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DISEASE NOTES OR NEW RECORDS

Cyclamen soft rot caused by *Erwinia chrysanthemi* in Argentina

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Abstract. Cyclamen plants from a commercial greenhouse were affected by a severe wilt. Symptoms began with a foliar yellowing, ending with the death of affected plants. Corms had internal rotten areas. The pathogen was identified as *Erwinia chrysanthemi*. Symptoms were reproduced on artificial inoculations. Bacteria did not need wounds to penetrate roots or corms.

Because of its winter blooming, cyclamen (*Cyclamen persicum*, Primulaceae) is one of the main flowering potted plants produced in commercial greenhouses in Argentina. There are maxi (traditional large), midi and mini type F1 hybrids (Dole and Wikins 1999).

During the winter of 2001, cyclamen plants of the mini type from a commercial greenhouse located in Nono, Córdoba Province, Argentina, were affected by a severe wilt. Symptoms began with a yellowing of the foliage, followed by the collapse of the petioles and the necrosis of the leaves, ending with the death of affected plants. There were no external symptoms on the corms but, in transverse sections, they had internal rotten areas and the vascular system appeared necrotic (Fig. 1). Wilt symptoms did not resemble those of any disease described for cyclamen in Argentina. The objectives of the present work were to identify the soft-rot causing agent, and to determine its possible ways of penetration.

Corms were carefully washed with running tap water, superficially disinfected with 70% ethanol and cut in half with a sterile scalpel. Isolations were made by either placing small internal pieces of corm tissue on potato-dextrose agar (PDA), or by macerating tissues on sterile distilled water and streaking on nutrient agar (NA) plates. Plates were incubated at 25°C for 48 h. Bacteria were consistently isolated on PDA and NA. Individual colonies from the NA plates were

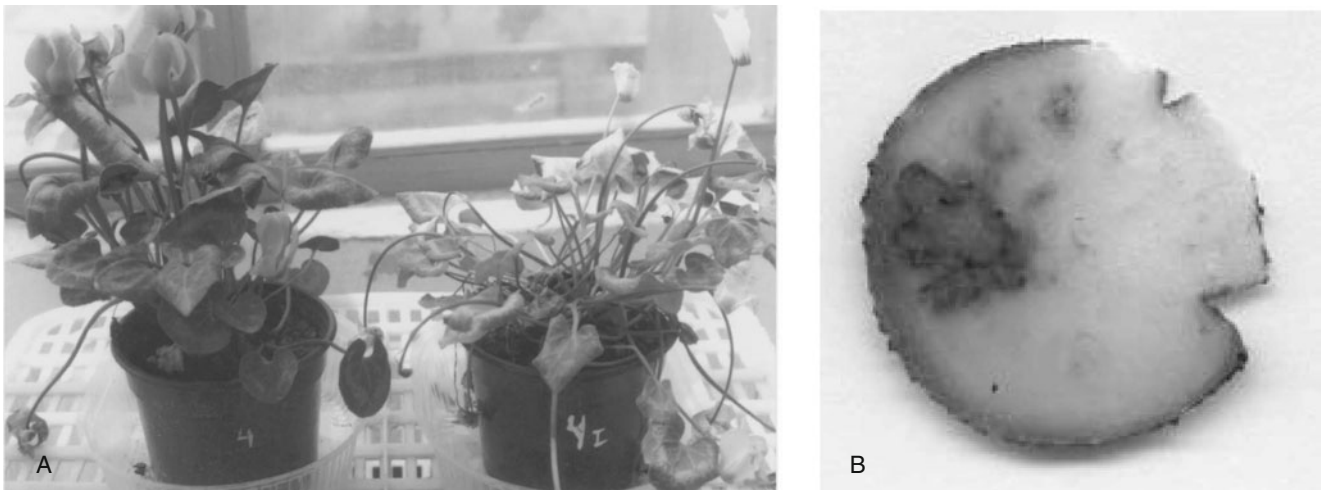

Fig. 1. (*A*) Leaf yellowing and wilt of *Cyclamen persicum* plants affected by *Erwinia chrysanthemi*. (*B*) Transverse cut of a corm showing necrosis of the vascular system with internal rotten areas.

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re-streaked to ensure purity. To identify the causal agent, tests described by Schaad (1988), Dickey (1979) and Janse and Ruissen (1988) were performed.

Pathogenicity tests were conducted on healthy cyclamen plants of the mini type using a suspension (10^7 cfu/mL) of the isolated bacterium on undamaged or wounded tissues (roots, corms or petioles). Roots were watered with the bacterial suspension after clipping their tips with scissors; corms were inoculated by opening a 3-mm-deep hole with a sterile scalpel and swabbing with a piece of cotton wool dipped in the inoculum suspension; petioles were scraped with a sterile scalpel and sprayed with the bacterial suspension. Inoculum was also applied onto undamaged roots, corms or petioles as described. Control plants were treated in the same way, but were watered, swabbed or sprayed with sterile distilled water. For each treatment, three plants in individual pots were used (12 treatments $\times$ 3 replicates = 36 plants). All plants were kept in plastic bags for 48 h after inoculation, and maintained in the greenhouse throughout the experiment. Symptoms were evaluated 3 weeks after inoculation. To verify the cause of the symptoms observed, the pathogen was re-isolated from symptomatic plants.

The isolated bacteria produced round, mucoid, convex, white colonies on nutrient agar. Bacteria were bacilliform and Gram negative. They did not fluoresce on King B medium, and did not produce urease or degrade starch. Strains were catalase positive, oxidase negative, facultative anaerobic and produced gas from glucose. They hydrolysed pectate, reduced nitrate, produced indole and the bacterium was sensitive to erythromycin (15 μg). Gelatine was liquefied after 14 days of incubation.

Three weeks after inoculation, all plants inoculated on the roots or corms with, or without, wounds had a wilt associated with corm rot. Only one plant inoculated on a wounded petiole was wilted. Plants inoculated on undamaged petioles remained healthy throughout the experiment. None of the control plants had wilt symptoms. Bacteria isolated from symptomatic inoculated plants gave the same reactions as the original strains, confirming Koch's postulates.

Based on the cultural, physiological and biochemical reactions performed, the bacterium was identified as *Erwinia chrysanthemi* (Schaad 1988). The symptoms observed and the characteristics of the isolated bacteria also correspond to those described by Daughtrey *et al*. (1995). This pathogen causes soft rots on several ornamental crops worldwide (Daughtrey *et al*. 1995; Janse and Ruissen 1988). Symptoms can be confused with those incited by *E. carotovora* subsp. *carotovora*, which can also cause crown, leaf and flower rot on cyclamen (Dole and Wikins 1999). *E. chrysanthemi* differs from *E. carotovora* by its sensitivity to erythromycin and its ability to produce gas from glucose (Daughtrey *et al*. 1995). Also, indole production is negative for *E. carotovora* (subspp. *carotovora*, *atroseptica* and *betavasculorum*), *E. cypripedii* and *E. rhapontici*, and is considered variable for *E. chrysanthemi* (Dickey 1979; Schaad 1988). However, in a thorough analysis of *E. chrysanthemi* considering the original host of the strains, Dickey (1979) determined that all *E. chrysanthemi* strains were indole positive, except for strains isolated from *Syngonium podophyllum*. Thus, indole reaction can also be considered as a key difference between *E. chrysanthemi* and other Erwinias.

An isolate with similar characteristics was obtained from wilted cyclamen plants from another crop located in Pablo Nogués (Buenos Aires Province).

To our knowledge, this is the first report of *Erwinia chrysanthemi* affecting cyclamen in Argentina. An isolate was lodged in the Banco Nacional de Microorganismos, Cátedra de Microbiología, Facultad de Agronomía, Universidad de Buenos Aires, Argentina as BNM-0271.

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