

Disease Notes

First Report of *Pestalotiopsis maculans* Causing Necrotic Leaf Spots in Nursery Plants of *Arbutus unedo* and *Ceratonia siliqua* in Spain. A. Trapero, M. A. Romero, R. Varo, and M. E. Sánchez, Departamento de Agronomía, ETSIAM, Universidad de Córdoba, Apdo. 3048, 14080 Córdoba, Spain. Plant Dis. 87:1263, 2003; published on-line as D-2003-0812-01N, 2003. Accepted for publication 14 July 2003.

Forestation of agricultural lands has led to a great increase in the production of plants in forest nurseries in southern Spain. During a disease survey of several nurseries, performed in 1998 and 1999, a necrotic leaf spot was found causing defoliation in seedlings of two mediterranean forest species, the ericaceous shrub *Arbutus unedo* and the leguminous tree *Ceratonia siliqua*. The affected plants were 9 to 24 months old and growing in fertilized peat in containers. Symptoms on both species consisted of large necrotic lesions (up to 20 mm in diameter) that were located mainly, but not exclusively, along the leaf margin. On *A. unedo*, necrotic spots were bordered by a red halo, while on *C. siliqua* the halo was dark. The fungus consistently isolated from both hosts was identified as *Pestalotiopsis maculans* (Corda) Nag Raj (= *Pestalotiopsis guepinii*), based on morphological characters (1). Acervular conidiomata (up to 200 µm in diameter) developed on the necrotic lesions of leaves incubated at 100% relative humidity and 20 to 24°C, and in cultures on potato dextrose agar (PDA) at 20 to 24°C over the course of 7 days. All isolates had 5-celled smooth conidia. Apical and basal cells were hyaline, while the three median cells were brown; the upper two were darker than the lower one. Conidia were 22 to 30 µm (mean length) and 5 to 9 µm (mean width). There were typically three (range 1 to 4) apical appendages averaging 17 µm long. The average basal appendage was 6 µm long. One-year-old seedlings and detached leaves from healthy field trees of *A. unedo* and *C. siliqua* were sprayed to runoff with an aqueous conidial suspension (10^6 conidia ml⁻¹) of two isolates of the fungus. All inoculated and control plants sprayed with water only were incubated in a growth chamber at 100% relative humidity and 20 to 24°C for 48 h and then in the same growth chamber at 50 to 80% relative humidity or in the greenhouse (10 to 30°C and 40 to 80% relative humidity) until symptoms developed. Detached leaves were incubated to prevent desiccation in a humid chamber at 100% relative humidity and 20 to 24°C for 2 months. After a period of 2 to 4 months, lesions developed on all inoculated leaves but not on noninoculated controls. Lesion morphology on both hosts was similar to that observed in naturally infected plants in the nurseries. *P. maculans* was reisolated from lesions of all infected leaves but not from control leaves. Although other species of *Pestalotiopsis* have been reported infecting leaves of *C. siliqua* (1), to our knowledge, this is the first report of a *Pestalotiopsis* sp. on *A. unedo* and of *P. maculans* on *C. siliqua*.

Reference: (1) T. R. Nag Raj. Coelomycetous Anamorphs with Appendage-Bearing Conidia. Mycologue Publications, Waterloo, Ontario, Canada, 1993.

First Report of *Cucumber mosaic virus* in *Helleborus foetidus* in France and Italy. L. Cardin and J. P. Onesto, INRA, IPSMV, Phytopathologie, Villa Thuret, BP2078, F-06606 Antibes Cedex, France; and B. Moury, INRA, Station de Pathologie Végétale, Domaine St Maurice, BP94, F-84143 Montfavet Cedex, France. Plant Dis. 87:1263, 2003; published on-line as D-2003-0804-01N, 2003. Accepted for publication 7 July 2003.

Helleborus foetidus L. (bear's foot) is a perennial plant from the family Ranunculaceae that is common in chalky soils of southern and western Europe. It is grown in gardens for its palm-shaped leaves and early flowers. In 1995, yellow-to-white oak leaf and line patterns in leaves of *H. foetidus* plants were observed in Hunawihhr (Alsace, France). The same symptoms were observed in plants in Entrevaux, Biot, and Gourdon (Provence-Alpes-Côte d'Azur, France) in 2000 and 2001, in Triora (Liguria, Italy) in 2002, and on cv. Western Flisk in a nursery in Nice (Provence-Alpes-Côte d'Azur, France) in 2002. Samples collected from these six locations contained six isolates that were further

characterized. Sap extracted from symptomatic plants was mechanically inoculated onto *Nicotiana tabacum* cvs. Xanthi-nc and Samsun, *Chenopodium quinoa*, *C. amaranticolor*, *Vigna unguiculata* cv. Black, and *Cucumis sativus* cv. Poinsett. Symptoms exhibited by the inoculated plants indicated infection by *Cucumber mosaic virus* (CMV). Sap extracted from symptomatic plants reacted positively in double-antibody sandwich-enzyme-linked immunosorbent assays (DAS-ELISA) to antibodies raised against CMV (2). Isometric particles (approximately 30 nm) were observed with an electron microscope in crude sap preparations from infected plants. Following purification of the suspect virus from infected *N. tabacum* (2) and treatment with formaldehyde (1), each isolate was shown to belong to group II of CMV strains (1,3) by double-immunodiffusion analysis. Following isolation from local lesions on *V. unguiculata*, the Hunawihhr isolate was grown in cv. Xanthi-nc plants and back-inoculated to 2-year-old uninfected seedlings of *H. foetidus* by aphids (*Myzus persicae*) or mechanical transmission. Mechanical transmissions were also performed with sap extracted from cv. Xanthi-nc plants infected with the D strain, which belongs to group I of CMV strains (3). Three months postinoculation, symptoms previously described in the original plants were observed in 3 of 10 mechanically inoculated plants and in 2 of 14 aphid-inoculated plants (Hunawihhr isolate), whereas no symptoms could be seen in any of the six plants inoculated with the D strain. On the basis of DAS-ELISA, 7 of 10 plants mechanically inoculated and 7 of 14 plants aphid inoculated with the Hunawihhr isolate were infected with CMV, whereas 3 of the 6 plants inoculated with the D strain were infected with CMV. To our knowledge, this is the first report that *H. foetidus* is a natural host for CMV. Beyond the direct impact of the disease induced by CMV on *H. foetidus*, this perennial and widespread plant species can be an important reservoir of CMV.

References: (1) J. C. Devergne and L. Cardin. Ann. Phytopathol. 7:225, 1975. (2) J. C. Devergne et al. Ann. Phytopathol. 10:233, 1978. (3) M. J. Roossinck. J. Virol. 76:3382, 2002.

***Subanguina radicolica*, the Root-Gall Nematode Infecting *Poa annua* in New Brunswick, Canada.** N. A. Mitkowski and N. Jackson. Department of Plant Sciences and Entomology, University of Rhode Island, Kingston 02881. Plant Dis. 87:1263, 2003; published on-line as D-2003-0807-01N, 2003. Accepted for publication 27 July 2003.

Poa annua frequently is found as the dominant turfgrass species on golf course putting greens grown in the range of cool-season grasses. While not intentionally established, it is an aggressive weed in stands of bentgrasses (*Agrostis* spp.). When significant encroachment of *P. annua* occurs, it often is maintained indefinitely. In May 2003, *P. annua* putting greens at the Riverside Country Club in Rothesay, New Brunswick, Canada showed signs of an unidentified disease. Putting greens were slow to green up and large chlorotic patches were evident across affected areas. When roots were examined, extensive galling was observed. Galls were slender and often twisted in appearance. Upon dissection of washed galls, hundreds of eggs were exuded into the surrounding water droplet, and mature male and female nematodes were observed. Further morphological examination of males, females, and juvenile nematodes demonstrated that they were *Subanguina radicolica* (Greef 1872) Paramanov 1967 (1,2). Each *P. annua* plant had an average of four galls (with a range of two to nine) primarily located within the uppermost centimeter of the soil. Of 18 *P. annua* putting greens, four were affected by the nematode and displayed the same damage symptoms. *S. radicolica* has been identified from American beachgrass in Rhode Island and from *P. annua* in Oregon, but to our knowledge, this is the first report of the nematode affecting *P. annua* on a golf course in eastern North America.

References: (1) W. F. Mai and P. Mullin. Plant-Parasitic Nematodes: A Pictorial Key to Genera. Cornell University Press, Ithaca, New York, 1996. (2) G. Thorne. Principles of Nematology. McGraw-Hill Book Company, Inc., New York, 1961.

(Disease Notes continued on next page)

Disease Notes (continued)

Sclerotinia Stem and Crown Rot of Corn-Salad Caused by *Sclerotinia minor* in California. S. T. Koike, University of California Cooperative Extension, Salinas 93901. Plant Dis. 87:1264, 2003; published on-line as D-2003-0805-01N, 2003. Accepted for publication 20 July 2003.

Corn-salad or lamb's lettuce (*Valerianella locusta*) is a specialty leafy green vegetable that is grown commercially in California and is harvested fresh for use in salads. In 2001, field plantings of corn-salad in coastal California showed symptoms and signs of a previously undescribed disease. Initial symptoms consisted of a light tan discoloration at the crown and lower leaf attachment areas. Once this discoloration was observed, the crown rapidly developed a soft rot, attached leaves wilted, and the entire plant collapsed. White mycelium and small (0.5 to 3.0 mm in diameter), irregularly shaped, black sclerotia formed on the crowns and lower leaves. Isolations from symptomatic crowns, mycelium, and sclerotia produced colonies of *Sclerotinia minor* (1). Seven-week-old corn-salad plants grown in a peat moss-based rooting medium in pots were used to test pathogenicity. Sclerotia from six corn-salad isolates from the Salinas Valley were inserted into slots made in the potting mix adjacent to the crowns of plants. Sclerotia were not placed in slots for control corn-salad. All test plants were incubated in a greenhouse at 21 to 23°C. After 4 weeks, inoculated corn-salad plants wilted and collapsed, and *S. minor* was reisolated from necrotic crown and stem tissues. Uninoculated plants were asymptomatic. Using the same method, sclerotia from one lettuce (*Lactuca sativa*) isolate were used to inoculate corn-salad plants that produced similar symptoms. All experiments were repeated and results were similar. To our knowledge, this is the first report of corn-salad as a host of *S. minor* in California and the United States. The susceptibility of corn-salad to *S. minor* from lettuce indicates that this crop might contribute to inoculum levels and lettuce drop incidence for the extensive lettuce plantings in the Salinas Valley.

Reference: (1) C. L. Patterson and R. G. Grogan. Plant Dis. 72:1046, 1988.

Outbreak of Tobacco streak virus Causing Necrosis of Cucumber (*Cucumis sativus*) and Gherkin (*Cucumis anguria*) in India. M. Krishnareddy, Devaraj, Lakshmi Raman, Salil Jalali, and D. K. Samuel, Division of Plant Pathology, Indian Institute of Horticultural Research, Hessaraghatta Lake PO, Bangalore-560089, India. Plant Dis. 87:1264, 2003; published on-line as D-2003-0804-03N, 2003. Accepted for publication 6 June 2003.

Cucumber (*Cucumis sativus* L.) and Gherkin (*Cucumis anguria* L.) are important cucurbitaceous vegetables grown in India for slicing and pickling. During the 2000 to 2002 rainy season and summer, a new virus disease, causing yield losses of 31 to 75% in Bangalore, Bellary, Davanagiree, and Tumkur districts of Karnataka State, infected cucumber and gherkin. Symptoms were tip necrosis characterized by necrotic lesions on leaves, and a general leaf and stem necrosis extending to mid veins, petioles, flower buds and tip, eventually resulting in dieback of vines. Tissue extracts from symptomatic leaves of cucumber and gherkin were mechanically inoculated on several herbaceous indicator plants (cowpea, cucumber, pepper, Zinnia, watermelon, *Chenopodium amaranticolor*, sunflower, *Nicotiana glutinosa*, *N. tabacum*, and *Gomphrena globosa*). On most hosts, symptoms of chlorotic or necrotic lesions followed by mottle or systemic necrosis were observed. Back-inoculation from the symptomatic indicator plants onto cucumber and gherkin resulted in symptoms typical of those observed in the field. Electron microscopic examination of leaf-dip preparation and ultra thin sections of virus infected plant samples showed the presence of isometric particles 25 to 28 nm in diameter. Similar types of particles were observed when infected samples were trapped in immunosorbent electron microscopy with polyclonal antibodies specific to Tobacco Streak virus (TSV) but not to Watermelon silver mottle virus (WSMV). Enzyme-linked immunosorbent assay tests using leaf extracts of field-collected samples and sap-inoculated plants showed positive reaction to antibodies of TSV (1) but not to antibodies of Cucumber mosaic virus, WSMV, Watermelon bud necrosis virus, Papaya ring spot virus W strain, and Zucchini yellow mosaic virus. Reverse transcription-polymerase chain reaction (RT-PCR) of RNA extracts of infected samples of field and

inoculated symptomatic plants was done by using primers derived from TSV RNA3 specific for the coat protein (CP) region of TSV (2). A 800-bp specific DNA fragment was amplified from infected cucumber and gherkin but not from healthy control plants. Sequence analysis of cloned PCR fragments revealed nucleotide identities of 99% with TSV isolates from cotton, mungbean, sunhemp, and sunflower (GenBank Accessions Nos. AF515824, AF515823, AF515825, and AY061929) and 88% with TSV-WC (GenBank Accession No. X00435). On the basis of host range, serological relationship, electron microscopy, and sequence analysis of the CP region, the virus was identified as a strain of TSV. To our knowledge, this is the first report of natural occurrence of TSV on cucumber and gherkin in India.

References: (1) A. I. Bhat et al. Arch. Virol. 147:651, 2002. (2) B. J. C. Cornelissen et al. Nucleic Acids Res. 12:2427, 1984.

First Report of *Rhizoctonia solani* AG-2-4 on Carrot in Georgia. D. R. Sumner and S. C. Phatak, University of Georgia Coastal Plain Experiment Station, Tifton 31793; and D. E. Carling, USDA/ARS, 533 East Fireweed Ave, Palmer 99645. Plant Dis. 87:1264, 2003; published on-line as D-2003-0814-02N, 2003. Accepted for publication 14 July 2003.

Anastomosis group-2-4 (AG-2-4) of *Rhizoctonia solani* Kühn was formally described in 2002 (1), but it was first collected in 1983 in Georgia from corn (*Zea mays* L.) exhibiting symptoms of crown and brace root rot. Although occasionally present on diseased corn roots, the role that isolates of AG-2-4 play in crown and brace root rot of corn is not specifically known (2). More recently, as part of a broad multi-year (1996 to 2000) survey of root diseases in field grown carrot (*Daucus carota* L.), isolates of *R. solani* AG-2-4 were recovered from diseased carrot plants in various stages of growth from fields of sandy loam soil at many locations in southern Georgia, including commercial fields in Coffee and Tift counties. During the 1996 to 2000 growing seasons, 123 isolates of *Rhizoctonia* sp. (including multinucleate and binucleate types) were collected from lesions on developing and mature carrot roots. Of these, 34% were AG-2-4, 10% were AG-2-2IV, 6% were AG-4, and 32% were binucleate *Rhizoctonia* sp. The remaining 18% were lost prior to AG typing. An additional 40 isolates were collected from carrot seedlings or soil and of these, 55% were AG-4, 18% were AG-2-2IV but none were AG-2-4. Virulence on carrot seedlings by two isolates of AG-2-4 (777R1P5-SL2 and 758C) was compared with virulence of isolates of AG-4, AG-2-2IV, AG-2-1, and binucleate *Rhizoctonia* sp. Carrot seeds soaked for 5 min in 0.5% NaOCl were planted in petri dishes containing moist autoclaved sandy loam soil. Each dish was inoculated in the center with a 10-mm-diameter disk cut from a 9-day-old potato dextrose agar (PDA) culture of the appropriate isolate. Petri dishes were placed in a 26°C incubator for 9 days, and then the seedlings were rated for disease. Virulence on mature carrot root tissue was also assessed on the same set of isolates. Cross sections of carrot roots (5 to 10 mm thick) were surface disinfested in 0.5% NaClO for 5 min. Three cross sections were placed on moist filter paper in sterile petri dishes and each was inoculated with a 5-mm-diameter disk of inoculum cut from 8- to 10-day-old cultures growing on PDA. All treatments were rated for damage following incubation on a lab bench at 21 to 24°C for 7 days. Isolate 777R1P5-SL2 caused moderate damage to seedlings but minimal rotting of mature carrot root tissue. Isolate 758C did no damage to either seedlings or root tissue. The AG-4 and AG-2-2IV isolates killed all seedlings and caused extensive rot on mature root tissue. The AG-2-1 isolate caused moderate damage to seedlings and mature root tissue, whereas isolates of binucleate *Rhizoctonia* sp. damaged neither seedling nor mature root tissue. These data suggest that some isolates of *R. solani* AG-2-4 may be capable of causing minor damage to carrot seedlings in the field in Georgia, but isolates of *R. solani* AG-4 and AG-2-2IV pose greater threat to seedlings and mature roots of carrot. Published data shows that isolates of AG-2-4 can kill seedlings of lettuce, cauliflower, and broccoli in the laboratory (1). *R. solani* AG-2-4 also may be capable of killing these crops in the field, all of which are grown commercially in Georgia. To our knowledge, this is the first report of *R. solani* AG-2-4 on carrot in Georgia.

References: (1) D. E. Carling et al. Phytopathology 92:43, 2002. (2) D. R. Sumner and D. K. Bell. Phytopathology 72:86.

First Report of Infection of Bermudagrass by *Bipolaris sorokiniana* in the Southeastern United States. R. G. Pratt, USDA, ARS, Waste Management and Forage Research Unit, P.O. Box 5367, Mississippi State, MS 39762. Plant Dis. 87:1265, 2003; published on-line as D-2003-0801-01N, 2003. Accepted for publication 8 July 2003.

Bipolaris sorokiniana (Sacc.) Shoemaker is a major foliar and root-infecting pathogen of cool-season forage and turf grasses and small grains in the southeastern United States (2). In North America, *B. sorokiniana* has been reported from bermudagrass (*Cynodon dactylon* [L.] Pers.) once in California in 1961 (1), and rarely from other warm-season grasses in the southeastern United States. In May, July, September, and October 2002, *B. sorokiniana* sporulation was observed on leaves of common bermudagrass exhibiting necrotic lesions and dieback in waste application fields on three commercial swine farms in Chickasaw, Lowndes, and Webster counties, MS. Leaves were collected (50 per farm per month), surface-disinfested, plated on water agar, and observed for fungal sporulation on leaf surfaces after 7 to 10 days (3,4). The pathogen was detected on 1 to 3 farms each month in leaves that were infected with numerous other dematiaceous hyphomycetes (3,4). Three randomly selected single-spore isolates of *B. sorokiniana* from each of bermudagrass and annual ryegrass (*Lolium multiflorum* Lam.), collected at the Webster County farm, were compared for select features of morphology and pathogenicity on bermudagrass. Isolates differed significantly in growth rates, amount of sporulation, and spore sizes on cornmeal agar, but differences were not consistently related to hosts of origin. In plants inoculated by atomizing equal quantities of spores (2.8×10^4 /ml) onto foliage, isolates of *B. sorokiniana* from bermudagrass and ryegrass both caused significantly ($P = 0.05$) more severe foliar necrosis after 10 days than *B. cynodontis* (5 pots of seeded plants per treatment in each of two experiments). *B. sorokiniana* was reisolated from disinfested, symptomatic bermudagrass leaf tissue following inoculations. To our knowledge, this is the first report of *B. sorokiniana* on bermudagrass in North America outside of California (1) and indicates that this pathogen is highly virulent on bermudagrass in the southeastern United States (3,4). Of potentially greater importance is the fact that one of the most common and widespread forage and turf grass species in the southeastern United States can serve as an alternate host for maintenance and increase of inoculum of *B. sorokiniana* during summer months.

References: (1) R. M. Endo. Plant Dis. Rep. 45:869, 1961. (2) D. F. Farr et al. Fungal Databases. Systematic Botany and Mycology Laboratory, On-line publication. ARS, USDA, 2003. (3) R. G. Pratt. Agron. J. 92:512, 2000. (4) R. G. Pratt. Plant Dis. 85:1206, 2001.

First Report of *Peronospora arborescens* as the Causal Agent of Downy Mildew on *Papaver nudicaule* in Italy. A. Garibaldi, A. Minuto, D. Bertetti, and M. L. Gullino, DIVAPRA—Patologia vegetale, Via Leonardo da Vinci 44, 10095 Grugliasco, Italy. Plant Dis. 87:1265, 2003; published on-line as D-2003-0804-02N, 2003. Accepted for publication 6 July 2003.

Iceland poppy (*Papaver nudicaule* L.) is increasingly grown on the Italian Riviera for export as a cut flower. During the spring of 2003, leaves with irregular, brown, angular spots were collected from a commercial crop grown outdoors near Ventimiglia (northern Italy) with temperatures ranging from 3 to 14°C. Leaves of infected plants appeared curled and blistered; the infected portions of the leaves turned chlorotic. On both surfaces of infected leaves, a characteristic gray, furry growth was evident, particularly at the center of the necrotic areas. Infected leaves eventually died without dropping. Basal leaves with poor air circulation were the most severely affected by the disease. Microscopic observations revealed conidiophores branching dichotomically at least five times. Conidiophores ended with sterigmata bearing single conidia. Conidia measured 19 to 24 × 16 to 18 μm and were elliptical to near spherical and hyaline. Oospores were not present. The pathogen was identified as *Peronospora arborescens* based on the morphological characteristics (2). Pathogenicity was confirmed by inoculating 60-day-old healthy *P. nudicaule* plants with a conidial suspension (1×10^5 conidia per ml). Five plants were used as replicates. Inoculated and noninoculated plants were maintained in a growth chamber at 15°C and 90 to 95% relative humidity. After 7 to 10 days, typical symptoms of downy mildew developed on inoculated plants. *Peronospora arborescens*

was observed on infected leaves. Noninoculated plants did not show symptoms. To our knowledge, this is the first report of *Peronospora arborescens* on *P. nudicaule* in Italy. *Peronospora arborescens* was previously reported on *P. nudicaule* and on many other species of *Papaver* (*P. somniferum*, *P. dubium*, *P. caucasicum*, *P. rhoeas*, *P. setigerum*, and *P. argemone*) in several countries (1,2).

References: (1) P. J. Cotteril and I. G. Pascoe. Australas. Plant Pathol. 27:263, 1998. (2) S. M. Francis. No. 686 in: Descriptions of Pathogenic Fungi and Bacteria. CMI, Kew, Surrey, U.K., 1981.

First Report of Fusarium Wilt of Lettuce Caused by *Fusarium oxysporum* f. sp. *lactucae* in Arizona. M. E. Matheron, University of Arizona, Yuma Agricultural Center, Yuma 85364; and S. T. Koike, University of California Cooperative Extension, Salinas 93901. Plant Dis. 87:1265, 2003; published on-line as D-2003-0806-01N, 2003. Accepted for publication 23 July 2003.

A new wilt and root rot disease was observed in 6 and 11 commercial fields of lettuce (*Lactuca sativa*) in western Arizona during the fall of 2001 and 2002, respectively. Distance between infested sites ranged from approximately 0.5 to 39 km. Five head lettuce cultivars as well as a red leaf lettuce cultivar were affected. Disease symptoms included yellowing and wilting of leaves, as well as stunting and plant death. The cortex of the crown and upper root of infected plants usually was decayed and reddish brown. Disease symptoms first appeared at the time of plant thinning and continued to develop up to plant maturity. *Fusarium oxysporum* was consistently isolated from symptomatic plant roots. Seeds of cv. Lighthouse were planted in nonsterile vermiculite within 3.0-cm-square × 7.0-cm-deep cells in a transplant tray and thinned to a single plant per cell. When the first true leaves were emerging, 10 individual seedlings were inoculated with a single-spore isolate of *F. oxysporum* recovered from diseased lettuce root cortex tissue. Inoculum was prepared by growing the fungus on potato dextrose agar in 100-mm-diameter × 15-mm-deep plastic petri dishes at 28°C with a 12-h photoperiod under fluorescent light. Once the fungus completely covered the agar surface, 50 ml of sterile distilled water was added to the dish, and the mycelia and conidia on the surface were scraped off the agar and suspended in the water. This fungal suspension was decanted, and a 2-ml aliquot containing 1.8×10^5 CFU was pipetted into the vermiculite near the stem of each lettuce seedling. Ten plants grown in noninfested vermiculite served as uninoculated controls. After inoculation, plants were maintained in a growth chamber at 28°C with a 12-h photoperiod under fluorescent light for 3 weeks. Symptoms of yellowing, wilt, vascular decay, and often plant death developed during the incubation period on all inoculated plants but not on control plants. *Fusarium oxysporum* was consistently reisolated from inoculated plants but not from uninoculated plants. The experiment was repeated and yielded the same results. A wilt and root rot disease of lettuce attributed to *F. oxysporum* f. sp. *lactucae* was first reported in Japan in 1967 (3) and subsequently in the United States (San Joaquin Valley of California) in 1993 (2), and Italy in 2002 (1). The researchers of the U.S. report did not cite the earlier work from Japan and described the pathogen as *F. oxysporum* f. sp. *lactucum*. The Arizona isolate used to demonstrate pathogenicity was of the same vegetative compatibility group as an isolate of the pathogen from lettuce in California reported in 1993. Several companies grow and harvest lettuce in Arizona and California. At the end of production and harvest in the fall, tractors, implements, and harvesting equipment are transported from the San Joaquin Valley in California to western Arizona. The similarity between the isolate of *F. oxysporum* f. sp. *lactucae* from western Arizona and the San Joaquin Valley of California suggest a possible introduction of the pathogen into Arizona from California, perhaps on soil adhering to farm equipment. To our knowledge, this is the first report of *F. oxysporum* f. sp. *lactucae* infecting lettuce in Arizona.

References: (1) A. Garibaldi et al. Plant Dis. 86:1052, 2002. (2) J. C. Hubbard and J. S. Gerik. Plant Dis. 77:750, 1993. (3) T. Matuo and S. Motohashi. Trans. Mycol. Soc. Jpn. 8:13, 1967.

(Disease Notes continued on next page)

Disease Notes (continued)

Occurrence of Rust Disease Caused by *Puccinia lagenophorae* on *Cineraria* in the Americas. J. R. Hernández, USDA-ARS, Systematic Botany and Mycology Laboratory, Beltsville, MD 20705; M. Daughtrey, Cornell University, Long Island Horticultural Research and Extension Center, Riverhead, NY 11901; and J. Jens, New York State Department of Agriculture and Markets, Cutchogue 11935. *Plant Dis.* 87:1266, 2003; published on-line as D-2003-0725-01N, 2003. Accepted for publication 6 July 2003.

Cineraria (*Pericallis* × *hybrida* B. Nord.), sometimes referred to as *Senecio cruentus* (Masson ex L'Hér.) DC., Asteraceae, is native to the Canary Islands. This important ornamental plant is cultivated worldwide and is of significant economic importance in the United States as a flowering potted plant. In February 2002, rust pustules were observed on cineraria plants cultivated in a greenhouse in Suffolk County, New York. The rust was identified as *Puccinia lagenophorae* Cooke (2). Aecioid sori were up to 1 mm in diameter and densely clustered, yellowish, and amphigenous but mainly hypophyllous on swollen, rounded, infected areas. The peridium is cupulate, lacerate, and composed of colorless cells measuring 10.5 to 20 × 18 to 33 µm that are verrucose on the outer surface and finely verrucose on the inner surface. Spores are sessile, 12 to 18 × 10.5 to 17 µm, yellowish, minutely verrucose with conspicuous, loosely attached globules, and in long chains in fresh material. Telia were not observed. Signs and symptoms were reproduced by rubbing healthy leaves of *Pericallis* × *hybrida* with a rust-infected leaf of *Pericallis* × *hybrida* bearing aecia sori. Plants were enclosed in a plastic bag for 18 h and held at 21°C. Aecioid sori developed within 10 days. Voucher specimens have been deposited in the U.S. National Fungus Collection (BPI 841952 and 841953). Common groundsel (*S. vulgaris* L.) infected by *P. lagenophorae* was observed outside the greenhouse in which the infected cineraria were discovered (W. L. Bruckart and A. Senesac, unpublished), and we postulated that the cineraria in the greenhouse might have been infected by spores from those infected weeds. Cross-infectivity was tested by rubbing leaves of groundsel on which aecia sori of *P. lagenophorae* were present on five leaves on each of three healthy cineraria plants. Incubation conditions were the same as described above. Aecioid sori developed 10 days after inoculation on four leaves of one plant. The control plants remained healthy. The inoculation was repeated on two plants and aecia sori developed on leaves of both plants. To our knowledge, this is the first report of *P. lagenophorae* on cineraria in the Americas. *P. lagenophorae* is autoecious, native to Australia and New Zealand, and has approximately 60 known host species (2). Recently, it was reported in California on common groundsel (3) and English daisy (*Bellis perennis* L.) (1).

References: (1) S. T. Koike and M. Scholler. *Plant Dis.* 85:562, 2001. (2) M. Scholler. *Sydowia* 49:177, 1997. (3) M. Scholler and S. T. Koike. *Plant Dis.* 85:335, 2001.

First Report of *Plasmopara petroselinii* on Parsley in Belgium. C. Crepel and S. Inghelbrecht, Department of Crop Protection, Agricultural Research Center, Burg Van Gansberghelaan 96, B-9820 Merelbeke, Belgium. *Plant Dis.* 87:1266, 2003; published on-line as D-2003-0813-01N, 2003. Accepted for publication 28 July 2003.

Plasmopara petroselinii (Oomycetes) was identified on parsley (*Petroselinum crispum* subsp. *crispum* cv. Petra) (Apiaceae) in Belgium during the winters of 2001 and 2002. The fungus was present in numerous fields, especially on parsley grown in plastic tunnels. Losses were sometimes dramatic and similar to disease problems in France and Switzerland where 80 and 50 ha, respectively, were found infected (1). Initial symptoms consisted of white spots on the upper leaf surface. As the disease progressed, the spots enlarged, became angular, and turned yellow. At the location of leaf spots, white-to-grayish white mycelium developed on the lower surface of the leaves. Eventually the leaves and leaf stalks rotted. The pathogen was identified at the Centraalbureau voor Schimmelcultures (CBS) (Utrecht, the Netherlands) as the downy mildew organism *P. petroselinii* (= *P. umbelliferarum* pro parte = *P. nivea* pro

parte = *P. crustosa*), based on morphological characteristics. Sporangia were papillate, lemon-shaped, almost hyaline, and 9 to 20 µm long, and produced on tree-like sporangiophores (100 to 420 × 6 to 8.5 µm) that were monopodially branched at approximately right angles. The sporangiophores usually bear three sterigma (4 to 19 × 2 to 3 µm) that narrow toward the tip (2). Prophylactic actions are the primary method to prevent the disease. Fungicides based on propamocarb can be used as a curative control method. To our knowledge, this is the first report of *P. petroselinii* on parsley in Belgium.

References: (1) E. Béliard and J. Thibault. *Phytoma* 554:2, 2002. (2) M. Brandenburger. Page 451 in: *Parasitische Pilze an Gefäßpflanzen in Europa*. Fischer Verlag, Stuttgart, Germany, 1985.

First Detection of *Phytophthora ramorum* Mating Type A2 in Europe. Sabine Werres, Federal Biological Research Centre for Agriculture and Forestry, Institute for Plant Protection in Horticulture, Messeweg 11/12, D-38104 Braunschweig, Germany; and Daphné De Merlier, Agricultural Research Centre, Department of Biological Control and Plant Genetic Resources, Rue de Liroux, 4, B-5030 Gembloux, Belgium. *Plant Dis.* 87:1266, 2003; published on-line as D-2003-0814-01N, 2003. Accepted for publication 7 August 2003.

Since its original isolation in 1993, *Phytophthora ramorum* has become an important pathogen. Initially, it was determined to be the causal agent of a twig blight of *Rhododendron* spp. in Germany and the Netherlands (3). Around the same period, symptoms and mortality on oak (*Quercus* spp.) and tanoak (*Lithocarpus densiflorus*) were associated with *P. ramorum* in California (2), where the disease was named sudden oak death. Subsequently, *P. ramorum* has been detected on a wide range of forest trees and shrub species in the United States. In Europe, the pathogen has spread to many countries, primarily on nursery plants of *Rhododendron* and *Viburnum* spp., and recently, on *Camellia japonica*, *Kalmia latifolia*, *Pieris formosa* var. *forrestii*, *P. japonica*, *Leucothoe* sp., *Syringa vulgaris*, and *Taxus baccata*. *P. ramorum* has not been observed in European forests. *P. ramorum* is heterothallic, and initial in vitro mating studies on agar media suggested that only the A1 mating type occurred in Europe, while only the A2 mating type was present in the United States (4). However, an isolate collected in 2002 in Belgium (1) appears to be the A2 mating type. This isolate (CBS 110901, Centraal Bureau voor Schimmelcultures, Baarn, the Netherlands) originated from an imported *V. bodnantense* plant at an ornamental nursery. A hyphal tip culture (BBA 26/02) of this isolate produced no oogonia on carrot piece agar after 6 weeks in pairing tests with other *Phytophthora* species of mating type A2. When paired with mating type A1 of *P. cambivora*, *P. cinnamomi*, *P. cryptogea*, and *P. drechsleri*, however, oogonia were observed in all pairings within 6 weeks. The number of oogonia was low in all pairings but was highest in pairings with *P. cryptogea*. No oospores were produced after 6 weeks between *P. ramorum* isolates BBA 26/02 and BBA 9/95 (from the holotype, mating type A1), but gametangia were observed when these isolates were paired on *Rhododendron* sp. twigs. Normal oogonia were produced on the outgrowing mycelium when pieces from these twigs were placed on carrot piece agar. The shape and size of the oogonia produced on carrot piece agar after pairing with *P. cryptogea* and on *Rhododendron* sp. twigs after pairing with *P. ramorum* BBA 9/95 were similar (24 to 34 µm, mean 29.6 µm and 25 to 33 µm, mean 30.6 µm, respectively). To our knowledge, this is the first observation of *P. ramorum* mating type A2 in Europe.

References: (1) D. De Merlier et al. *Plant Dis.* 87:203, 2003. (2) D. M. Rizzo et al. *Plant Dis.* 86:205, 2002. (3) S. Werres et al. *Mycol. Res.* 105:1166, 2001. (4) S. Werres and B. Zielke. *J. Plant Dis. Prot.* 110:129, 2003.

First Report of A1 Mating Type of *Phytophthora ramorum* in North America. E. M. Hansen, P. W. Reeser, W. Sutton, and L. M. Winton, Department of Botany and Plant Pathology, Oregon State University, Corvallis 97331; and N. Osterbauer, Oregon Department of Agriculture, Salem 97301. *Plant Dis.* 87:1267, 2003; published on-line as D-2003-0808-01N, 2003. Accepted for publication 1 August 2003.

Phytophthora ramorum is known in Europe and the western United States (1). In Europe, it is found in nurseries and landscape plantings. In the United States, it has been confined to coastal forests, and in California, it is found in a few horticultural nurseries. All European isolates tested have been A1 mating type, while all North American isolates were A2 mating type (2). Amplified fragment length polymorphism markers also indicated that the populations on the two continents are distinct, and nearly all North American isolates are from one clone (Kelly Ivors, unpublished). In June 2003, *P. ramorum* was isolated from diseased *Viburnum* and *Pieris* spp. cultivars from a Clackamas County nursery in northern Oregon and diseased *Camellia* sp. cultivar from a Jackson County nursery in southern Oregon. Representative isolates were submitted to the American Type Culture Collection, Manassas, VA. As part of the effort to determine the origin of these new infestations, we tested the nursery isolates for mating type. Seven Oregon nursery isolates, three Oregon forest isolates (from the predominant North American clone), and two European isolates were paired. Agar plugs from 3-day-old colonies were placed in close proximity on carrot agar plates, and then the plates were examined for oogonia after 3 and 10 days as advised by C. M. Brasier (personal communication). Oogonia and antheridia typical of *P. ramorum* (2) formed when isolates from the Clackamas County nursery were paired with the Oregon forest isolates and also when isolates from the Jackson County nursery were paired with the European isolates. Gametangia also formed in pairings between Oregon forest isolates and European isolates, but not in any other combinations. We developed polymerase chain reaction (PCR) primers for four microsatellite loci and determined allele sizes for the same set of isolates (unpublished). Microsatellite alleles of the Clackamas County isolates were identical to the European tester isolates, and alleles of the Jackson County isolates were identical to the Oregon forest isolates. These results indicate that the recent Oregon nursery infestations are of separate origins. The Clackamas County isolates are A1 mating type and have microsatellite alleles like the European testers, but according to shipping records, the nursery has received no host nursery stock directly from Europe. However, host nursery stock has been received from a Canadian nursery. The Jackson County isolates are of A2 mating type and have microsatellite alleles like the forest isolates of Oregon, which is consistent with the reported origin of these plants from a California nursery. These preliminary microsatellite results need to be validated against a larger isolate set but are congruent with the mating type results. The Oregon nursery infestations highlight the dangers of unregulated or underregulated transport of host nursery stock from infested areas to noninfested areas. All host plants from infested nursery blocks at the affected Oregon nurseries have been destroyed by incineration, and a monitoring program has been implemented. Other host nursery stock on site has been taken "off-sale" pending verification that it is disease free, per the United States Department of Agriculture, APHIS requirements.

References: (1) J. M. Davidson et al. On-line publication. doi:10.1094/PHP-2003-0707-01-DG. *Plant Health Progress*, 2003. (2) S. Werres et al. *Mycol. Res.* 105:1155, 2001.

e-Xtra*

First Report of a *Labyrinthula* sp. Causing Rapid Blight Disease of Rough Bluegrass and Perennial Ryegrass. M. W. Olsen, D. M. Bigelow, and R. L. Gilbertson, Department of Plant Pathology, University of Arizona, Forbes 204, Tucson 85721; and L. J. Stowell and W. D. Gelernter, PACE Turfgrass Research Institute, 1267 Diamond St., San Diego, CA 97109. *Plant Dis.* 87:1267, 2003; published on-line as D-2003-0729-01N, 2003. Accepted for publication 6 July 2003.

A *Labyrinthula* sp. was isolated from symptomatic rough bluegrass (*Poa trivialis* L.) and perennial ryegrass (*Lolium perenne* L.) from a golf course in Arizona. Initial symptoms were a water-soaked appearance and rapid collapse of small patches of turf foliage. The affected turf died, and patches coalesced to form large dead areas after several weeks. The symptoms were those of the disease recently termed "rapid blight" for which the causal agent has not been identified (1). Rapid blight was first observed in southern California in 1995 and has become increasingly problematic in 10 other states on several cool-season turfgrasses (1). In Arizona, it is associated with high salinity irrigation water. In microscopic examinations of symptomatic *P. trivialis* and *L. perenne* leaf tissue from November 2002 to February 2003, fusiform or spindle-shaped vegetative cells (4 to 5 × 15 to 20 μm) were observed in leaf cells. These cells are consistently associated with rapid blight (1) and are typical in size and shape of those described for *Labyrinthula* spp. (3,4). The fusiform cells were cultured in 1% horse serum water agar medium made with irrigation water (electrical conductivity [EC] = 3.5 to 4.0 dS/m) from a golf course in central Arizona with rapid blight. The cells readily formed colonies on this medium and exhibited gliding motility along a network of hyaline slime filaments as previously described for the genus *Labyrinthula* (3,4). Koch's postulates were fulfilled by inoculating *P. trivialis* and *L. perenne* seedlings with *Labyrinthula* sp. isolated from naturally infested *P. trivialis* in two experiments. The grasses were started from seed and grown as a lawn in containers in the laboratory. Both experiments were repeated once. In the first experiment, infested autoclaved leaf pieces of *P. trivialis* were used as inoculum. Inoculated leaf pieces were placed within each of several bundles of 4 to 6 leaves and held loosely in place with a 0.5-cm wide ring of tygon tubing. Seedlings were irrigated with sterilized irrigation water from the golf course (EC = 4.0 dS/m). In the second experiment, agar discs from *Labyrinthula* sp. colonies on 1% horse serum agar were used as inoculum by placing the agar discs in contact with leaves. Seedlings were irrigated with sterile tap water adjusted to 4.0 dS/m using synthetic sea salt (Instant Ocean, Aquarium Systems, Inc., Mentor, OH) Leaf tissue of all inoculated seedlings became water soaked within 3 to 7 days and collapsed within 10 days in both experiments. Fusiform cells were observed in inoculated leaf tissue cells, and the *Labyrinthula* sp. was reisolated from 100% of selected symptomatic seedlings. Control seedlings treated with noninfested leaf pieces or sterile agar pieces did not develop symptoms, and no fusiform cells were isolated from the leaf tissue. *Labyrinthula* spp. are usually associated with marine systems (3). *Labyrinthula zosterae* D. Porter & Muehlst. has been identified as the causal agent in a marine grass wasting disease (2), but to our knowledge, no *Labyrinthula* spp. have been described as pathogens of terrestrial plants.

References: (1) S. B. Martin et al. *Phytopathology* (Abstr.) 92:(suppl)S52, 2002. (2) L. K. Muehlstein et al. *Mycologia* 83:180, 1991. (3) K. S. Porkorny. *J. Protozool.* 14:697, 1967. (4) D. Porter. *Handbook of Protoctista*. Jones and Bartlett, Boston, MA, 1990.

*The e-Xtra logo stands for "electronic extra" and indicates this Disease Note online contains supplemental material not included in the print edition.

(Disease Notes continued on next page)

Disease Notes (continued)

First Report on the Presence of *Leptosphaeria maculans* Pathogenicity Group-3, the Causal Agent of Blackleg of Canola in Manitoba. W. G. D. Fernando and Y. Chen, Department of Plant Science, University of Manitoba, Winnipeg, MB R3T 2N2, Canada. Plant Dis. 87:1268, 2003; published on-line as D-2003-0808-02N, 2003. Accepted for publication 23 July 2003.

Blackleg, caused by *Leptosphaeria maculans* (Desmaz.) Ces. & De Not. (anamorph = *Phoma lingam*) (Tode:Fr.) Desmaz., is an economically important and serious disease of canola (*Brassica napus* L.) in Australia, Europe, and Canada. *L. maculans* isolates can be categorized into four pathogenicity groups (PGs) on the basis of the interaction phenotypes (IP) on the differential canola cvs. Westar, Glacier, and Quinta (1) by using a standard screening protocol in the greenhouse. PG1 isolates are weakly virulent and PG2, PG3, and PG4 isolates are highly virulent. In Manitoba, *L. maculans* population consists mainly of PG2 (virulent on cv. Westar; avirulent on cvs. Glacier and Quinta) and a few PG1 isolates (avirulent on all three differentials). The Oilseed Pathology Lab in the Department of Plant Science, University of Manitoba examines the pathogenic variability of blackleg isolates obtained from Manitoba each year. In 2002, the blackleg-resistant cv. Q2, was found to be severely infected in Roland, Manitoba. The canola stubble collected from a coop trial plot (Roland, Manitoba) and a farm in East Selkirk (60 km northeast of Winnipeg, Manitoba) was isolated for the blackleg fungus. Small pieces of stubble were cut from the pseudothecia forming section and surface sterilized with 1% sodium hypochlorite solution for 3 to 5 min and then rinsed in sterile distilled water. V8 agar medium containing 1% streptomycin sulphate was used to culture the isolates under continuous cool-white fluorescent light for 14 days. Pure cultures of the pathogen were isolated and characterized as *L. maculans* by means of colony morphology, pycnidia, and microscopic observations of pycnidiospores. Pycnidiospores that formed on V8 plates were flooded with 10 ml of sterile distilled water and then harvested by filtering through sterilized Miracloth and kept at -20°C. The isolates were passed once through cv. Westar to maintain their virulence. The PG test was performed with the three differential cultivars. Two additional cultivars, Q2 (resistant to PG2 isolates) and Defender (moderately resistant to PG2 isolates), were included for comparisons. Twelve 7-day-old cotyledons of each differential cultivar grown in Metro Mix were wound inoculated with a 10- μ l droplet of pycnidiospore suspension (1×10^7 pycnidiospores per ml). Inoculated cotyledons were maintained in the greenhouse (16/21°C night/day and a 16-h photoperiod). The experiment was repeated twice. Disease severity on cotyledons was assessed 12 days postinoculation by using a 0 to 9 scale (2). All five isolates from Roland and East Selkirk were highly virulent on Glacier (6.4 to 7.7), Q2 (7.1 to 8.2), and Defender (7.2 to 8.4), but intermediately virulent on Quinta (4.5 to 5.4). This clearly indicated that these isolates were of PG3. Isolates of PG2 have been predominant in Manitoba for the past 25 years, and highly virulent isolates belonging to PG3 had not been detected previously. To our knowledge, this is the first report of the presence of PG3 in *L. maculans* in Manitoba.

References: (1) A. Mengistu et al. Plant Dis. 75:1279, 1991. (2) P. H. Williams. Crucifer Genetics Cooperatives (CrGC) Resource Book, University of Wisconsin—Madison, 1985.

First Report of Brown Rot in Wine Grapes Caused by *Monilinia fructicola* in Canada. P. L. Sholberg and P. D. Haag, Agriculture and Agri-Food Canada, Pacific Agri-Food Research Centre, Summerland, BC, V0H 1Z0, Canada; and S. Hambleton and H. Boulay, Agriculture and Agri-Food Canada, Biodiversity (Mycology and Botany), Ottawa, ON, K1A 0C6, Canada. Plant Dis. 87:1268, 2003; published on-line as D-2003-0808-03N, 2003. Accepted for publication 30 July 2003.

A survey was conducted in 2001 and 2002 to determine incidence of fruit pathogens in wine grapes (*Vitis vinifera*), an important crop in the southern interior of British Columbia (BC), Canada. Grape clusters were sampled every 2 weeks from June to October at eight vineyard sites located from Osoyoos in the south to Kelowna, approximately 100 km to the north. In the laboratory, the berry clusters were surface disinfested for 0.5 min in 70% ethanol, followed by 1 min in 0.5% sodium hypochlorite, and rinsed twice in sterile distilled water. The berries were placed on potato dextrose agar (PDA) amended with 15 ml/liter of 85% lactic acid and incubated at 20°C for 1 week. During the 2002 survey, a fungus resembling *Monilinia fructicola* (G. Wint.) Honey was observed sporulating on immature 'Pinot noir' grapes from Kelowna that were sampled on 14 August. Later in the growing season, a similar fungus was detected on 'Riesling' grapes from Summerland sampled on 11 September. There was no evidence of brown rot near the vineyard in Kelowna, but diseased stonefruit were present near the vineyard in Summerland. Subsequent identification of the fungus from 'Riesling' as *M. fructicola* was based on morphological characters and DNA sequence data for the internal transcribed spacer (ITS) regions of the nuclear ribosomal rRNA genes. The sequenced isolate was deposited in the Canadian Collection of Fungus Cultures as DAOM 231119, and the ITS sequence was accessioned in GenBank as AY289185. Colony growth on PDA was rapid and in concentric rings with the colony margin complete, microconidia abundant, and macroconidia 12 to 13 μ m long. Macroconidia germinated with a long germ tube before branching. These characteristics distinguished this fungus from *M. laxa*, a closely related species that is slow growing with lobed colony margins, produces few microconidia, and germ tubes that branch close to the conidium (1). The complete ITS sequence for DAOM 231119 was a 100% match to other sequences deposited for *M. fructicola* (Z73777, AF010500, and U21815). On the basis of comparisons of available data, ITS sequences for *M. fructicola* (three complete ITS, seven partial ITS) and *M. laxa* (8 complete ITS, 10 partial ITS) differed consistently at four nucleotide positions. The fungus identified as *M. fructicola* was tested for pathogenicity on mature surface-sterilized 'Pinot noir' and 'Riesling' grapes. Under humid conditions, buff-colored sporodochia bearing conidia developed over the surface of the infected berries. This indicates that *M. fructicola* can cause decay of wine grapes and could be confused with bunch rot caused by *Botrytis cinerea*. Previously, *M. fructicola* was reported on grapes in Oklahoma, but likely these grapes were not *Vitis vinifera* (2). To our knowledge, this is the first report of brown rot caused by *M. fructicola* on wine grapes in North America.

References:(1) L. R. Batra. World Species of *Monilinia* (Fungi): Their Ecology, Biosystematics and Control. Mycologia Memoir No. 16. Gerbrüder Borntraeger, Berlin/Stuttgart, 1991. (2) D. A. Preston. Host Index of Oklahoma Plant Diseases, Tech. Bull. No. 21. Oklahoma Agricultural and Mechanical College, Agricultural Experiment Station, Stillwater, 1945.