

Pathological variability of *Colletotrichum capsici* isolates associated with anthracnose of chilli (*Capsicum annuum* L.)

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ABSTRACT

The four different pathovar of anthracnose of chilli caused by Colletotrichum capsici causing were isolated from different places of chilli growing areas namely KC1, KC2, KC3 and KC4. Fungal isolates from distinctive chilli fields causing typical die back symptoms were identified as belonging to the C.capsici. Pathogenicity test validated that four isolates were causal agents of chilli anthracnose disease and KC4 was distinguished highly pathogenic to chilli fruits. To know the virulence isolates fruits were inoculated and results suggested maximum fruit rot intensity were observed in isolate KC4 and found to be more aggressive with disease intensity of (60.15%), having brown colour colony on PDA and also having highest conidia size of 26.36 x 4.08 µm.

Kew words: Colletotrichum, isolates chilli, Anthracnose.

INTRODUCTION

India is one of the leading producers and exporter of chilli which contributes about thirty-five percent of total spice production. Chilli can be used in diversified forms such as green and red chilli which has C vitamin and A vitamin (Saimbhi *et al.*, 1977). The occurrence of disease caused by various causal agents such as fungus, bacteria and viruses which is directly reduce total yield of crop. More than 40 fungal pathogens attack in chilli crops. (Rangaswami ,1979). Amongst various fungal stresses the genus *Colletotrichum capsici* (Syd) Butler and Bisby,) is the serious pathogen causing a devastating disease which known as anthracnose and has national importance. The fungus spreads in all over chilli growing dominant areas of India resultant in disease occurrence level among 66-84 percent & suffering produce harm up to 12-50 per cent(sharma *et al* 2005; Manandhar *et al.*,1995,Poonpolgul and Kumphai,2007;pakdeeveraporn *et al.*,2005; Saxena 2016). The genus *Colletotrichum* is the most devastating causal agent of chilli (Yang *et al.* 2012, b). C.

capsici also has been described the one of the major problems under Agro climatic zones (shenoy et al.,2007).

The Per cent disease intensity on crop is determined by the strain of the pathogen. **Sharma et al., (2005)** recounted the presence of fifteen strains of *Colletotrichum* by inoculated of green chilli fruit which were based on occurrence of disease symptom development. (**Montri et al., 2009**) also reported the showed aggressiveness of *Colletotrichum. capsici* isolates by measurement such as per cent lesion size, appearance of necrotic and existence of fruiting body acervuli on *Capsicum* species. Thus, some evidence like spreading of pathotype in chilli cultivating regions can be helpful for accurate taxonomic identification and well characterization of *C. capsici* for the purpose of developing new varieties and plantdisease managing (**Freeman et al., 1998**).

Materials and Methods:

The Current research work were conducted in Lovely professional university, India in the controlled condition, using completely randomise design (CRD).

Isolation of test pathogen:

Anthrachnose infected chilli fruits were collected from different geographical region of India. With the help of sterile surgical blade small infected tissue were cut along with some healthy portion. These pieces were disinfected in 1% sodium hypochlorite (NaOCl) for ten seconds & finally washed three times with distilled water under aseptic conditions. To remove the extra moisture these small pieces were placed on sterilized blotting paper. Single piece was kept PDA containing Petri plates and were kept in B.O.D incubator at temp $25 \pm 2^{\circ}\text{C}$ for 7 days and observed at regular intervals to detect of the mycelium growth of the fungus (**Akhtar and Singh, 2007; Ratanacherdchai et al., 2010; Ramaswamy and Mahadevan, 1999; Abang 2006**).

Purification of pathogen

All the pathogen strains were purified by using single spore isolation method (**Choi et al.,1999 & Rangaswami and Mahadevan,1999**) with minor modification.

Pathogenicity test

By Pin pricking method:

Pathogenicity tests were done by using plug inoculation method suggested by Thind and Jhooty, 1990. Before inoculation, healthy fruits of chilli were washed with 70% (v/v) ethanol

to remove external infection. 10 µl spore suspension of *Colletotrichum capsici* were taken from ten days old culture were inoculated on injured chilli fruit. The observation was taken at regular interval as lesion diameter of infected fruits (**Ratanacherdchai et al., 2007**) Per cent disease intensity (PDI) was analysed by using disease rating scale (**Mayee and Datar 1986; Santosh Reddy and Nargunda, 2015**).:

Disease Survey and occurrence

Per cent disease incidence of chilli Anthracnose incidence was recorded by scoring five plants in each microplot using 0-9 scale [**Yang, J., 1999.**]

Results: Pathogenicity test

Four *Colletotrichum capsici* strains were used for testing of virulence of the pathogen. Strains were used in this experiments were namely KC1, KC2, KC3 and KC4. The pathogen was associated with the host confirmed to be *Colletotrichum capsici* based on symptomatology and study of the causal agent.

Table 1 . Pathogenicity test of four pathotypes

Pathotypes	Area of sample	Disease intensity (%)	Aggressiveness of isolates
Kc1	Uttar Pradesh	43.82	++
Kc2	Karnataka	46.45	++
Kc3	Karnataka	31.57	+
Kc4	Andhra Pradesh	60.15	+++

Aggressiveness of the *Colletotrichum* isolates : Out of four isolates the isolate KC4 was significantly found to be most virulent which recorded the highest fruit rot infection (60.15) followed by KC2 (46.45) per cent .least aggressiveness were observed in KC3 (31.57) per cent. The disease reaction of all strains were categorised given the subsequent disease grade (**Singh et al., 1993**).

Fig No 1 Pathogenicity test in Lab condition



A. Inoculated with spore suspension of *C.capsici* in plastic tray, B. Lesions formed on Red chilli C. Lesions formed on green chilli.

Spraying of inoculum on chilli plant: The culture suspension was inoculated on leaves and the chilli plants were enclosed with brown paper to provide moisture throughout the experiment Data of symptoms on fruits & leaves were recorded at regular interval

Table 2: Disease score of *C.capsici* to calculate PDI

Grade	% lesioned area covered by inoculated fruit	Reaction
0	0	-
1	1-10	+
3	11-25	++
5	26-50	++
7	51-75	+++
9	>75	++++

+ NO virulence, + less Virulent, ++ moderately virulent, +++ virulent & ++++ highly virulent

Fig 2 : Pathogenicity test in pot under playhouse condition



Healthy plant

diseased plant

Fig : 3 Symptoms of *C.capsici* on shoot and fruits under field condition.



A Chilli Die-back symptom
ring on fruit

B. Sporulation on fruit

C. Sunken concentric

Discussion

Anthrachnose is accountable for main losses in chilli growing areas (Pakdeeveraporn *et al.*, 2005). The *Colletotrichum capsici* which harm chilli crop (Butler,1918) is recognized to reduce the production of chilli in major growing areas. *Colletotrichum capsici* is very dominant in each corner of the country and is considered to be a critical problem of chilli

(Grover and Bansal, 1968, Chowdhury, 1957). The investigation by (Cannon *et al.*, 2000) was confirmed the species *C. capsici* accountable for causing anthracnose of chilli. The present study focused on pathogenicity, was used to identify and distinguish the pathovar of *C. capsici* causing anthracnose. The study displayed that the behaviour of *C. capsici* strain were similar with respect to appearance of disease. Variances in disease intensity of *Colletotrichum* pathovar have been observed by earlier (Taylor *et al.*, (2007). The Pathogenicity test validated that four isolates were causal agents of chilli anthracnose disease and KC4 was identified highly pathogenic to chilli fruits and shown the same symptom of fruit rot. The current outcomes are conformity with the finding of (Desai and Prasad 1955) and the similar results were found by Sutton 1973 and Kulshrestha *et al.*, (1976).

Conclusion:. These outcomes shown that *Colletotrichum* strains are in different in their aggressiveness and varied in disease intensity in chilli crop & all the isolate were found to associate with anthracnose of chilli which were isolate from different parts of India. These results also helping to identify the pathogenic behaviour of pathogen. .

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