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Colletotrichum capsici, is the most threaten causal agent causing Anthracnose, Die-Back and Fruit Rot of chilli (*Capsicum annum* L.) in Odisha

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Colletotrichum capsici (Sydo) Butler and Bisby causing anthracnose, die-back and fruit rot of chilli (*Capsicum annum* L.) have been described. *C.capsici* was identified from various parts of Odisha according to their cultural, morphological characters and pathogenenicity test and squash mount study under microscope.

Key words: *Capsicum annum, Colletotrichum capsici,* chilli

INTRODUCTION

In Odisha, chillies are grown throughout the year in all the 30 districts. It is cultivated in an area of 75.53 thousand hectare with total production of 852 thousand million tonnes with average yield of 64.32 kg/ha in both *Kharif* and *Rabi* seasons (Odisha agriculture statistics-2009-10) Among the districts, Keonjhar stands first in production 4.16 thousand million tonnes followed by Ganjam 4.02 million tonnes and Anugul 3.95 million tonnes in both the seasons while the lowest production is observe in the districts of Puri (0.28) million tones, Nayagarh 0.46 million tonnes and Khordha 0.47 million tonnes.

Like other spices and vegetable crops, chilli is also subjected to the attack of several diseases and insect pests., besides unfavourbale and adverse climatic conditions which are suspected to be determined to the yield of this crop. Among the various diseases, die-back and ripe fruit rot, bacterial spots, mosaic and leaf curl of chilli are most important in decreasing the yield to a great extent. Among the fungal diseases, Friut rot and Die back caused by *Colletotrichum capsici,* is commonly affecting to the chilli crop in all the district during the growth period.

MATERIALS AND METHODS

Diseased specimens of chilli fruits, leaf and stem from districts of Odisha. during April - May 2008-09.

Collection and preservation of specimens

The specimens were collected and kept in specimen tubes, paper packets and polythene bags. A field note on the habitat of the host and the symptoms produced was prepared during the collection. Fresh specimens were photographed and then dried between blotting papers with powdered napthalen or paradichlorobenzene and were pressed in few layers of papers. After the specimens were properly pressed and dried, they were preserved in paper packets along with small napthalen powder and properly accessioned.

Associations of Colletotrichum capsici in chilli seeds

Fruit rot infected ripe fruit of chilli were collected during disease survey in 10 different agro climatic

zones of Odhisha. The samples (1.0 kg each) were collected adjacent to the research stations/ Krishi Vigyan Kendras of OUAT. The samples were dried separately and the seeds were collected and preserved for future study.

Two hundred seeds were taken from each sample to study the association of any fungi on/in seeds. One hundred seeds were distinfected separately with 0.1% mercuric chloride solution for 1-2 minutes, followed by through washing with sterile water for three times. The rest 100 seeds of each sample were kept without surface sterilizatrion. one hundred seeds of each category (surface sterilized and surface unsterilized) were placed on moist blotters and the remaining 100 seeds of each were plated in PDA medium (de Tempe, 1953 and 1963). At the rate of 10 seeds per plate with three replications of each. The moist blotters and Petriplates were incubated in an incubator at 28°C ±1. Observation on the occurrence of fungal colonies in different samples were recorded after seven days of incubation (Fig. 1a and 1b)

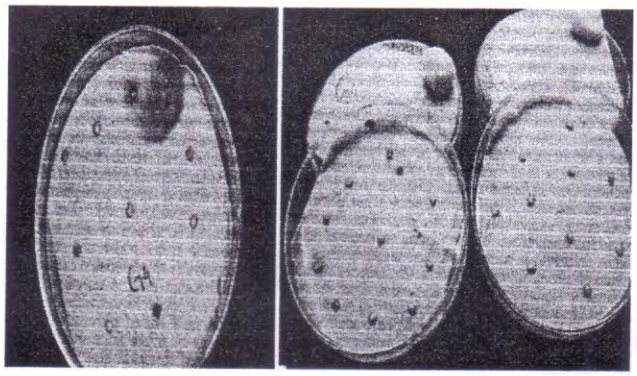

Fig.1a and 1b : Showing the association of *Colletotrichum capsici* in chilli seeds

Study of Symptomatology

In order to study the symptoms of die-back and fruit rot diseases of chilli caused by *Colletotrichum capsici,* susceptible and popular chilli variety Pandra Mircha, which is extensively grown in Rayagada district of Odisha was selected healthy and diseased (fruit rot) fruits of the variety were collected from the Adopted village of KVK, Rayagada Gunupur (Korma and Bijapur) during May, 2007 (Fig 2a., 2b, 2c and 2d). The crop was grown during *rabi*, 2007-2008 at the Instructional farm of Krishi Vigyan Kendra, Rayagada, (Gunupur), in the districts of Rayagada and also at adjoning farmers' field. Nursery beds were raised separately for healthy and diseased seeds in the month of November, 2007-08 for growing the crops in *rabi* seasons, following the normal package of practices transplantation of 4 wk old seedlings done separately in plots of 9.0 m x 3.0 m in a spacing of 45 cm x 30 cm with

Fig.2a,2b,2c,2d : Showing leaf spot, die back, fruit rot and wilting of chilli crop in the field condition.

three replications. The trial plots were kept out of fungicidal contact for allowing the pathogen for its normal growth and infection without possibility of hindrance. Steps were taken to observe the appearance of different symptoms on seeds, seedlings, grown up plants and fruits under natural conditions.

Isolation of fungus

The samples were brought to the laboratory in a sealed plastic bag. Lesions were excised, surface sterilized with 70% ethanol for 30 sec. rinsed with distilled water and dried on filter papers. They were cut into smaller pieces of approximately 5 x5 mm size and placed in Petri dishes containing potato dextose agar (PDA) and incubated at room, temperature in the dark for 48 to 72 hr. Fungus was isolated and maintained in slants for preservation. Isolation of fungus from infected fruit was also made following the standard procedure as mentioned earlier. To obtain the pure culture of fungus a single spore isolation method was used. A healthy spore was collected from the suspension of spores by observing under compound microscope. Spore

was transfored to a PDA medium are kept in an incubator for growth.

Pathogenicity

The pathogenicity test for some of the diseases was carried out on healthy potted plants. These plants were inoculated with mycelia and spore suspension of respective fungal pathogens grown in 7

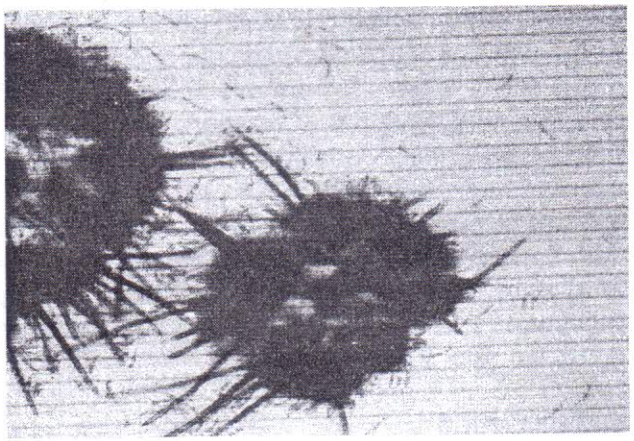

Fig. 3 : Showing Long setae and acervuli and conidia of *Colletotrichum capsici*

days old pure culture. The mycelia and spore suspention was atomized over injured leaves of test plants which were kept in a moist chamber for 24 hours after inoculation. Observations on the development of characteristic symptom on the test plants were taken. Control plants were left in each case and were atomized only by sterile distilled water and kept under identical conditions.

The pathogenicity test was carried out on potted healthy plants with mycelial and spore suspension (6×10^6 CFU.ml^{-1}) of 7 days old culture. The inoculated plants were spots appeared on the inoculated leaves which gradually enlarged to large necrotic patches on the leaves, from which the pathogen was re-isolated.

Identification of the Fungus

The fungus was identified by conventional method basing on the colony characteristics, nature of growth, shape and size of fruiting body i.e. acervuli ,conidia and presence of setae etc with the help of compound microscope and LEICA - DM 3000 (German) a phase contrast bright field microscope and identified as *C.capsici* as described and correlated by Gilman, 1956 ; Sparrow and Sussman, 1973 ; Chowdhry et al., 2000 and Sarbhoy, 2006.

Study of cultural characteristics

The fungus isoalted from the diseased parts of chilli fruits were subjected to incubation at a temperature of 28^{0}C±1 for growth and sporulation. The fungus was grown in Petriplated by transferrlng 5 mm of mycelial disc of 7-day-old pure culture. The charcteristics of the fungal colony, including colour, shape and growth habits of vegetative and reproduction parts were observed by naked eye and also by compound microscope.

Study of morphological and taxonomic characteristics

Morphological identification : An inoculum loop was used to scratch the mycelium of the colonies and mounted on the slides. The compound microscope (Unilab - RH81) and LEICA - DM 3000 (German), a phase contrast bright field microscope were used to observe the colony colour and form, appressorium features, shape, size and colour of spores from different isolates. The measurements of mycelia, conidia and acervuli were recorded by using stage and ocular micrometers. The detailed measurements and other microscopic characters were also recorded from 50 readings of each. Identification was made by comparing the characterstics of fungi following Charlie *et al.* (2001) and Deacon (1998).

On the basis of morphological as well as cultural charcters the pathogen was identified as *Colletotrichum curcumae* (Sydo) Butler and Bisby (Butler and Bisby 1993), In Odisha, the present species is also related to *Colletotrichum capsici* in regularly producing setae, in the host as well as in culture, but it differs from the latter in conidial characters having slightly curved conidia with one end rounded and the other end bluntly pointed and with three oil globules instead of one oil globule. The setae in the present species are bluntly pointed unlike in *C. capsici* where the setae are sharply pointed and conidia are perfectly bent with pointed ends and two oil globules. Moreover variations amongs the isolates of *C. capsici* have been also reported by Palarpawar and Ghurde (1993). As per the description of these authors the present isolate has similarities with isolate -II as regards conidial and setae characters and absence of formulatuion of setose sclerotia at periphery of colony which are formed in isolated -I of the species. The conidial size in isolate-I was more in comparison to isolate-II.

RESULTS AND DISCUSSION

Identification of the fungus

Ripe chilli fruits infected with the fruit rot disease were collected and thin transverse section of the affected parts were prepared and critically examined under microscope to job serve the morphological characters of the fungus.

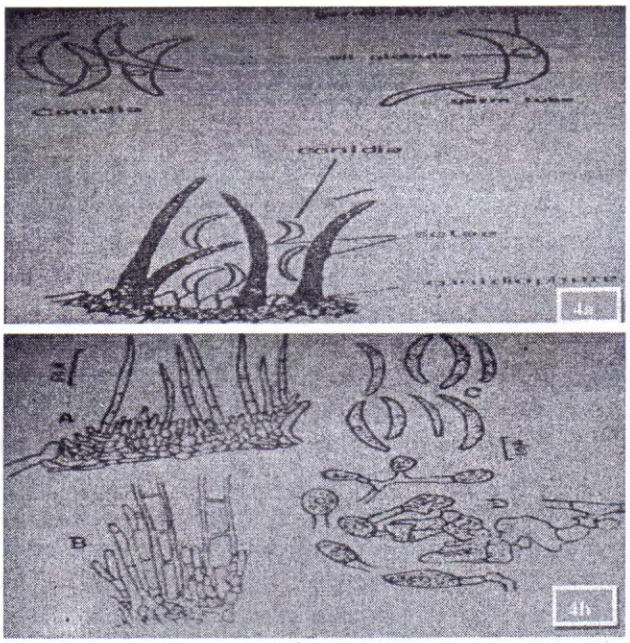

Fig. : 4a and b : Camera lucida photograph showing setae, falcate conidia and short conidiophores

The pure culture of the fungus was also maintained in the Petriplates and also in plants after isolation from the disease samples and the cultural characteristics were observed.

Study of cultural characteristics

The fungus in pure culture produced colonies with profuse cottony mycelium, initially white, later turning into a ashy black in colour with blackish islands as fruiting bodies interspersed on the mycelial mat which were produced after 10-12 days.

Study of morphological and taxonomic characteristics

The mycelium was septate, profusely branched, inter and intracellular, pale olivaceous in colour measuring, 1.78-3.56 µm diameter averaging 2.67µm in diameter. The acervulus was found composed of long setae (Fig.-3), conidiophores and conidia on isodiametric stromactic tissue. The setae were scattered with tubuler structure sparingly septate, thinner at the base with pointed tips, dark brown with tips light brown in colour and measured, 119.8 -179.2 x 3.6 - 5.4 µm averaging,149.5 x 4.5 µm. The conidiophores were short, hyaline, multicelled, club shaped, arising from the stromatic tissue and measuring 9-30 x 2-4 µm averaging 19.5 x 3 µm in size. The conidia were falcate curved with narrow ends, hyaline, aseptate each with an oil globule, at the centre (Fig. 4a and 4b). They measured, 21.67 - 27.80 and 2.96 - 3.78 µm averaging, 24.73 x 3.37 µm in size.

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