Cuba, Jamaica, and the Dominican Republic, and on the mainland from central Mexico south to Bolivia and northwestern Brazil (2). In October 1998, symptoms of black Sigatoka (2) were observed on several different cultivars in a collection at the University of Florida's Tropical Research and Education Center (TREC) in Homestead (25°30' N, 80°30' W). During preliminary surveys, the disease was found at four of eight locations in a 15 km² area to the north of TREC. Disease severity, rated as the youngest leaf spotted (YLS), averaged 4.8 on the most susceptible cultivar, Rajapuri, at one of the locations. The extent and history of damage at this site indicated that black Sigatoka had been there for at least 3 to 4 years. The prevailing east to west winds in the Caribbean, and highly variable incidence and severity of the disease also suggested that the pathogen had been introduced to the area on infected seed pieces (suckers) rather than by wind or rain-blown ascospores from Cuba or other affected areas (1). The presence of the disease was confirmed after the following characteristics of the pathogen's anamorph, *Paracercospora fijiensis*, were observed on affected leaves: simple conidiophores occurring singly or in groups of two to six with one to several septa, scars, and usually a broadened base; and conidia much more abundant on lower leaf surfaces, straight to variously bent with one to several septa and a conspicuous scar at the base. Single-ascospore cultures were recovered from Rajapuri and are stored at CIRAD/FLHOR in Montpellier. This is the first time black Sigatoka has been reported in the continental United States. Banana is a minor but significant tropical fruit crop in southern Florida, with fruit valued at over \$2.5 million per annum. Production from Hua moa, Silk, and other important cultivars will probably be affected as the disease becomes established in this part of the state.

References: (1) R. H. Stover. *Plant Dis.* 64:750, 1980. (2) J. C. Tejerina et al. *Plant Dis.* 81:1332, 1997.

(Disease Notes continued on next page)

First Report of Aerial Blight Caused by *Pythium myriotylum* on Tomato in Florida. P. D. Roberts and R. Urs, University of Florida, Southwest Florida Research and Education Center, Immokalee 34142; and R. J. McGovern, University of Florida, Gulf Coast Research and Education Center, Bradenton 34203. Florida Agricultural Experiment Station, Journal Series No. R-06525. Plant Dis. 83:301, 1999; published on-line as D-1998-1217-02N, 1998. Accepted for publication 14 December 1998.

In September 1996 and 1997, diseased tomato seedlings were observed with symptoms of an aerial watery rot on leaves, petioles, and stems. Tomato cvs. Sanibel and 10097 from commercial fields in southwest Florida (Collier and Lee counties) and west central Florida (Manatee County) exhibited similar symptoms that occurred at an incidence of 15 to 18% about 4 weeks after transplanting and resulted in plant death. Microscopic examination of symptomatic tissue revealed the presence of mycelium and oogonia typical of *Pythium* spp. A fungus was consistently isolated from four plants sampled from each site onto a medium selective for *Pythium* spp. and maintained in pure culture on V8 juice agar at 28°C. The isolates were identified as *Pythium myriotylum* Drechs. based on the following morphological data: lobate sporangium, 12 to 13 µm wide; vesicle 15.4 to 19.4 µm in diameter; exit tube 54 to 90 µm long, oogonium 23 to 30 µm in diameter; and oospore 21 to 26 µm in diameter (1,2). Pathogenicity tests were conducted with two isolates from diverse regions within Florida by spray inoculating the leaves and shoots of 6- to 8-week-old tomato seedlings with a sporangial suspension of 1×10^4 sporangia per ml. Noninoculated plants served as controls. Plants had 24 h of pre- and post-dark period, day/night temperatures of 28/21°C, a 14-h photoperiod, and near 100% relative humidity in a growth chamber. The foliage of inoculated tomato plants exhibited symptoms identical to those observed in field samples 24 h after inoculation and 100% mortality within 72 h. The reisolated fungus was morphologically identical to the original isolate. Noninoculated plants remained asymptomatic. The unusual rainfall recorded at some sites, such as in Manatee County in September 1997, was 36% higher than the 40-year average and may have contributed to the incidence of this previously undescribed foliar blight.

References: (1) Anonymous. C.M.I. Descriptions of Pathogenic Fungi and Bacteria No. 118. (2) T. Watanabe. Pictorial Atlas of Soil Fungi. Lewis Pub., London. p. 71.

First Report of Tomato Bushy Stunt Virus Isolated from Lettuce. H.-Y. Liu, J. L. Sears, C. Obermeier, G. C. Wisler, E. J. Ryder, and J. E. Duffus. USDA-ARS, 1636 East Alisal Street, Salinas, CA 93905; and S. T. Koike, University of California Cooperative Extension, Salinas 93901. Plant Dis. 83:301, 1999; published on-line as D-1999-0104-01N, 1999. Accepted for publication 17 December 1998.

In recent years a disease causing dieback and necrosis of Romaine and leaf lettuce has become increasingly important in California and incidence is becoming more widespread. This disease has been primarily found in areas where soil has been dredged from a river or in flooded land. Tomato bushy stunt virus (TBSV) isolates have been isolated from roots and leaves of symptomatic lettuce. The particles are isometric with a diameter of 30 nm. Double-stranded RNA (dsRNA) profiles are identical to the tomato and *Prunus* isolates of TBSV. However, spurs are formed in agar double diffusion tests when antisera to the tomato and *Prunus* isolates were used. A similar dieback disease of lettuce was observed in several counties of California during the mid-1980s. Symptoms of this disease are very similar to those described for the "brown blight" disease of lettuce reported in the 1920s (1), including severe stunting of plants and extensive chlorosis, mottling, and necrosis of older leaves. Plants infected early in their development may die. Although inoculation under greenhouse conditions has not reproduced the dieback disease in lettuce, TBSV has been consistently isolated from field-grown, symptomatic lettuce. The question of whether this new dieback disease of lettuce is caused only by lettuce isolates of TBSV or if some other viruses are also involved needs further studies.

Reference: (1) I. C. Jagger. Phytopathology 30:53, 1940.

Occurrence of *Colletotrichum acutatum*, Causal Organism of Strawberry Anthracnose in Southwestern Spain. B. de los Santos G^o de Paredes and F. Romero Muñoz, C.I.F.A. "Las Torres-Tomejil," Apartado

Oficial, Alcalá del Río - 41200, Sevilla, Spain. Plant Dis. 83:301, 1999; published on-line as D-1998-1223-01N, 1998. Accepted for publication 16 December 1998.

A wilting disease of strawberry (*Fragaria × ananassa* Duchesne 'Oso Grande') was observed in production fields in Huelva, southwestern Andalucía, Spain. Crowns of wilted plants developed a reddish brown, firm rot. Longitudinal sections of roots of diseased plants showed a black coloration of the cortex. Symptoms also included blighting of flowers due to floral infection and rotting of green and ripening fruits. On the fruit, the round, firm sunken lesions were covered with masses of salmon-colored spores. Lesions on the leaves were round, ranged in diameter from 5 to 10 mm, and were light brown with purple edges. Infected tissues were surface disinfected in 20% sodium hypochlorite, blotted dry, and plated on water agar. Dishes were incubated at 25°C; hyphal tips made of fungi growing from lesions were transferred to potato dextrose agar (PDA) and incubated under cool-white fluorescent light at 25°C. One fungus was isolated consistently from lesions on different tissues. It was identified as *Colletotrichum acutatum* by morphological characteristics (2) and enzyme-linked immunosorbent assay (1). On PDA, the mycelium of the fungus was white to gray and covered with salmon spore masses. Colony diameters on PDA averaged 45 mm after 7 days at 25°C. Hyaline, aseptate conidia were cylindrical, fusiform, and intermediate in shape and averaged 12.8×4.2 µm in size. The fungus produced setae on infected fruit. Inoculations of strawberry cvs. Oso Grande and Chandler with conidial suspensions (10^6 conidia per ml) of *C. acutatum* produced lesions on the fruit, petioles, and crowns, and caused wilt. The pathogen was reisolated from lesions on the inoculated plants. This is the first report of *C. acutatum* causing strawberry anthracnose in Huelva.

References: (1) T. A. Cooke et al. EPPO Bulletin 25:57, 1995. (2) B. C. Sutton. The Coelomycetes. CMI, Kew, England, 1980.

First Report of Grapevine Downy Mildew (*Plasmopara viticola*) in Commercial Viticulture in Western Australia. S. J. McKirdy, I. T. Riley, and I. J. Cameron, AgWA, Western Australia 6983; and P. A. Magarey, SARDI, South Australia 5333. Plant Dis. 83:301, 1999; published on-line as D-1998-1221-01N, 1998. Accepted for publication 14 December 1998.

Despite the suitability of climate, Western Australia was one of the few grape (*Vitis vinifera* L.) growing areas free of grapevine downy mildew (*Plasmopara viticola* (Berk. & M. A. Curtis) Berl. & De Toni in Sacc.). Area freedom had been maintained by restricting the movement of host material and machinery from outside the state and fungicide use in Western Australia vineyards had been considerably less. *P. viticola* was detected in 1997 in 14 of 15 vines growing at Kalumburu, a remote community in the semi-arid tropics of Western Australia, and was eradicated. In October 1998, grape leaves with oilspots typical of downy mildew were received from a grower in the Swan Valley near Perth, one of the main production areas of Western Australia. Sporangia were hyaline and ellipsoid (14×11 µm), were borne on treelike sporangiophores, and were consistent with those described for *P. viticola* (1). This is the first record of *P. viticola* in commercial viticulture in Western Australia. A response plan for exotic diseases was activated and after 2 weeks of surveillance the disease was found in 45 of 70 vineyards surveyed of the 280 vineyards in the Swan Valley. Given the extent of spread, eradication of downy mildew was not considered possible. Weather data for August to October 1998 indicated the likelihood of several infection periods from budburst to flowering when the disease was first detected. Crop loss will be considerable in many vineyards. *P. viticola* was also found in bench-grafted cuttings in pots in leaf consigned from the Swan Valley to several other areas in August 1998. Downy mildew was found in other areas only in association with these consigned vines. An industry code of practice, including hygiene, is being developed to slow the rate of spread of *P. viticola* in Western Australia.

Reference: (1) Anon. C.M.I. Descriptions of Pathogenic Fungi and Bacteria No. 980, 1989.

(Disease Notes continued on next page)

Disease Notes (continued)

First Report of Powdery Mildew of Pepper in North-Central Mexico. R. Velásquez-Valle, INIFAP-Zacatecas, Apdo. Postal #18 Calera, Zacatecas, México, CP 98500 and P. Valle-García, Centro de Ciencias Agropecuarias-UAA, Ave. Universidad 940, Aguascalientes, México, CP 20100. Plant Dis. 83:302, 1999; published on-line as D-1999-0104-02N, 1999. Accepted for publication 3 December 1998.

Pepper plants showing general yellowing and white spots on the lower surface as well as brown spots on the upper surface of the lowest leaves were observed during the 1998 crop season in the states of Chihuahua, Zacatecas, and Aguascalientes (north central area of México). Light defoliation was observed. Samples were collected in Chihuahua and Zacatecas to identify the pathogen, which showed navicular conidia. A conidial suspension was sprayed on pepper varieties, but only Hungarian Yellow Wax showed powdery mildew lesions. The average size of the conidia from Zacatecas was $61.2 \times 18.2 \mu\text{m}$ while those from Chihuahua measured $58.2 \times 17.6 \mu\text{m}$, which is coincident with the description given for an *Oidiopsis* sp. The perfect stage is *Leveillula taurica* (1). Cleistothecia were not observed. In Zacatecas the disease was not detected in tomato fields or in sowthistle (*Sonchus oleraceae*) plants contiguous to, or within, infected pepper fields. Powdery mildew incidence ranged from 35 to 80% in Ancho type and from 0 to 55% in Mirasol type pepper fields in Zacatecas. The number of powdery mildew lesions per leaf in most cases varied from 1 to 3. Incidence was higher in fields where the canopy was closed and warmer and wetter conditions may have allowed the disease to progress.

Reference: (1) J. B. Jones et al. 1991. Compendium of Tomato Diseases. American Phytopathological Society, St. Paul, MN.

A New Virus on Maize in Nigeria: Maize Mild Mottle Virus. G. Thottappilly, International Institute of Tropical Agriculture (IITA), Ibadan, Nigeria; W. P. Qiu, J. S. Batten, J. N. Hughes, and K.-B. G. Scholthof, Department of Plant Pathology and Microbiology, Texas A&M University, College Station 77843. Plant Dis. 83:302, 1999; published on-line as D-1999-0104-03N, 1999. Accepted for publication 31 December 1998.

Maize (*Zea mays*) and itch grass (*Rottboellia cochinchinensis*) plants exhibiting a mild mosaic or mottle were collected from a farmer's field near Mokwa, Nigeria, in 1993. Icosahedral virions (approximately 28 to 30 nm) were purified from symptomatic tissue by differential centrifugation in 0.1 M phosphate buffer, pH 7.0. The virions are composed of one single-stranded positive-sense RNA of approximately 4,000 nucleotides and, as estimated by 12.5% sodium dodecyl sulfate-polyacrylamide gel electrophoresis (SDS-PAGE), a capsid protein of approximately 28 kDa. The virus was readily detected in infected plants by enzyme-linked immunosorbent assay and immunoblot assays with a polyclonal rabbit antibody derived from purified virions. A partial cDNA library was generated with random primers. Sequence analyses of a cDNA clone representing a portion of the putative replicase gene aligned most closely with related sequences of viruses within the Tombusviridae. In particular, a region of 78 predicted amino acids surrounding the "GDD" replicase motif shares 73% identity with panicum mosaic virus and 61% identity with maize chlorotic mottle virus. The virus is readily transmitted by mechanical inoculation to sweet and dent corn, millet, and wheat. Currently it is not considered of economic importance in Nigeria. The data suggest that "maize mild mottle virus" is a newly identified virus infecting maize.

First Report of *Sclerotinia sclerotiorum* on Common Ragweed (*Ambrosia artemisiifolia*) in Europe. Gy. Bohár and L. Kiss, Plant Protection Institute of the Hungarian Academy of Sciences, P.O. Box 102, Budapest, H-1525. Plant Dis. 83:302, 1999; published on-line as D-1999-0106-01N, 1999. Accepted for publication 4 January 1999.

Common ragweed (*Ambrosia artemisiifolia* L.) is reported as a host of *Sclerotinia sclerotiorum* (Lib.) de Bary in North America (2,4), but not in Europe. A Hungarian survey of fungal diseases of ragweed in 1994 did not find sclerotinia rot of common ragweed (*A. artemisiifolia* var. *elatior* (L.) Descourt.) (1). In autumn 1998, mature ragweed plants, 1 to 1.5 m tall, were collected from the borders of four sunflower (*Helianthus annuus* L.) fields in which sclerotinia rot of sunflower was frequently observed during the season, and also from six other roadside sites in Hun-

gary. Ragweed plants exhibiting symptoms characteristic of sclerotinia rot, i.e., wilting foliage and light brown, dry lesions on the stems, were found only near two sunflower fields. Black, round to irregular or oblong sclerotia were also observed on the infected ragweed plants both externally on the stem lesions and internally, in the pith cavity. Sclerotia measured up to 5 mm in diameter and were 5 to 14 mm long. After isolation on potato dextrose agar, the pathogen produced abundant aerial mycelium and large sclerotia characteristic of *S. sclerotiorum*. To confirm pathogenicity, potted seedlings and mature plants of ragweed were inoculated in the greenhouse with autoclaved wheat grains colonized with mycelia of *S. sclerotiorum* placed 0.5 to 1 cm from the collar of the test plants. Seedlings were killed in 2 to 3 days while mature plants wilted after 5 to 6 days. In a field test, six mature plants were inoculated by attaching mycelial disks to their stems with Parafilm. These plants wilted 12 to 14 days after inoculation. The pathogen was reisolated from all diseased plants. This is the first report of *S. sclerotiorum* on common ragweed in Europe. Nonsclerotial mutants of the fungus (3) are being produced to be tested as potential biocontrol agents of common ragweed, which has become not only the most widespread, but also the most important allergenic plant species in Hungary since the early 1990s.

References: (1) Gy. Bohár and L. Vajna. Növényvédelem 32:527, 1996. (2) G. J. Boland and R. Hall. Can. J. Plant Pathol. 16:93, 1994. (3) G. J. Boland and E. A. Smith. Phytopathology 81:766, 1991. (4) D. F. Farr et al. 1989. Fungi on Plants and Plant Products in the United States. American Phytopathological Society, St. Paul, MN.

Occurrence of Witches'-Broom, a New Phytoplasma Disease of Acid Lime (*Citrus aurantifolia*) in India. D. K. Ghosh, A. K. Das, and Shyam Singh, National Research Centre for Citrus, P. B. No. 464, Shankarnagar P.O., Nagpur - 440010; S. J. Singh, Indian Institute of Horticultural Research, Bangalore 560089; and Y. S. Ahlawat, Indian Agricultural Research Institute, New Delhi - 110012. Plant Dis. 83:302, 1999; published on-line as D-1999-0111-01N, 1999. Accepted for publication 23 December 1998.

In India, acid lime (*Citrus aurantifolia* (L.) Swingle) is one of the most important citrus fruits grown. It constitutes nearly 20% of the total citrus production. During 1995, an unusual type of disease was observed on a 6-year-old acid lime plant in an orchard in the Nagpur District in eastern Maharashtra. It was named witches'-broom disease (WBD) to reflect the most conspicuous symptom. Other symptoms included small chlorotic leaves, highly proliferated shoots, and shortened internodes. Leaves dropped prematurely and infected twigs were distorted. In advanced stages, infected branches had dieback symptoms. WBD of lime has been reported from Oman and UAE (1) and the causal phytoplasma was designated "*Candidatus* Phytoplasma aurantifolia" (2). Subsequent surveys in 1995-1998 revealed disease incidences as high as 5% in Maharashtra and in other major acid-lime-growing states—Andhra Pradesh, Tamilnadu, and Karnataka. After the grafting of infected acid lime shoots, disease symptoms developed on Troyer citrange, rough lemon, and Rangpur lime, but not on sweet orange (mosambi), mandarin (Nagpur), or trifoliolate orange. The WBD agent was transmitted from infected acid lime to periwinkle (*Catharanthus roseus*) plants and vice versa by dodder (*Cuscuta reflexa*). Ultrathin sections of leaf midrib of infected acid lime plants were fixed on copper grids, stained with uranyl acetate and lead acetate, and examined in a JEM 100S transmission electron microscope. Numerous bodies having the characteristic morphology of phytoplasmas were observed in phloem sieve tubes of acid lime in diseased but not in healthy leaves. The phytoplasmal bodies ranged from 100 to 800 nm in diameter and were bounded by a poorly defined membrane. Freehand transverse sections of young internode regions of a WBD-infected periwinkle plant were stained in DAPI (4', 6 diamidino-2-phenylindole; 1.0 $\mu\text{g/ml}$) and were observed with a fluorescent microscope (Leica). An intense bluish-white fluorescence in the phloem elements of diseased periwinkle and its absence in healthy samples were consistent with the presence of phytoplasmas. This is the first report of phytoplasma-induced witches'-broom disease of acid lime in India.

References: (1) M. Garnier et al. Plant Dis. 75:546, 1991. (2) L. Zreik et al. Int. J. Syst. Bacteriol. 45:449, 1995.

(Disease Notes continued on next page)

First Report of Pea Enation Mosaic Virus Affecting Lentil (*Lens culinaris*) in Syria. Khaled M. Makkouk and Safaa G. Kumari, Virology Laboratory, Germplasm Program, International Center for Agricultural Research in the Dry Areas (ICARDA), P.O. Box 5466, Aleppo, Syria; and Bassam Bayaa, Faculty of Agriculture, University of Aleppo, Syria. *Plant Dis.* 83:303, 1999; published on-line as D-1999-0107-01N, 1999. Accepted for publication 5 January 1999.

Symptoms suggestive of virus infection in lentil (*Lens culinaris* Medik.) fields in Dara'a in southern Syria have been observed, in epidemic proportions, almost annually since 1994. A similar epidemic was observed on many lentil genotypes at the ICARDA farm, near Aleppo, as well as in other locations in northern Syria during 1998. Symptoms included growth reduction and rolling of leaves, accompanied by mottling with tip wilting or necrosis. Field symptoms were reproduced on lentil cv. Syrian Local upon mechanical inoculation of plants with inoculum from symptomatic field plants. Transmission tests showed that the disease agent can be transmitted from lentil to lentil, pea (*Pisum sativum* L.), and faba bean (*Vicia faba* L.) plants by the pea aphid (*Acyrtosiphon pisum* Harris) in a persistent manner. More than 500 symptomatic lentil plants were collected and tested for the presence of 14 different viruses by the tissue-blot immunoassay (TBIA) (2). Around 80% of the samples reacted only with antiserum to pea enation mosaic virus (PEMV), a Dutch isolate (E1540) provided by L. Bos, Wageningen, The Netherlands (1). Surveys conducted during the 1997/1998 growing season showed that PEMV was widely distributed in the major lentil-growing areas of Syria: some lentil fields had more than 50% virus incidence. This is the first record of PEMV naturally infecting lentil in Syria.

References: (1) K. Mahmood and D. Peters. *Neth. J. Plant Pathol.* 79:138, 1973. (2) K. M. Makkouk and A. Comeau. *Eur. J. Plant Pathol.* 100:71, 1994.

First Report of Tomato Yellow Leaf Curl Virus in Réunion Island.

M. Peterschmitt and M. Granier, CIRAD, BP 5035, 34032 Montpellier Cedex 1, France; R. Mekdoud, SPV, 2 route Ligne Paradis, 97410 Saint Pierre, Réunion, France; A. Dalmon, LNPV, Domaine Saint Maurice, BP 94, 84143 Montfavet Cedex, France; O. Gambin, FDGDEC, 16 rue Millius, 97400 Saint Denis, Réunion, France; J. F. Vayssieres and B. Reynaud, CIRAD, 7 chemin de l'IRAT, 97410 Saint Pierre, Réunion, France. *Plant Dis.* 83:303, 1999; published on-line as D-1999-0108-01N, 1999. Accepted for publication 4 January 1999.

In September 1997, stunting, reduced leaf size, leaf curling, and yellow margins were observed on tomato plants on a farm on the south coast of Réunion, a French island belonging to the Mascarenes archipelago. To our knowledge, these symptoms appeared to be characteristic of a tomato yellow leaf curl virus (TYLCV) infection. Diseased plants gave positive reactions with a triple antibody sandwich-enzyme-linked immunosorbent assay (TAS-ELISA), using ADGEN antibodies specific for begomoviruses (1). The serological results were confirmed by polymerase chain reaction (PCR) with a pair of degenerate primers—MP16, 5'-CCTCTAGATAATATTAC(C/T)(G/T)(G/A)(A/T)(T/G)G(G/A)CC-3' and MP82, 5'-CGGAATTC(T/C)TGNAC(C/T)TT(G/A)CANGNCC(T/C)T C(G/A)CA-3'—designed by Malla Padidam (ILTAB, San Diego, CA) to amplify a region of the A component of begomoviruses, between the intergenic conserved nonanucleotide sequence (TAATATTAC) and the first 5' quarter of the capsid protein gene. A 500-bp PCR product was obtained from a symptomatic plant but not from a healthy looking one. After cloning the PCR product in a pGEM-T Easy vector (Promega, Madison, WI) and sequencing it with plasmid-specific primers (SP6, T7), the sequence was compared with the sequences of the NCBI data base, with the use of BLAST. Nineteen sequences among those producing the highest scoring segment pairs were compared with each other and with the 500-bp PCR product from Réunion by the Clustal method of MegAlign (DNASTAR, London). The Réunion sequence (AJ010790) was at least 94% similar to sequences of TYLCV isolates from the Dominican Republic (AF024715), Cuba (AJ223505), and Israel (X15656, X76319 for the mild clone). Based on these results, it appeared that the analyzed tomato plant was infected by a geminivirus isolate belonging to the Israeli

species of TYLCV. A preliminary survey was carried out from December 1997 to April 1998 in both outdoor and protected tomato crops. Infected plants were detected by TAS-ELISA in 52 of the 123 locations visited. Severe economic losses were observed: 14 locations with 60 to 100% yield reduction and 11 locations with 40 to 60% yield reduction. All the infected samples were collected in the leeward coast, which is the driest region of the island. Although *Bemisia tabaci* (Gennadius) has been recorded since 1938 in Réunion (2), it has been observed on tomato crops only since 1997 and population levels were low compared with those of *Trialeurodes vaporariorum* Westwood. During the first six months of 1998, *B. tabaci* was found on *Euphorbia heterophylla* L., *Lantana camara* L., *Solanum melongena* L., *S. nigrum* L., and *Phaseolus vulgaris* L. These host plants often occur near infected tomato crops.

References: (1) S. Macintosh et al. *Ann. Appl. Biol.* 121:297, 1992. (2) L. Russell and J. Etienne. *Proc. Entomol. Soc. Wash.* 87:202, 1985.

First Report of Sinaloa Tomato Leaf Curl Geminivirus in Costa Rica.

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In October 1998, geminivirus-like symptoms were widespread in tomato plantings near Turrialba, Costa Rica. Isolates from several fields were experimentally transmitted to tomato seedlings with whiteflies from a *Bemisia tabaci* (Genn.) colony maintained at CATIE, which resulted in interveinal chlorosis and leaf curling symptoms indistinguishable from those observed in the field. Total DNA was extracted from leaves of 16 of these experimentally inoculated plants and assayed by polymerase chain reaction (PCR) for the presence of begomovirus DNA with the degenerate primers AV324 and AC889 (2) to amplify the core region of the coat protein gene (core Cp). PCR yielded the expected size core Cp fragment (576 bp) from 16 of 16 samples. The core Cp fragments of six samples were cloned and sequenced. A comparison of the core Cp sequences with reference begomovirus sequences indicated all Costa Rican isolates were >95% identical to Sinaloa tomato leaf curl geminivirus reported in 1994 from Sinaloa, Mexico (STLCV-SINALOA). Virus identity was confirmed by multiple sequence alignments of the viral coat protein gene (Cp) and the common region (CR) sequences of A and B components (CR-A and CR-B), respectively, with analogous reference begomovirus sequences. Cp and CRs were obtained by PCR, and amplicons were cloned and sequenced as described (1). The Cp open reading frame (ORF; 756 nucleotides) (AF110515) identified within the A component amplicon shared 92.9% sequence identity with STLCV-SINALOA Cp (AF040635). The CR sequences of the A (AF110516) and B (AF110517) components (163 nucleotides) shared 98.2% sequence identity with each other, suggesting that they were amplified from the cognate A and B components of the same virus. Further, the CR-A and CR-B components contained the same putative Rep binding site, TGGGGT-AA-TGGGGT, which was also identical to that of STLCV-SINALOA. The mean percent divergences between viral Cp and CR amplicons ($n = 6+$) ranged from 98 to 100%. Collectively, STLCV-like symptoms in tomato, >92% identity between viral Cp sequences, and identical CR iterons indicate that the Costa Rican tomato virus is STLCV, or a closely related strain. This is the first report of an STLCV-like begomovirus in tomato in Costa Rica (STLCV-CR).

References: (1) A. M. Idris and J. K. Brown. *Phytopathology* 88:648, 1998. (2) S. D. Wyatt and J. K. Brown. *Phytopathology* 86:1288, 1996.

(Disease Notes continued on next page)

Disease Notes (continued)

First Report of the Presence of a Phytoplasma in Tomato in Mauritius. A. Dookun, S. Aljanabi, S. Saumtally, and L. J. C. Autrey. Mauritius Sugar Industry Research Institute, Reduit, Mauritius. *Plant Dis.* 83:304, 1999; published on-line as D-1998-0115-01N, 1998. Accepted for publication 29 December 1998.

In recent years, tomato plants (*Lycopersicon esculentum* Mill., cultivar MST 32/1) grown in Mauritius were observed with symptoms characteristic of a phytoplasmal infection. Young plants exhibited stunted growth, with a bunched top symptom on one or more stems. The leaves were small with a curled margin, sometimes were purplish, and were clustered as a rosette. On older plants, other leaf symptoms, such as necrotic vein, mosaic, and vein clearing, that were typical of viral infection were observed. In March to June 1998, research was initiated to determine whether a phytoplasma or a virus was associated with the problem. Virus screening was carried out by double antibody sandwich enzyme-linked immunosorbent assay (DAS-ELISA) with the following antisera: tomato mosaic virus (TMV), tomato leaf curl virus (TLCV), tomato spotted wilt virus (TSWV), cucumber mosaic virus (CMV), potato virus X (PVX), potato virus Y (PVY), and potato leaf roll virus (PLRV). The presence of a phytoplasma was assayed by the polymerase chain reaction (PCR) with universal primers that amplify the 16S rDNA sequences. Oligonucleotide pairs RU3 and FU5 (1) and P1 and P6 (2) were evaluated separately. Template DNA was prepared from young leaves with the Nucleon Phyta-Pure Plant DNA extraction kit (catalog no. RPN 8511, Amersham Life Science, Buckinghamshire, U.K.). Amplification of a 880-bp or a 1,400-bp product with the two primer pairs confirmed the presence of a phytoplasma in the tomato plants with the bunched top symptoms. No amplification was obtained from plants that had both bunched top and necrotic symptoms on the leaves and that were found to be infected with PVY by ELISA. To determine whether phenolics and other inhibitors from the virus-induced necrotic tissue interfered with the detection of phytoplasmas, plant DNA from these tissues was spiked with known phytoplasmal DNA. Amplification was not successful, confirming the presence of PCR inhibitors. Further purification of the DNA with phenol/chloroform before precipitation helped to obtain DNA of a purer quality from which a positive band representing the sequence of a phytoplasma was amplified. This is the first report of a phytoplasma in tomato in Mauritius.

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First Report of Benomyl Resistance in *Didymella bryoniae* in Delaware and Maryland. K. L. Everts, Department of Natural Resource Science and Landscape Architecture, University of Maryland, Lower Eastern Shore Research and Education Center, Salisbury 21801, and University of Delaware, Research and Education Center, Georgetown 19947. *Plant Dis.* 83:304, 1999; published on-line as D-1999-0115-02N, 1999. Accepted for publication 12 January 1999.

Gummy stem blight, caused by *Didymella bryoniae* (Auersw.) Rehm, is the most severe foliar disease of watermelon, *Citrullus lanatus* (Thunb.) Matsum. & Nakai, in eastern Maryland and southern Delaware. The fungicide benomyl is used in combination with chlorothalonil to manage gummy stem blight. Under conducive environmental conditions, yield losses are high even when fields are sprayed weekly. Resistance of *D. bryoniae* to benomyl has been reported in New York State and South Carolina (1). Gummy stem blight-infected leaves and stems were collected from nine and three fields in Wicomico County, MD, and Sussex County, DE, respectively, in 1996. Infected tissue was also collected from two Wicomico County fields in 1997. One single-spore subculture was obtained to represent each field. Agar plugs were taken from actively growing subcultures and inverted on a 25% (quarter strength) potato dextrose agar medium amended with 0 and 33.1 mg of benomyl per liter, the concentration of benomyl that reduced relative colony diameter of four resistant isolates in New York and South Carolina by 50% (1). Two replicate plates were used per experiment and each experiment was repeated once. After 6 days of growth at 21°C in the dark, the colony diameter was measured. Isolates were classified as sensitive if they were unable to grow, moderately sensitive if colony diameter was reduced 40 to 60%, and resistant if colony diameter was reduced less than 10% on

the benomyl-amended media, compared with unamended media. Isolates that had previously been tested were used as sensitive (W03) and moderately sensitive (NY1) standards (1). In 1996, two isolates were sensitive, four isolates were moderately sensitive, and six isolates were resistant to benomyl. One isolate from 1997 was resistant and the other was moderately sensitive. This is the first report of resistance to benomyl within the *D. bryoniae* population in eastern Maryland and southern Delaware.

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Lisianthus Crown and Stem Rot Caused by *Fusarium avenaceum* in Central Italy. S. Pecchia, Dipartimento di Coltivazione e Difesa delle Specie Legnose, Sez. Patologia Vegetale, Università degli Studi di Pisa, Via del Borghetto 80, I-56124 Pisa, Italy. *Plant Dis.* 83:304, 1999; published on-line as D-1999-0118-01N, 1999. Accepted for publication 15 January 1999.

Lisianthus (*Eustoma russelianum*, syn. *E. grandiflorum*), a member of the Gentian family, is grown in the greenhouse in Italy as a commercial cut-flower crop. Its cultivation is a relatively new industry with high economic potential. However, disease problems have been observed on various greenhouse-planted lisianthus in Central Italy (Tuscany): symptoms of wilting, tan leaf flecks, tan stem discoloration, crown and stem rot, and death. White mycelia and orange sporodochia were also commonly observed on diseased crowns and stems. *Fusarium avenaceum* isolates were recovered from diseased tissues. Four single-spore isolates were selected for artificial inoculation. Pathogenicity tests were conducted in the greenhouse on 16- to 17-week-old *E. russelianum* plants, cvs. Mariacki, Echo, and Heidi, by placing a colonized agar plug, or drops of a conidial suspension, at the crown previously wounded with a sterile scalpel. Moist cotton was fastened to the wound site. Control plants were inoculated with sterile agar plugs or drops of sterile, distilled water. Five plants per cultivar and per treatment were used for each isolate. All plants developed typical disease symptoms within 1 or 2 weeks when inoculated with mycelial plugs or conidial suspensions, respectively. Control plants developed no symptoms. Koch's postulates were completed by consistently reisolating the pathogen from inoculated plants. This is the first report of *F. avenaceum* on lisianthus in Italy and is similar to reports of the disease in California and Florida (1,2).

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Rust Disease on Lemongrass in California. S. T. Koike, University of California Cooperative Extension, Salinas 93901; and R. H. Molinar, University of California Cooperative Extension, Fresno 93702. *Plant Dis.* 83:304, 1999; published on-line as D-1999-0118-02N, 1999. Accepted for publication 15 January 1998.

Lemongrass (*Cymbopogon citratus*) is a minor crop grown for its edible stem and oil. The plant is grown both commercially, by specialty producers, and noncommercially, particularly by homeowners of various Asian communities. For several years, a rust disease has affected the lemongrass plantings in coastal and inland California. Symptoms consist of elongated, stripelike, dark brown lesions that develop on both sides of leaf surfaces. Only lesions on abaxial leaf surfaces erupt and develop dark cinnamon brown uredinial pustules. Lesion development can be substantial, and coalescing lesions result in significant foliage death. Ellipsoidal urediniospores measured 22 to 28 µm by 22 to 25 µm and contained 3 to 4 germ pores in an equatorial configuration. Uredinia contained clavate paraphyses. Teliospores were not observed. Based on the morphology of the uredinia and urediniospores, the rust was identified as *Puccinia nakanishikii* (1). Rust on lemongrass has been observed during various seasons, but this is the first report identifying the causal pathogen in California. A *Darluca* mycoparasite species was often observed in uredinia of infected lemongrass from the coastal counties. In the United States, *P. nakanishikii* has also been reported on lemongrass in Hawaii (2).

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