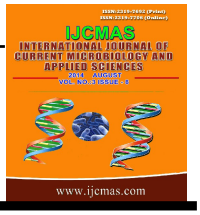
Int.J.Curr.Microbiol.App.Sci (2014) 3(8) 296-302

International Journal of Current Microbiology and Applied Sciences
ISSN: 2319-7706 **Volume 3 Number 8 (2014) pp. 296-302**
http://www.ijcmas.com



Original Research Article

Studies on Rhizome rot pathogen in *Curcuma longa*

V.Sarathi[1*], Senthil kumar[1], R.Senthil kumar[2] and A.Panneerselvam[3]

[1]PG and Research Department of Microbiology, J.J. College, Pudukkottai, Tamil Nadu, India
[2]Department of Microbiology, PG Extension, Bharathidasan University, Perambalur, India
[3]Department of Botany and Microbiology, A.V.V.M.Sri Pushpam College, Poondi, India
**Corresponding author*

A B S T R A C T

Turmeric is a major foreign exchange earning crop. This crop is highly susceptible to rhizome rot infection caused by fungi. Samples of disease infected rhizomes of turmeric were collected from 5 different agricultural fields in Pudukkottai District.Samples were screened for isolation and identification of rhizome rot causing fungal pathogen. The isolate was found in *Pythium aphanidermatum* belonging to Phycomycetes is a well-known devastating pathogen of many vegetables, fruits, rhizomes, grasses and ornamental crops in several parts of the world with a wide host range. Pathogenicity testing was done according to Koch's postulates. Hence, this is the comprehensive report on identify of major fungal pathogens associated with rhizome rot.

Keywords

Rhizome rot, *Pythium aphanidermatum*, Spices *Curcuma longa*

Introduction

Turmeric, the 'golden spice' is prone todiseases like rhizome rot caused by *pythium aphanidermatum* (Rathaiah, 1982) and the crop is Zingiberaceae member like *Curcuma longa* in some turmeric growing tracts of five different villages of PudukkottaiDistrict of Tamilnadu. Turmeric is a perennial herbaceous monocotyledon spice and also an important foreign exchange earning crop (Erwin and Ribeiro, 2010). Turmeric is mainly exported in dry and powder forms (India spice 2009). Although turmeric is a high return crop but rhizome rot poses a persistent threat to the cultivation and storage of turmeric. About 50-80% losses during storage have been reported due to this disease (Nirmal, 1992). Traditionally, a large number of farmers cultivated turmeric in the region, but many gave up its cultivation owing to the frequent rhizome rot disease that destroyed the crops (Access, 2008).

The genus *Pythium aphanidermatum* belongs to the class Phycomycetes of kingdom Stramenopila. *Pythium* is a serious pathogen of many vegetables, fruits, grasses, rhizomes and ornamental crops in several parts of the world (Hendrix, 1973 andPlaats-Niterink, Vander 1981). *Pythium aphanidermatum*(Edson) Fitzp. is a serious pathogen in many horticultural crops in

warmer areas with a broad host range (Plaats-Niterink and Vander 1981). A pathogen with wide host range can survive over several seasons of cultivation symptomatically or asymptomatically on its host plants. One of the most useful approaches of disease management to this problem is crop rotation. But the susceptibility of the rotated crop to this pathogen may lead to failure of the crop production. Hence the information regarding the host range of the pathogen over rotated crop will be helpful for the better management of the disease.

Materials and Methods

Sample collection

Infected turmeric rhizomes were collected in sterilized polythene bags from five different villages of Ganapathipuram, Adhanakkottai, Pudunagar, Manjapettai and Thethuvasalpatti of Pudukkottai District, Tamilnadu, India. The infected rhizomes were washed by tap water and surface sterilized with help of 0.01% mercuric chloride solution for 1 minute. The rhizomes are stored at refrigerator in sterile condition.

Isolation and identification of fungal pathogen isolated from infected turmeric

For isolation of *Pythium aphanidermatum* the infected rhizomes were cut into small pieces and surface sterilized with 0.01% mercuric chloride solution for 1minute then rinsed in sterile distilled water. After surface sterilization the rhizome pieces were placed on sterile filter paper to remove moisture then the pieces placed on Petriplates containing Potato Dextrose Agar medium (Jeffers and Martin, 2010). The plates were incubated at 25±2°C for 72 hours. After incubation period the plates were examined the fungal colony was observed (Plaats-Niterink and Vander 1981).

Morphological identification

The main and traditional methods for identifyingfungal species are based on morphological and physiological studies (Taechowisan *et al*., 2008). Identification of *Pythium aphanidermatum* was based on standard keys suggested by (Plaats-Niterinkand Vander 1981) and Waterhouse (1967). Slides were prepared from these cultures and stained with lacto phenol cotton blue according to Parija and Prabhakar (1995) and examined under the light microscope.

Inoculum preparation

To prepare the inoculum of pathogen *Pythium aphanidermatum*, the potato dextrose broth was prepared and sterilized at 121ºC for 15 minutes. After sterilization the broth was allow to cool and the 6mm diameter of *Pythium aphanidermatum*pure culture was inoculated Erley'n Mayer flask containing PD broth. The pure culture of pathogen was already inoculated in PD agar plates. After inoculation the flasks are stored orbital shaker at 150 rpm for 7days.

Pathogenicity test

The sterilized potting mixture containing soil, sand and farm yard manure (1:1:1) was filled in six numbers of clay pot. The healthy rhizomes of turmeric were planted in all pots filled with sterilized potting mixture.The six pots were separated in to two sets, one set was control and another one set was treated. The 5ml of liquid culture of *Pythium aphanidermatum* was inoculated in second set of three pots containing 40 days older healthy turmeric plants. The plants without inoculum served as control (Johnston and Booth, 1983).

Three replicates were kept for each test. The water was irrigated in pots once in a week, trial pot was protected from insects and animals throughout the cultivation period under controlled condition. After 15 days of inoculum introduced the plants were observed the development of water soaked lesions on pseudo stem and subsequent yellowing of the leaves. The rhizome rot symptoms showed by these plants were observed carefully and were recorded at regular intervals.

Reisolation of the pathogen

The plants which showed symptoms of rhizome rot, rhizome were collected and used for the reisolation of the pathogen to prove the pathogenicity. The infected samples were brought to the laboratory and the infected portions including roots and rhizomes were used for isolation. These were washed thoroughly with tap water to remove the adhered soil. Small bits excised from the diseased portions along with some healthy portions were surface sterilized with 0.01% mercury chloride (or) with 75% ethanol for 1 minutes and then rinsed with sterile distilled water and placed on Petri plates containing potato dextrose agar and incubated at 27ºC for 72 hours. After incubation period the mycelial growth was observed on PD agar plates then the organism was identified by using standard methods.

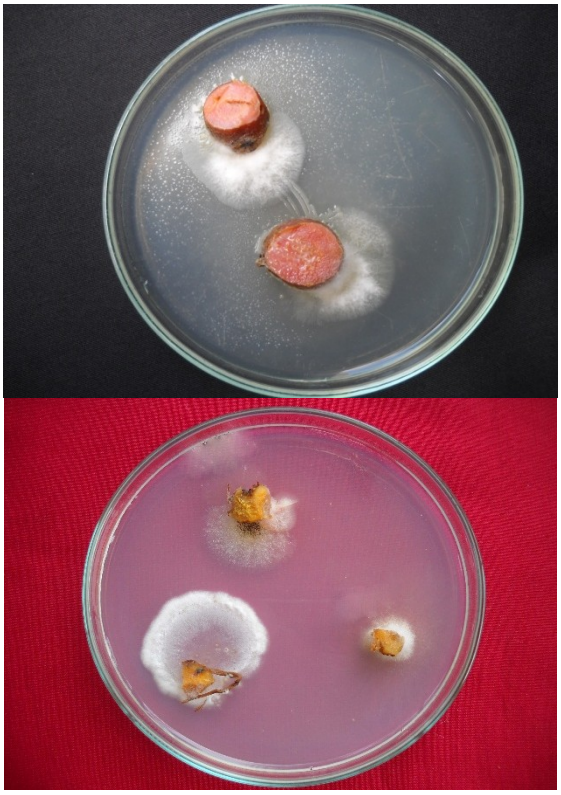
Results and Discussion

In this study, the recovered isolates from diseased turmeric rhizomes were identified on their morphological characteristics as well as colony growth was identified as, *Pythium aphanidermatum*. The symptoms showed after 15 days were typically as in the case of *Curcuma longa*. The leaves of affected plants exhibit gradual drying along the margins. This ultimately results in complete drying of all the leaves. The collar region of the pseudostem becomes soft and water soaked, resulting in collapse of the plant. The root system is very much reduced and its tissues are also affected. In severe condition, the infection spreads to rhizomes which decompose and turn into a decaying mass of tissues. The development of rhizomes is poor. The disease may appear in isolated plants or may involve several adjacent clumps resulting in appearance of diseased patches in the field.

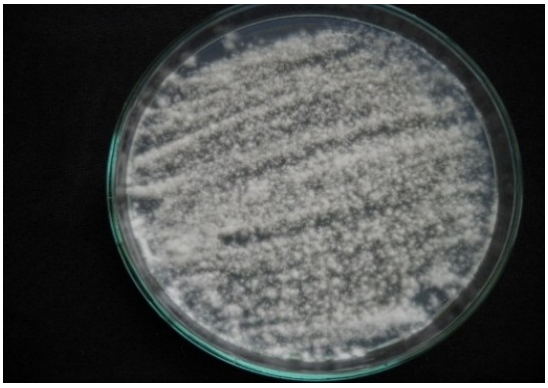
Pythium aphanidermatum

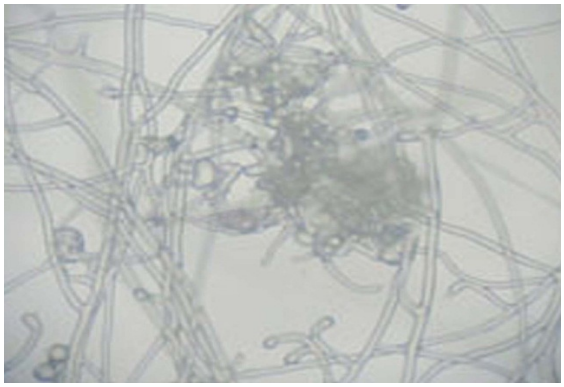
The hyphae are hyaline and the mycelium has no crass walls. Main hyphae upto 75µm wide and Oospores aplerotic, spherical, smooth, average 41µm in diameter. Sporangia are lobate consisting of terminal complexes of swollen hyphal branches of various length and germinated by extension of long exit tube and vesicle formation and zoospore discharge. Vesicles average 41µm in diameter. The presence of apluerotic Oogonium, Oogonia globose, terminal, smooth, average 28 µm in diameter, with straight Oogonial stalks. Antheridia typically intercalary usually diclinous and average 41µm long and 11µm wide, commonly 1 pair Oogonium. It produces white colored aerial mycelium on PDA medium.

Plate-1 Isolation of pathogen from infected rhizome



Pure culture and Microscopic observation of*Pythium aphanidermatum*



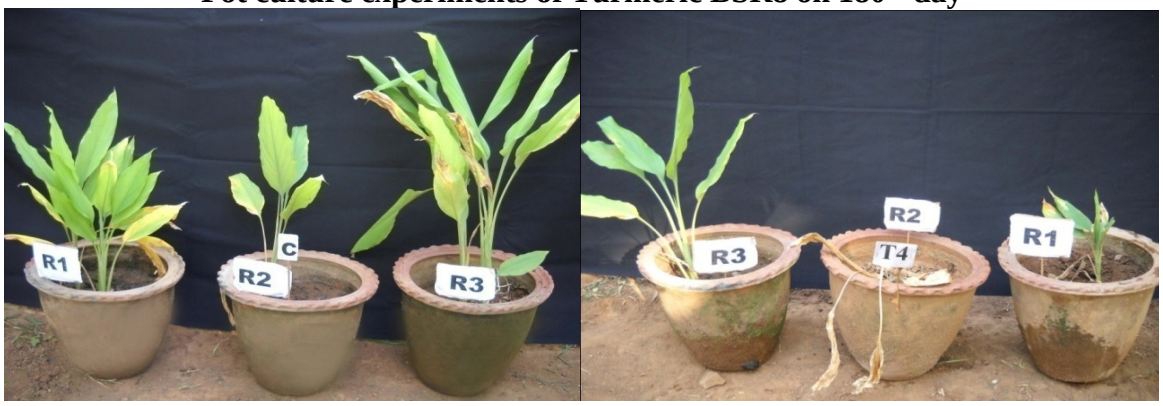


Pathogenicity test

Pot culture experiments of Turmeric Bhavani Sachar 8 (BSR8) on 60th day

Plate-2 Pot culture experiments of Turmeric BSR8 on 120th day

Pot culture experiments of Turmeric BSR8 on 180th day



C –Control, T4 – *Pythium aphanidermatum* treated and R1, R2 and R3 – Replication Pots

In the present study, the pathogenicity test is restricted to members of the family Zingiberaceae since they are rotated with turmeric in five different villages of Pudukkottai Districts of Tamil Nadu. In the present study *C. longa* were found to be hosts for *P. aphanidermatum* causing rhizome rot. *P. aphanidermatum* is a major constraint for the production of healthy rhizome all over the world, sometimes causing total failure of crop (Fagaria *et al*., 2006).

The pathogen was identified in this study have ability to grow on a wide range of substrates and have efficient mechanisms for dispersal as well as they can survive in the soil and in plants for many years as sporangiospores (Schulz *et al*., 2000). Therefore, it is very important to develop proper management practices to control this type of fungi. The results of this study revealed that *P. aphanidermatum* which are pathogenic to turmeric are found abundant in farms of five different villages of Pudukkottai District. In this investigation we observed that the severity of the disease is high due to high temperature condition which is favorable for the spread of especially *P.aphanidermatum*.

The identification of the fungi causing the decay of turmeric rhizomes is the first step towards further studies to develop an integrated crop management program to prevent and control rhizome rot in the fields.

References

Erwin, D.C. and Ribeiro, O.K. (1996). Phytophthora Diseases worldwide. The *American Phytopathological Society,* ST Paul, MN.

Fagaria, M.S., Choudhary, B.R. and Dhaka R.S. (2006). Vegetables crops production technology, Vol. II, Kalyani Publisher, New Delhi, Pp. 223-227.

Hendrix, F.F. and Campbell, W.A. (1973).*Pythium* as plant pathogens. Annua, Rev, Phytopathol. 11 77-98.

Jeffers, S.N. and Martin S.B. (2010). Comparison of two media selective for *Phytophthora* and *Pythium* species. *Plant Dis*. 70: 1038-1043.

Johnston, A. and Booth, C. (1983). Plant Pathologist's Pocketbook, second ed., Commonwealth Mycological Institute, Kew, p. 439.

Nirmal, K., Samsuddin, K. and Ratnambal, M.J. (1992). Plant, Cell tissue and Organ culture. 29:71-74.

Parija, W. and Prabhakar, P.K. (1995). Evaluation of lacto phenol cotton blue for wet mount preparation of feces. *J. Clin. Microbiol*., 33: 1019-1021.

Plaats-Niterink and Vander (1981). Monograph of the genus *Pythium. Stud. Mycol.* 21: 1-24.

Rathaiah, Y., (1982). Rhizome rot of turmeric. *Indian Phytopathol.* 35: 415-417.

Schulz, D., Lambrecht, S. and Wolf, G. (2000). Biological control of *Pythium ultimum* in sugar.

Taechowisan, T., Wanbanjob, A., Tuntiwachwuttikul, P., Shen, Y. amd Lumyong, S. (2008). Synergistic activities of 4-arylcoumarins against Phytopathogenic fungi, *Res. J. Microbiol.,* 3: 237-245.

Waterhouse, G.M. (1967). Key to *Pythium pringsheim. Mycol. Pap.* 109: 1-15.