

A NEW REPORT OF *PYTHIUM OLIGANDRUM* FROM PAKISTAN

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Abstract

Pythium oligandrum a well known mycoparasite of pathogenic Oomycetes and other fungal plant pathogens has been isolated for the first time from Pakistan. *P. oligandrum* is characterized by haustoria or hook shaped hyphae, toruloid or sub-globose sporangial structures, intercalary ornamented or spiny oogonia, aplerotic oospores and persistent antheridia.

Introduction

Several genera of Oomycetes contain economically important plant pathogens. Of these *Phytophthora* and *Pythium* include some of the world's most destructive plant pathogens which account collectively for multibillion dollar losses in world cash crops (van West *et al.*, 2003). However, some species in the genus *Pythium* can be beneficial, functioning as biological control agents protecting against phytopathogenic *Pythium* species (Martin & Loper, 1999). The well known oomycetous mycoparasite *P. oligandrum* Drechsler is capable of suppressing various soilborne oomycetes and fungal plant pathogens (Al-Rawahi & Hancock, 1998; Benhamou *et al.*, 1997, 1999; Madsen & de Neergaard, 1999; Paul, 1999; Picard *et al.*, 2000; Ribeiro & Butler, 1995).

During the present studies on Oomycetous fungi of Sindh, *P. oligandrum* was isolated from the rhizosphere of cucurbits growing in Malir, Karachi. The culture has been deposited at the Karachi University Culture Collection (KUCC) as KUCC-OOP-03018. This isolate closely resembles the *P. oligandrum* Drechsler with few exceptions. It appears to be the first report of *P. oligandrum* from Pakistan (Sultan *et al.*, 1997), which is described and illustrated herein.

Materials and Methods

Soil samples were collected at random from a depth of 0-5 inches of soil. Samples from a field were mixed to obtain composite sample. Isolation was done by baiting technique (Harvey, 1925). Soil sample kept in a polyethylene bag was moistened by the addition of sterile water and mixed thoroughly to get a paste like consistency. With the help of a sterilized teaspoon, the soil was placed at one side in each of three sterilized Petri plates and 10-15 ml sterilized water was added. Two grass blades (3 cm long) were placed in each Petri plate, one near the soil and the other away from the soil. The Petri plates were incubated at room temperature. After 5-8 days faint halo of fungal threads were observed on the baits. The baits were rinsed in sterilized water to remove soil particles, and placed into fresh sterilized Petri plates half-filled with sterile water and new fresh baits were added. After 2 days of incubation, the baits colonized by Oomycetous fungi were transferred on the corn-meal agar (CMA) medium amended with 100 ppm PCNB for purification. Radial growth and colony characteristics were observed on potato

dextrose agar (PDA), corn-meal agar (CMA), potato-carrot agar (PCA) (Plaats-Niterink, 1981) and corn-meal dextrose agar (CMDA) (Lodhi *et al.*, 2004) at 25°C. Water culture of fungi was prepared by adding an inoculum disc and grass blades to sterile water in a Petri plate and incubating it at 25°C. After the production of sporangia, zoospore and sexual structures fungi were identified after reference to Sparrow (1960) Plaats-Niterink (1981) and Dick (1990). Biometric values viz., aplerotic index, ooplast index and wall index were determined after Shahzad *et al.*, (1990).

Taxonomic Description

Morphological characteristics: Main hyphae up to 7µm wide. Tips of the hyphae haustoria are hook shaped. Sporangia consist of irregular complexes of filamentous toruloid, swollen and sub globose structures. Sporangia when globose 12x20 µm in diameter and toruloid structures up to 38x12 µm in size. Encysted zoospores up to 10 µm. Oogonia mostly intercalary occasionally terminal, ornamented, spiny (19-)22-24(-27) (av. 23) µm in diam. Spines 4-6 µm long and 2-3 µm wide. Antheridia 1-2 per oogonium, persistent, even with aborted oogonia, mostly declinous and lateral in attachment. Oospores aplerotic, smooth, 17-23 (av. 20.5) µm in diam. Ooplast 10 µm in diam. Oospore wall 1-2 (av. 1.6) µm thick (Fig. 1).

Biometric value: Aplerotic index 71%, Wall index 40% and Ooplast index 20%.

Colony characteristics: The *P. oligandrum* produces submerged mycelial growth with distinct stellate pattern on PDA submerged growth with indistinct chrysanthemum/stellate pattern on PCA, on CMA indistinct chrysanthemum pattern with submerged growth and on CMAD chrysanthemum pattern with superficial mycelial growth.

Daily growth rate at 25 °C on PDA 3.7 mm, PCA 5.5 mm, CMA 5.5 and CMAD 5.7 mm.

Discussion

P. oligandrum was first isolated by Drechsler (1930) from diseased peas in USA. *P. oligandrum* is known to have multiple behaviors in its nature. It is either common soil fungus, or a strong or weak pathogen of a number of plant species or a biological control agent against the number of plant pathogenic fungi. It is recorded as a common soil fungus from Central Africa (Kobayashi *et al.*, 1977), South Australia (Vaartaja & Bumbieris, 1964 a, b), Hawaii (Klemmer & Nakano, 1964), USA (Hendrix & Campbell, 1970), Germany (Domsch *et al.*, 1968) and Netherlands (Plaats-Niterink, 1975). It is also known to cause crown-rot and damping-off of rhubarb and weakly pathogenic to a number of other plant species (Middleton, 1950, 1952). As a bio-control agent, *P. oligandrum* is an aggressive hyperparasite of a number of fungi such as *P. ultimum*, *P. debaryanum*, *Aphanomyces laevis*, *Phialophora radicola*, *Botryotrichum piluliferum*, *Gaeumannomyces graminis*, *Streptomyces* sp., *Rhizoctonia solani*, *Fusarium culmorum*, *Trichoderma viride* and *T. koningii* (Vesely, 1978 a, b; Deacon, 1976).

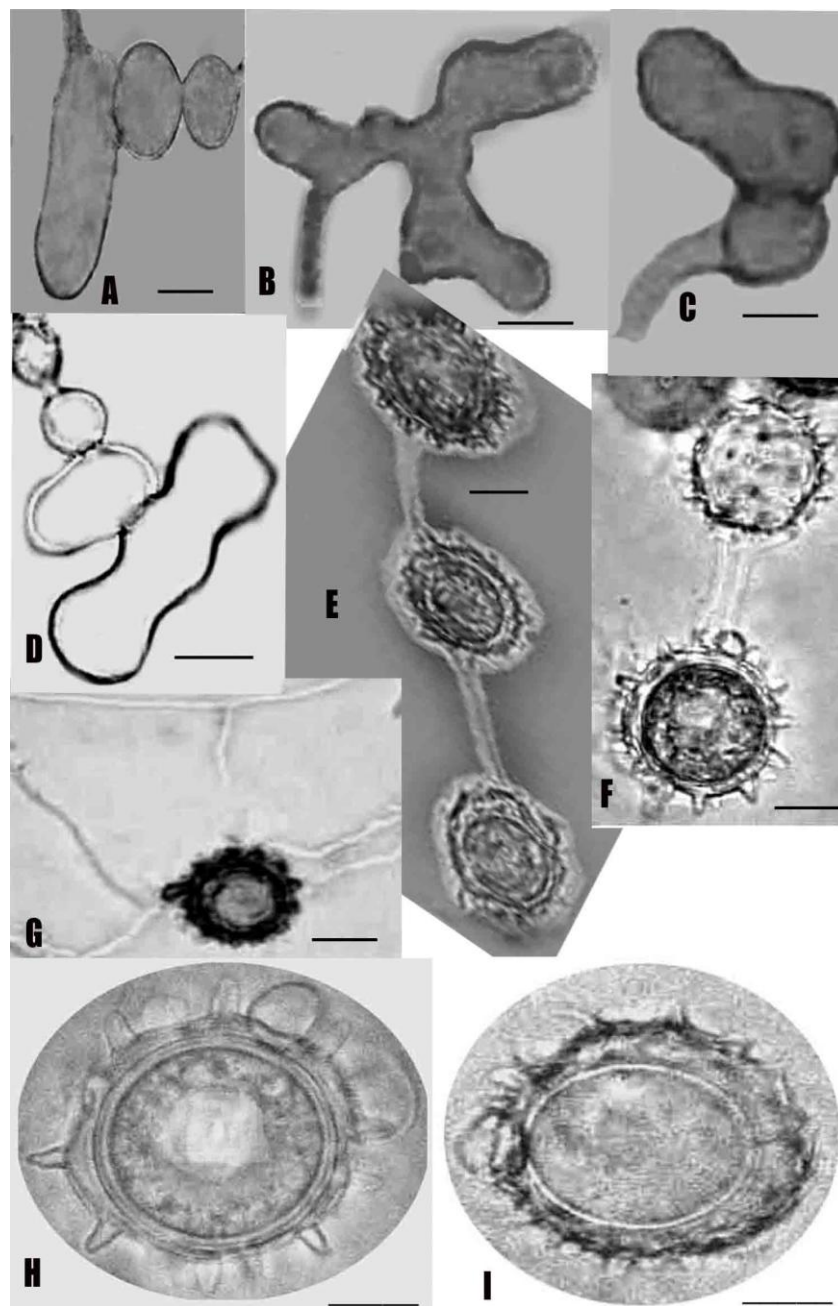


Fig. 1. A-C, Filamentous toruloid and sub globose sporangial structures; D, Empty sporangia; E-F, Ornamented, intercalary oogonia and aplerotic oospore; G, Oogonium with declinuous antheridia; H, Oogonium with two laterally attached persistent antheridia; I, Persistent antheridia with aborted oogonia and aplerotic oospore.
Scale bar = 10 μ m

There are two species of *Pythium* that have non-proliferating sporangia consisting of sub globose, elongated, or toruloid structures and ornamented oogonia less than 30µm in diam., with oospores diameter less than 25 µm. They are *P. oligandrum* and *P. acanthicum*. *P. acanthicum* differs with our isolates by its terminal oogonia, small oogonial spines, plerotic oospores and antheridial nature.

All characteristics of our isolates are in accordance with the description of *P. oligandrum* given by Plaats-Niterink (1981). Moreover according to Plaats-Niterink (1981) the antheridia are mostly lacking in *P. oligandrum*, whereas, in our isolate antheridia are always present with oogonium.

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