

DISEASE NOTES OR NEW RECORDS

Occurrence of *Cladosporium herbarum* on wheat leaves (*Triticum aestivum*) in Argentina

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Abstract. During the course of recent surveys in the wheat cropping area of Buenos Aires and Entre Ríos Provinces, Argentina, an unusual disease was found on wheat leaves. A fungus was isolated from the symptomatic tissues and identified as *Cladosporium herbarum* (Pers.:Fr) Link. Inoculations were carried out under greenhouse conditions to test pathogenicity and fulfil Koch's postulates. The symptoms of the disease and the causal agent are described.

Cladosporium Link.Fr was reported on wheat (Wiese 1985) as a common and mild parasite affecting dead or half-dead plant tissues in association with some other fungi. It often appears on the ear heads causing a greenish-black, mouldy growth on the affected parts (Farr *et al.* 1989). Wiese (1985) reported that it did not cause severe symptoms on leaves and stems of wheat but there are some reports indicating that moist and shady conditions could favour the occurrence of outbreaks of the disease on leaves (Arya and Panwar 1955).

During the last 5 years, leaf spot symptoms on wheat cultivars Buck Pingo, B. Biguá, B. Brasil and B. Poncho growing in the area of northeast Buenos Aires Province were commonly observed. On most plants, leaves showed symptoms sufficiently similar to those described for the complex of necrotrophic foliar pathogens (*Drechslera tritici-repentis*, *Septoria tritici*, *Alternaria triticumaculans* and *Bipolaris sorokiniana*) to suggest that any of these might have been involved. *C. herbarum* (Pers.:Fr) Link was previously reported in Argentina (Marchionatto 1948) on leaves and spikes of wheat and other grasses without describing the symptoms in detail. Since then, there have been no other reports of this disease on wheat leaves in Argentina.

Infected leaf samples were collected during an extensive survey conducted from September–November 2002. Samples were collected from different cultivars in farmers' fields and one Experimental Research Station across the wheat region of Buenos Aires and Entre Ríos Provinces, in five of the eight sites surveyed (Los Hornos, Nogoyá, Olavarría, Tandil, Victoria) (Fig. 1).

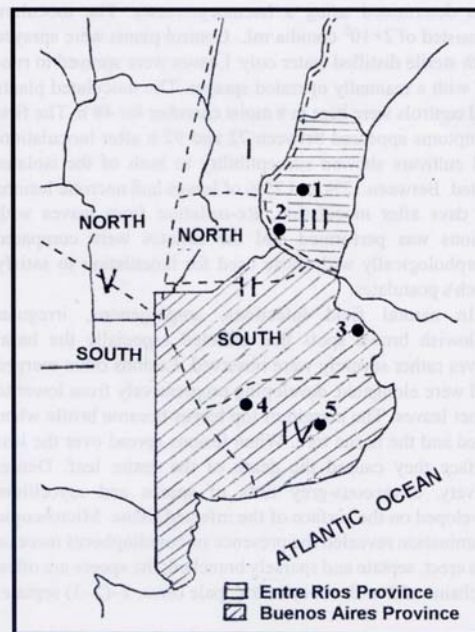


Fig. 1. Sites of the wheat area surveyed in 2002 where *Cladosporium herbarum* was detected. 1: Nogoyá; 2: Victoria (wheat area III); 3: Los Hornos (wheat area II South); 4: Olavarría; and 5: Tandil (wheat area IV).

Diseased leaves were collected, stored in paper bags and transported to the laboratory. The pathogen was isolated from typical necrotic symptoms. Small sections of diseased leaves were disinfested in 70% ethanol and 0.1% mercuric chloride for 1 min, then rinsed twice in sterile distilled water and placed on 2% potato-dextrose-agar (PDA). Petri dishes were maintained at 22°C. Cultural studies of the fungus were conducted on single-spore colonies grown in Petri dishes containing PDA, cultured at 20–22°C under cool-white fluorescent light supplemented with near UV with a 12 h photoperiod.

Inoculation experiments to confirm pathogenicity were performed in the greenhouse at 15–25°C and 80% relative humidity. Fifteen plants of each of the cultivars Buck Biguá, B. Brasil, B. Pingo and B. Poncho were grown in plastic pots (12-cm-diameter) with a standard potting mix. Plants were inoculated when they had reached the third expanded leaf stage and heading stage. Inoculum was prepared from 10-day-old cultures of *C. herbarum* (isolates Ch101 and Ch500) growing on PDA and was obtained by flooding each sporulating plate with sterile distilled water and gently scraping the fungal colony with a flame-sterilised scalpel to dislodge conidia. The conidial suspension was filtered once through a single layer of cheesecloth and spore concentration was determined using a haemocytometer. The inoculum consisted of 2×10^5 conidia/mL. Control plants were sprayed with sterile distilled water only. Leaves were sprayed to runoff with a manually operated sprayer. The inoculated plants and controls were kept in a moist chamber for 48 h. The first symptoms appeared between 72 and 92 h after inoculation. All cultivars showed susceptibility to both of the isolates tested. Between 12% and 75% of leaves had necrotic lesions 10 days after inoculation. Re-isolation from leaves with lesions was performed and the isolates were compared morphologically with those used for inoculation to satisfy Koch's postulates.

In natural field infections, amphigenous, irregular yellowish brown spots that affected especially the basal leaves rather severely were observed. Lesions often merged and were elongated, developing progressively from lower to upper leaves. The margin of top leaves became brittle when dried and the tissue tore. When lesions spread over the leaf surface they caused the death of the entire leaf. Dense, velvety, olivaceous-grey tufts of spores and mycelium developed on the surface of the infected tissue. Microscopic examination revealed the presence of conidiophores more or less erect, septate and sparsely branched; the spores are often in chains of 2 or 3, subcylindric, pale olive, 1–(2–3) septate,

10–15 × 4–7 µm. The teleomorph, *Mycosphaerella tulasnei* was not seen.

All wheat plants inoculated with *C. herbarum* in the greenhouse developed symptoms identical to those observed on naturally infected plants in the field. No differences in degree of infection were noted among the cultivars. Nevertheless, adult plants showed more severe symptoms than younger ones. No symptoms were observed in control non-inoculated plants. Isolation from symptomatic tissue has consistently yielded cultures of *C. herbarum*. The fungus sporulated on the diseased tissue in the Petri dishes. Comparison of morphological characteristics of *C. herbarum* isolates revealed no differences between field and glasshouse produced spores in the shape and size of conidia.

The isolates of *C. herbarum* have been lodged in the culture collection of the CIDEFI (Centro de Investigaciones de Fitopatología), Facultad de Ciencias Agrarias y Forestales de la Universidad Nacional de La Plata, Buenos Aires, Argentina, with the accession numbers 111-01, 209-02, 210-02, 212-02 and 215-02.

Inoculation studies proved that *C. herbarum* was the cause of this outbreak on wheat in Argentina. In the last few years, the increased incidence of the disease may be related to new cultural practices (reduced tillage, nitrogen fertilisation, irrigation), the use of new germplasm and favourable weather conditions. This contributed to a major spread, not only of *C. herbarum*, but also of the foliar complex of necrotrophic pathogens in general.

The fact that other wheat cultivars apart from those checked may also be susceptible to the pathogen, shows the importance of conducting thorough research to determine the reactions of those cultivars currently used in the Argentina cropping area.

References

- Arya HC, Panwar KS (1955) Some studies on a virulent strain of *Cladosporium herbarum* (Link) Fr. on wheat. *Indian Phytopathology* **8**, 176–183.
- Farr DF, Bills GP, Chamuris GP, Rossman AY (1989) 'Fungi on plants and plant products in the United States.' (APS Press: St Paul, Minnesota, USA)
- Marchionatto JB (1948) 'Tratado de Fitopatología.' (Ediciones Librería del Colegio, Editorial Sudamericana)
- Wiese MV (1985) 'Compendium of wheat diseases.' (APS Press: St Paul, Minnesota)

Accepted 13 February 2003