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New apiaceous hosts of *Sclerotinia sclerotiorum* in the Cerrado region of Brazil

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ABSTRACT

This paper reports for the first time in the Cerrado region of Brazil the occurrence of white mold caused by *Sclerotinia sclerotiorum* in coriander (*Coriandum sativum*), parsley (*Petroselinum crispum*) and Peruvian carrot (*Arracacia xanthorrhiza*). The disease was observed on coriander, in Cristalina, Goiás State, on parsley in Gama, the Federal District and on Peruvian carrot in Ibiá, Minas Gerais State. Pathogenicity tests demonstrated that the fungus *S. sclerotiorum* is the causal agent of the observed symptoms in these plants. The three isolates obtained from naturally infected plants were inoculated in coriander (cv. Verdão), parsley (cv. Lisa Gigante), Peruvian carrot (cv. Amarela de Senador Amaral) and carrot (cv. Forto Nantes) and they were pathogenic to these hosts. The fungus isolates were re-isolated from inoculated plants fulfilling Koch´s postulates and also confirming that *S. sclerotiorum* is a polyphagous pathogen.

Keywords: *Arracacia xanthorrhiza*, *Coriandrum sativum*, *Petroselinum crispum*, white mold, etiology.

RESUMO

Novas hospedeiras de *Sclerotinia sclerotiorum* na família Apiaceae na região do Cerrado do Brasil

Nesta publicação relata-se, pela primeira vez, na região de cerrado do Brasil, a ocorrência de mofo-branco, causado por *Sclerotinia sclerotiorum* em coentro (*Coriandum sativum*), salsa (*Petroselinum crispum*) e mandioquinha salsa (*Arracacia xanthorrhiza*), três espécies de hortaliças pertencentes à família Apiaceae. A doença foi detectada em coentro no município de Cristalina-GO, em salsa na cidade satélite do Gama-DF e em mandioquinha salsa no município de Ibiá-MG. Os testes de patogenicidade foram realizados em coentro, salsa e em mandioquinha salsa e o fungo foi reisolado das plantas inoculadas, completando-se assim os postulados de Koch. Quando os três isolados foram inoculados nas três espécies de plantas e também em cenoura (cv. Forto Nantes), todos os três isolados foram patogênicos, demonstrando a falta de especificidade por hospedeira de *S. sclerotiorum*.

Palavras-chave: *Arracacia xanthorrhiza*, *Coriandrum sativum*, *Petroselinum crispum*, mofo-branco, etiologia.

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The Apiaceae botanical family comprises several vegetable crops and some species used as medicinal plants or condiments. Among them the most important vegetable crops are carrot (*Daucus carota*), parsley (*Petroselinum crispum*), celery (*Apium graveolens*), Peruvian carrot (*Arracacia xanthorrhiza*) and coriander (*Coriandrum sativum*).

White mold, caused by the soil-borne fungus *Sclerotinia sclerotiorum* (Lib.) de Bary, is an important disease of cultivated plants and also of some weeds. The fungus *S. sclerotiorum* is a polyphagous species that has already been reported as a pathogen of more than 400 plant species around the world (Bolland & Hall, 1994). In Brazil there is a long list of hosts of the fungus *S. sclerotiorum* and it includes many vegetable crops, such as carrot, potato, tomato, green pepper, eggplant, lettuce and some brassicaceous species (Mendes *et al*., 1998).

White mold is a very serious problem in vegetable crops, especially when they are cultivated in contaminated wet soils and the weather is cool and wet (Reis & Lopes, 2007). The optimum temperature for disease development ranges from 15 to 21º C. The resistant structure of the pathogen, the sclerotium, can survive for a long time in the soil, needing high soil and air humidity and free water on the plants for germination and to infect its hosts (Lobo Júnior, 1999; Lobo Júnior *et al*., 2000).

Sclerotium germination can be either myceliogenic or carpogenic. In myceliogenic germination the mycelium is produced directly from the sclerotium and in carpogenic germination a fructification structure, the apothecium, is produced by the sclerotium. For the occurrence of carpogenic germination the sclerotium must receive enough light to deliver the stipes and develop the apothecia. Without enough light germination will be only myceliogenic and the mycelium will be able to penetrate the healthy tissues of the host plants when in contact with them (Tu, 1989).

In areas free of the pathogen when inoculum is not already present in the soil, an epidemy of white mold can be started by seeds that are either infected by the dormant fungal mycelium or contaminated by sclerotia that have been transported together with the seed lot. Dormant mycelia, present in the tegument and in the cotyledons of seeds, can remain viable for more than three years. When seeds are sown and, if the temperature and humidity are favorable, mycelia can develop and initiate a new infection cycle. Some contaminated seeds may not germinate but can

produce new mycelia and sclerotia (Tu 1998).

In all host plants the chemical control of white mold is hindered by the difficulty of reaching the infection sites of the fungus near the soil, because the pathogen is covered by the plant's leaves and stems. The wide host range of the pathogen makes crop rotation less successful in diseased management if fields are already contaminated with *S. sclerotiorum* (Reis *et al*., 2007). The non-host crops are practically restricted to gramineous plants, which may not be economically viable for the growers (Lobo Júnior, 1999).

This research had the aim of studying and reporting the etiology of crown and stem rot on Peruvian carrot, coriander and parsley plants in the Cerrado region of Brazil.

MATERIAL AND METHODS

Plant samples and pathogen isolation - In winter 2003 symptoms of white mold, crown rot, and stem rot, with the presence of sclerotia were observed on coriander plants (cv. Verdão) (Figure 1A). These symptoms were observed on coriander plants cultivated under central pivot in Cristalina, Goiás State. The coriander field had been planted to produce commercial seeds. In 2008, parsley (Figure 1B) and Peruvian carrot (Figure 1C) plants were observed with the same symptoms in the region of Gama, Federal District and in Ibiá, Minas Gerais State, respectively. Samples of those plants were taken to the Plant Pathology Laboratory of Embrapa Hortaliças, where the fungus was isolated in PDA medium amended with the antibiotic rifampicin (50 ppm).

Pathogenicity test - The pathogenicity test was carried out on the three host plant species with the respective isolate. The test plants used were the Peruvian Carrot cultivar Amarela de Senador Amaral (Embrapa Hortaliças), the coriander cultivar Verdão (Feltrin) and the parsley cultivar Lisa Gigante (Sakata). Because of the different cultivation cycles, parsley plants were inoculated 50 days after sowing, coriander plants 30 days after sowing and Peruvian carrot 60 days after planting. For the inoculation, one mycelium disc of the fungus (15 mm diameter) was taken from a seven day-old fungus colony in PDA. The mycelium discs were attached to the crown region of the test plants with the aid of a sterilized tooth pick. Control test plants received only one PDA disc, attached to the crown by a sterilized tooth pick. After inoculation, plants were kept in wet chambers putting a wet plastic bag on the plants for 48 h. During this period the pots were kept under the greenhouse benches to avoid direct exposure to the sun's rays and the increase in temperature into the wet chamber. Subsequently, plants were kept over the benches of the greenhouse during ten days. The complete experiment consisted of three treatments with four replicates. Each replicate was represented by one pot with two plants, cultivated in sterilized soil. Evaluation was carried out 10 days after inoculation when the presence of characteristic symptoms of white mold and formation of sclerotia on the lesions was observed. Three days before the evaluation, the symptomatic plants were put in a wet chamber again to stimulate the formation of white mold and fungal sclerotia on the lesions, due to the very low air humidity in the greenhouse (under 40%). After evaluation the pathogen was isolated in pure culture again. In addition, a second test was carried out where the three pathogen isolates were cross inoculated on the three plant hosts plus carrot plants (cv. Forto Nantes). This experiment was conducted in a factorial design 3 x 4 x 3 (three pathogen isolates, four plant species and three replicates).

RESULTS AND DISCUSSION

Isolates from naturally infected coriander, parsley, and Peruvian carrot produced fungal colonies that grew fast and were white in color on PDA medium. A number of sclerotia developed around the edges of dishes with these colonies after eight to 12 days of growth, under 12 hours of light. Sclerotia were beige colored at the beginning, turning black after some days of growing; they presented a large range of sizes (Figure 1D). The isolate from coriander presented sclerotia whose length ranged from 3.5 to 12.1 mm; the isolate from parsley presented length of sclerotia ranging from 2.5 to 13.2 mm; and the isolate from Peruvian carrot presented sclerotia length ranging from 3.9 to 10.3 mm. Under the microscope we observed that hyphae were septate and multinucleated. These characters are in accordance with those reported for the fungal species *S. sclerotiorum* (Kohn, 1979).

Three to four days after inoculation, test plants began to present symptoms of water-soaked lesions around the point of inoculation. The coloration of the lesions was initially beige, turning brown later. Ten days after inoculation most parsley plants (80%), coriander (80%) and Peruvian carrot (70%) had already died. Fragments of dead plants, when kept in a moist chamber, developed white mycelia and sclerotia typical of *S. sclerotiorum*. Control plants did not develop disease symptoms.

In the cross-inoculation assay all three isolates infected all three inoculated host plants and the carrot. In all tested plants the same symptoms were observed as described above. This is in accordance with other authors, who have reported the wide host range of this pathogen and its lack of host specificity (Farr *et al*., 1989; Bolland & Hall, 1994; Mendes *et al*., 1998; Reis *et al*., 2007). The wide host range of the pathogen has epidemiological implications and makes the management of white mold very difficult, especially in terms of crop rotation (Reis *et al*., 2007).

The pathogen was re-isolated from all inoculated plants, fulfilling Koch's Postulates. The fungal species *S. sclerotiorum* is reported in the literature as a pathogen of parsley in other countries (Anonymous, 1960; Bolland & Hall, 1994; Farr *et al*., 1989; Chang & Kim, 2003), but to the best of our knowledge this is the first report in Brazil. No other reports have been found of *S. sclerotiorum* causing white mold on coriander in Brazil or in other countries. For Peruvian carrot, this disease has already been reported

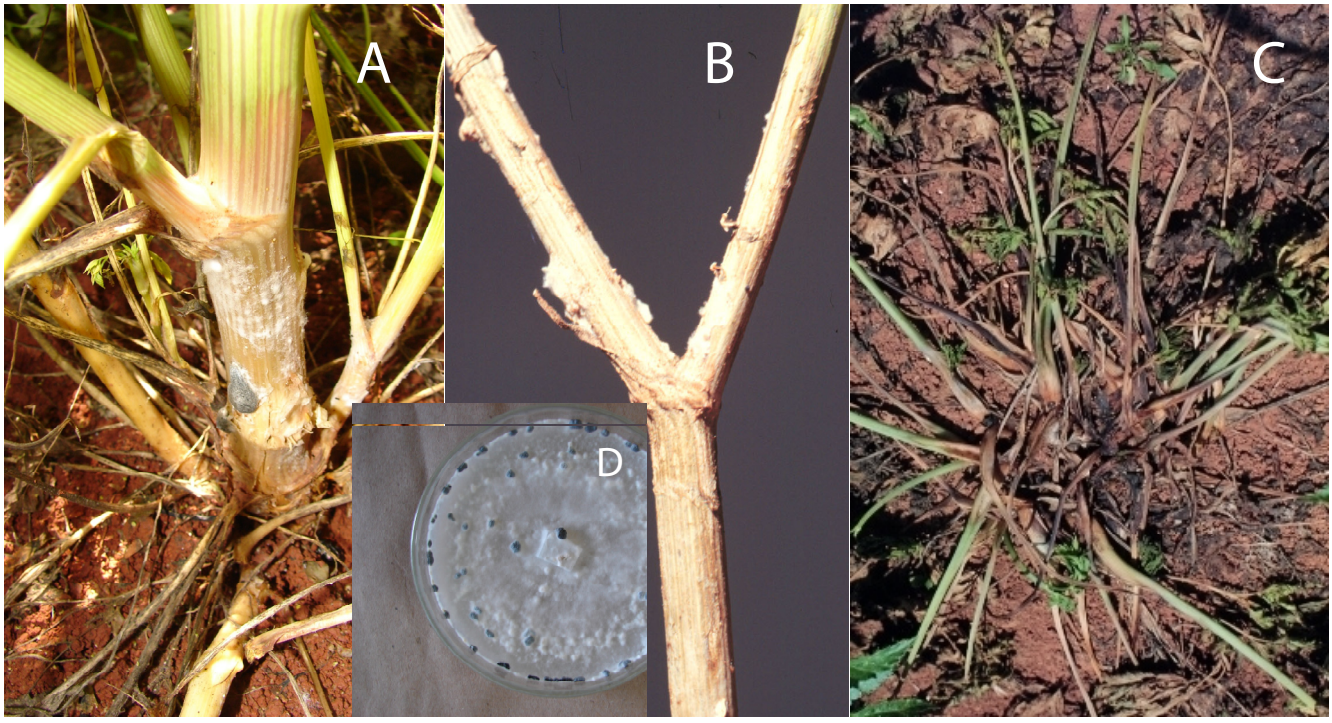


Figure 1. Symptoms of white mold caused by *Sclerotinia sclerotiorum* on coriander (A), parsley (B), and Peruvian carrot (C) and 14-day-old fungus culture in PDA (D) [sintomas do mofo-branco, causado por *Sclerotinia sclerotiorum*, em coentro (A), salsa (B) e mandioquinha salsa (C) e cultura do fungo em BDA com 14 dias de idade (D)]. Brasília, Embrapa Hortaliças, 2010.

in Espirito Santo State (Costa *et al*., 1987; Ventura & Costa, 1998) and in Peru (Barrantes, 1998); however, this is the first report for the Cerrado region of Minas Gerais State.

In this paper, we report for the first time the occurrence of white mold on coriander and parsley in Brazil, extending the host range of *S. sclerotiorum*. This report contributes to more information about the epidemiology of the disease and can be useful in implementing effective management strategies for the disease.

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