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# RED ROT OF SUGARCANE: MORPHOLOGICAL VARIABILITY IN SITAPUR DISTRICT OF UTTAR PRADESH

Vikash Pandey & D.N. Shukla
Bhargava Agricultural Laboratory, Department of Botany, Faculty of Science, University of Allahabad, Allahabad-211002, Uttar Pradesh, India.

**ABSTRACT**
Sugarcane is an important cash crop and used as the chief source of sugar grown tropical and subtropical region in India. Sugarcane production is challenged by various biotic and abiotic stresses among the biotic factors, red rot disease caused by *Colletotrichum falcatum* is a major disease leading to severe reduction in sugarcane production. Cultural, morphological studies conducted under in *vitro* in Oat Meal Agar (OMA) showed characteristic variation in their conidial and colony characters which were collected from various places in Sitapur District of Uttar Pradesh. Thus present study brings the cultural, morphological variations and virulence characters of *C. falcatum*.

**KEYWORDS:** Red rot, Morphology, Sugarcane.

## INTRODUCTION

Sugarcane (*Saccharum officinarum* L.), belong to the family *Poaceae*, is an economically important cash cum industrial crop grown in the tropical and sub-tropical region in India. Many biotic and abiotic stresses affected sugarcane quality and yield. Sugarcane diseases caused by fungi, bacteria, virus and mycoplasma, such as fungal diseases become major problems for the sugarcane growing countries. Red rots major fungal disease occur all sugarcane growing state in India. Red rot disease is the oldest serious fungal disease of sugarcane, generally called "Cancer" of sugarcane is caused by *Colletotrichum falcatum* Went. Disease incidence depends upon the varieties, localities and favourable environmental condition. The disease was first described from Java (now Indonesia) by Went (1893), who called the fungus, *C. falcatum* and named the disease as "het rood snot" meaning "red smut". The sexual stage of *C. falcatum* was later reported by Spegazzini (1896) in Argentina who named it *Physalospora tucumanensis.* Barber (1901) first observed this disease in Bihar (India) and Butler (1906) coined the name "Red Rot", the name by which it is known till date. Later, the red rot causal organism was reclassified by Von Arx and Muller (1954) and included in the genus *Glomerella* as *G. tucumanensis*. In India, the first documented epidemic of red rot occurred in 1895-1901 and in subsequent years a number of major outbreaks have been recorded as a regular event in the sub-tropical and tropical regions of the country (Satyavir, 2003). This disease has been hold accountable for 5 to 10% cane yield and sugar recovery loss worldwide. Red rot is considered as the major constraint for sugarcane production in India (Viswanathan and Samiyappan, 2008). It has been reported as damaging disease of sugarcane cultivars in Australia, Bangladesh, Pakistan, Taiwan, and USA (Viswanathan and Samiyappan, 2002). Red rot is widely distributed and has been reported in 68 sugarcane growing countries of the world (Bharti *et al.*, 2012). Red rot infection in cane causes a loss of total weight to about 29.07% leading to 30.8% loss in sugar recovery (Hussnain and Afghan 2006). The present study conducts cultural and morphological variability of *C. falcatum* based on their conidial and colony characteristics. In the present study, an attempt has been made to collect five isolates of *C. falcatum* Went prevalent in different sugarcane growing parts in Pilibhit district of Uttar Pradesh in India.

## MATERIALS & METHODS

### Survey and collection of disease samples

An extensive survey of sugarcane growing areas various localities Aira, Ajbapur, Kumbhi, Gulariya, Belrayan, Paliyakalan, Khambharkheda, Sampurnanagar and Gola Gokarannath in Lakhimpur (Khiri) districts of Uttar Pradesh were conducted during July and August months of 2012-13. Varieties of sugarcane red rot disease symptoms of were collected for 27 isolates of *C. falcatum*. Strains were isolated from lesions on infected stem pieces. Symptoms of red rot disease on these cultivars were recorded. Red rot infected sugarcane sample collected from directly farmer's fields.

**TABLE 1**. Incidence of red rot disease

| Districts | Localities | Varieties | Strains |
|---|---|---|---|
| Sitapur | Maholi | CoS 767 | CFSIMA |
| Sitapur | Biswan | CoS 8436 | CFSIBI |
| Sitapur | Hargaon | CoJ 64 | CFSIHA |
| Sitapur | Ramgarh | CoS 8436 | CFSIRA |
| Sitapur | Kamlapur | CoSe 92423 | CFSIKA |
| Sitapur | Jawaharpur | CoS 767 | CFSIJA |
| Sitapur | Mehmudabad | UP 9530 | CFSIME |

**Isolation of *Colletotrichum falcatum***

Infected canes were split open by sterilized knife and observed for reddish tissue and white transverse band. The red rot pathogen was isolated by tissue segment methods as described by Rangaswami (1958), three 5-5 mm pieces of tissue were taken from the margin of infected tissues, surface sterilized by dipping in 1% sodium hypochlorite for 1 min, immersed in 70% ethanol for 1 min and rinsed three times with sterilized water and finally dried in sterilized tissue paper (Abbas *et al.*, 2010). After 5 days of incubation, the plates having red sporulation were purified by sub-culturing. All the isolates were further purified by single spore technique (Riker and Riker, 1936). The fungus from the pure cultures obtained was examined microscopically in order to match it with the characters of the pathogen examined from the diseased samples. The pure cultures were maintained in Potato Dextrose Agar slants.

Samples were placed on water agar and incubated at room temperature (26 to 31°C). The growing edges of any fungal hyphae developing from the tissues were then transferred aseptically to oatmeal agar medium and fungi were identified following sporulation. Single spore subcultures were obtained for each isolate using the procedure described by Goh, (1999). When the fungus showed sporulation, spore masses were pieced off with a sterilized weir loop and streaked on the surface of water agar. After inoculating overnight at 29 ± 2°C on biological oxygen demand (BOD), single germinated spores were picked with a sterilized needle and transferred to oat meal agar (OMA) medium. The cultures of different isolates were maintained on OMA slants at 4°C for further studies.

**Morphological Characteristics**

Morphological characters of the colony viz., colony color, substrate color, margin of colony and topography were recorded through naked eye and spores *viz.*, size, color and shape of the conidia were observed in binocular microscope with oculars lens. The three replicate mean values examined and the range was determined.

**Results**

In order to find cultural, morphological variations of *Colletotrichum falcatum* among the different parts of Uttar Pradesh, an extensive survey was conducted in major sugarcane growing areas of Uttar Pradesh covering (Aira, Ajbapur, Kumbhi, Gulariya, Belrayan, Paliyakalan, Khambharkheda, Sampurnanagar and Gola Gokarannath) Lakhimpur (Khiri) 2012-2013.

**Cultural, Morphology Characteristic of *Colletotrichum falcatum***

The morphological characteristics of different isolates of *C. falcatum* on OMA medium were studied; significant variations were observed with respect to Conidial and Colony characteristics base.

**TABLE 2**. Conidial characteristic of *Colletotrichum falcatum*

| Strains | Conidial characteristics | | | |
|---|---|---|---|---|
| | Length (µm) | Width (µm) | Colour | Shape |
| CFSIMA | 24.2 | 4.6 | Hyaline | Falcate |
| CFSIBI | 25.1 | 4.7 | Hyaline | Falcate |
| CFSIHA | 25.4 | 4.5 | Hyaline | Falcate |
| CFSIRA | 24.7 | 4.8 | Hyaline | Falcate |
| CFSIKA | 23.8 | 4.5 | Hyaline | Falcate |
| CFSIJA | 25.9 | 4.5 | Hyaline | Falcate |
| CFSIME | 24.7 | 4.5 | Hyaline | Falcate |

Table shows that, the maximum conidial length of *Colletotrichum falcatum* was recorded for strain CFSIJA as 25.9 µm, which was followed by CFSIHA as 25.4 µm, CFSIBI as 25.1 µm, CFSIRA as 24.7 µm, CFSIME as 24.7 µm and CFSIMA as 24.2 µm, whereas the minimum colony growth of *C. falcatum* was recorded for strain CFSIKA as 23.8 µm. The maximum conidial width of *C. falcatum* was recorded for strain CFSIRA as 4.8 µm, which was followed by CFSIBI as 4.7 µm, whereas the minimum conidial width of *C. falcatum* was recorded for strains CFSIMA, CFSIHA, CFSIKA, CFSIJA and CFSIME as 4.5 µm.

**TABLE 3**. Colony characteristic of *Colletotrichum falcatum*

| Strains | Colony characteristic | | | | | |
|---|---|---|---|---|---|---|
| | Colony colour | Substrate colour | Margin | Topography | Colony (mm) | Sporulation |
| CFSIMA | White Orange | Pinkish Black | Smooth | Raised Fluffy | 88.1 | +++ |
| CFSIBI | Greyish | Black | Smooth | Raised Fluffy | 85.5 | +++ |
| CFSIHA | Greyish White | Dark Greyish | Smooth | Raised Fluffy | 86.8 | +++ |
| CFSIRA | White | Black | Smooth | Raised Fluffy | 89.0 | +++ |
| CFSIKA | Black | Black | Smooth | Raised Fluffy | 88.3 | +++ |
| CFSIJA | Black | Black | Smooth | Raised Fluffy | 86.9 | +++ |
| CFSIME | Greyish White | Black | Smooth | Raised Fluffy | 86.3 | +++ |

Table shows that, the maximum colony growth of *Colletotrichum falcatum* was recorded for strain CFSIRA as 89.0 mm, which was followed by CFSIKA as 88.3 mm, CFSIMA as 88.1 mm, CFSIJA as 86.9 mm, CFSIHA as 86.8 mm, CFSIME as 86.3 mm, whereas the minimum colony growth of *C. falcatum* was recorded for strain CFSIBI as 85.5 mm, The colony colour of *C. falcatum* was recorded for strain; white orange colour CFSIMA, greyish colour CFSIBI, greyish white colour CFSIHA and CFSIME, white colour CFSIRA, black colour CFSIKA and CFSIJA. The substrate colour of *C. falcatum* was recorded for strain; pinkish black colour CFSIMA, black colours CFSIBI, CFSIRA, CFSIKA, CFSIJA and CFSIME; dark greyish colour CFSIHA. All strains margin smooth and topography raised fluffy growth observed.

## DISCUSSION

Red rot caused by the fungus *Colletotrichum falcatum* is dreadful and seed transmissible stalk disease which spreads from place to place through the infected sugarcanes (Ramesh Sundar *et al.,* 2009; Malathi *et al.,* 2010). The length of conidia ranged from 25.9-23.8 µm. highest length of conidia was observed in CFSIJA strain (25.9 µm) and shortest conidia were recorded in CFSIKA isolates (23.8 µm). Width of the conidia ranged from 4.5 to 4.8 µm. highest conidia CFSIRA (4.8 µm) and Shortest conidia CFSIME (4.5 µm). Conidia were falcate shaped with a round apical end tapering towards the base. The colony radial growth 89.0-85.5 mm. maximum colony radial growth was observed in CFSIRA strain (89.0 mm) and minimum colony radial growth were recorded in CFSIBI strain (85.5 mm). Different colony colours *viz.,* white orange, greyish, greyish white, white and black colours were observed. All the isolate showed variation regarding substrate colour, margin, topography, colony diameter and sporulation (table-3). Morphological diversity has been found in four isolates of *C. falcatum* from SPF234, CO1148, BF162 and SHF242 (Abbas *et al*., 2010). Variability in cultural and morphological characters and virulence and development of physiological races has been attributed to hybridization, mutation conidial and hyphen fusions (Bharti *et al.,* 2011). Morphological variation revealed that there exists a wide variation among the isolates which is the basic method for characterization of different thirty isolates (Prema *et al.,* 2013). Similar results have been obtained by Malathi *et al*. (2011), in a study with large number of isolates the growth of isolates has direct correlation with pathogencity and it revealed that the tropical isolates were light colored, fast growing and highly sporulating types. Prema *et al.* (2011) observed wide variation in *C. musae* isolates with respect to cultural and morphological characters, the isolates produced blackish white, light pink and dark orange coloured colonies. Chona and Srivastava (1960) sub-divided each of these two races into groups and sub-groups based on the texture of the mycelium and the degree of sporulation. They found that isolations made from diseased canes from localities affected with the red rot epidemic invariably yielded light, highly sporulating strains whether isolated from the diseased stalk or midrib lesions and the dark sparsely sporulating isolates were only rarely encountered in epidemic areas. Two types of colony morphology were classified in which the light type was observed more frequently than the dark type in 15 isolates of Thailand (Sangdit *et al.,* 2014). Sutton (1992) reported that the conidial size of *C. falcatum* ranged between 15.5 and 26.5 µm in length and from 4 to 5 µm in width. Kalaimani (1995) examined six isolates of *C. falcatum* and found variation in the length and width between 30.62 to 37.65 µm and 6.69 to 8.46 µm, respectively. Mishra and Behera (2009) revealed significant variation in the size of conidia of *C. falcatum* from India where the dimensions varied between 23.94 and 30.83 µm in length and from 3.28 to 3.69 µm in width. Prihastuti *et al.* (2010) reported differences in the size of conidia (16–35 µm long, 4–5 µm wide).

## CONCLUSION

The length of conidia ranged from 25.9 to 23.8 µm. Width of the conidia ranged from 4.5 to 4.8 µm. Conidia was falcate shaped with a round apical end tapering towards the base. The colony radial growth ranged from 89.0-85.5 mm. Different colony colours *viz*., white orange, greyish, greyish white, white and black colours were observed. All the isolate showed variation regarding substrate colour, margin, topography, colony diameter and sporulation.

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