

Incidence of *Fusarium oxysporum* spp. *udum* on Pigeonpea in Bundelkhand Region, India

ABSTRACT

Survey was conducted of pigeonpea growing areas of 4 districts in Bundelkhand region of Uttar Pradesh during 2019-20. *Fusarium udum* was found constantly associated with the root samples. This indicates that this fungus, a well-known wilt pathogen, was primarily responsible for the wilt disease of pigeonpea. The average disease incidence ranged between 3.25% to 49.00% from district to districts. The average incidence percentage of wilted plants in Chitrakoot district was 39.06% followed by Banda district 25.67%, Hamirpur district 15.99% and Mahoba district 14.64% respectively. The maximum wilt (*Fusarium oxysporum*) incidences were recorded in Chitrakoot district followed by Banda, Hamirpur, and Mahoba. Chitrakoot isolate of *F. udum* was found more pathogenic and caused higher wilt incidence than other isolate. All the isolates differed in their radial growth colony characters on both solid media. It was found that PDA was the best medium in compare to PSA. The Chitrakoot isolate and radial growth was fast growing followed by others. Sporulation was moderate to excellent in different isolates. However, the maximum radial growth was a recorded-on PSA in Chitrakoot isolate and minimum radial growth in Mahoba isolate. The most distinguishing characteristic of the macro conidia are their strongly curved or hooked apices and measure 11-21.12 x 1.95 to 3.78 μ m.

Key Words- Survey, Isolates, Colony, PDA, PSA, Pathogenicity and Radial growth

1. INTRODUCTION

India is the leading producer of pulses in the world. India shares 25% of global production, 27% of world consumption and importer 14% of pulses in the world [4]. Major pulses viz., chickpea, pigeonpea, moong bean, black gram, lentil, peas and various kinds of beans are grown in India [1]. Pigeonpea and chickpea forms majority of share in total production of pulses. India ranks first in area and production of pigeonpea in the world contributing 80% and 67% in world's acreage and production, respectively [2]. It is the second most important pulse crop after chickpea [5]. It covered an area of around 42.29 lakh ha, producing 37.54 lakh tones with the average productivity of 806 kg/ha approximately during 2019-20 [2]. Maharashtra, Karnataka, Madhya Pradesh, Uttar Pradesh, Gujarat, Jharkhand, Telangana and Andhra Pradesh are the major producers of pigeonpea with more than 90% share in total pigeonpea production [3]. Karnataka has the highest area under pigeonpea (13 lakh ha), but the highest production was recorded in Maharashtra (9.71 lakh tones) [2]. Pigeonpea can be attacked by more than 100 pathogens [14]. These include fungi, bacteria, viruses, nematodes and phytoplasma. However, only a few of them cause economic losses [10] and the distribution of the major diseases is geographically restricted. The diseases of considerable importance at present are sterility mosaic, Fusarium wilt, Phytophthora blight, Macrophomina root rot, stem canker, and Alternaria blight on the Indian subcontinent. Wilt disease caused by *Fusarium oxysporum* f. sp. *udum* is the most important disease of pigeonpea worldwide. This is the most important soil born disease of pigeonpea and was first described in 1906 from Bihar state, India [7]. Although the disease first appears in patches in a field, it can extend to the entire field if pigeonpea is repeatedly cultivated in same field.

Even though plants are infected at an early stage, the wilt symptoms are not expressed until flowering and podding. The yield loss depends on the stage at which the plants wilt, it can approach 100%. When wilt occurs at the pre pod stage, about 67% when wilt occurs at maturity and 30% when it occurs at the preharvest stages [9]. The yield loss observed above 10-50% and in some years up to 90% in pigeonpea due to *Fusarium* wilt in farmers' fields [21]. Wilt incidence generally increases when the crop is ratooned or retained as perennial. The disease is seed borne and soils born. The annual losses due to wilt has been estimated at \$71 million in India and \$5 million in Eastern Africa.

2. MATERIAL AND METHODS

The laboratory experiment was conducted in the department of Plant Pathology, Collage of Agriculture, Banda University of Agriculture and Technology, Banda, and survey of *Fusarium oxysporum spp. udum* on pigeonpea fields of Banda, Mahoba, Hamirpur, and Chitrakoot districts of Bundelkhand region. Random roving method of survey was carried out to record the severity of *Fusarium* wilt in pigeonpea. The area having randomly selected of wilt incidence and after survey observations of disease incidence were recorded. The disease incidence was recorded at 20 randomly selected plants of each block. The percent disease incidence was assessed by the formula:

$$\text{Disease incidence} = \frac{\text{Total number of infected plants}}{\text{Total number of observed plants}} \times 100$$

Radial growth and colony characterization the sample was taken 5 disease plant (*Fusarium oxysporum spp. Udum*) is collected at different five location which are four corner and one middle part of the field at every block/ tehsil. After collection of diseased samples, the collar region of each sample was washed thoroughly in running tap water to remove the soil particles. Each sample diseased part (*Fusarium oxysporum spp. Udum*) was cut into small bits and at least 5 bits were surface sterilized by dipping in 0.1% mercuric chloride for 1 minute and rinsed three times in sterilized distilled water. The procedure was repeated three time for each sample. The petriplates was incubated at $25 \pm 2^{\circ}\text{C}$ till visible fungal colonies appeared [11]. Radial growth and colony characters were recorded on PDA and PSA solid media. These were sterilized in hot air oven at 160°C for 1 hour. 20 ml of sterilized media was transferred in each sterilized petriplates. All isolates were grown on PDA and PSA plates in triplicate. Inoculated petriplates were inoculated at $25 \pm 2^{\circ}\text{C}$ for 21days. Morphological character pigeonpea wilt pathogen was identified on the basis of their cultural and morphological characteristic. Slides were prepared with cotton blue stain and examined under the compound microscope for morphological characters of the fungus. The pathogenicity was tested at each Disease sample (*Fusarium oxysporum spp. Udum*) was grown up on PDA plates. The fungal soil (*Fusarium oxysporum spp. Udum*) 500g was collected at different five location which are four corner and one middle part of the field at every block/ tehsil. A

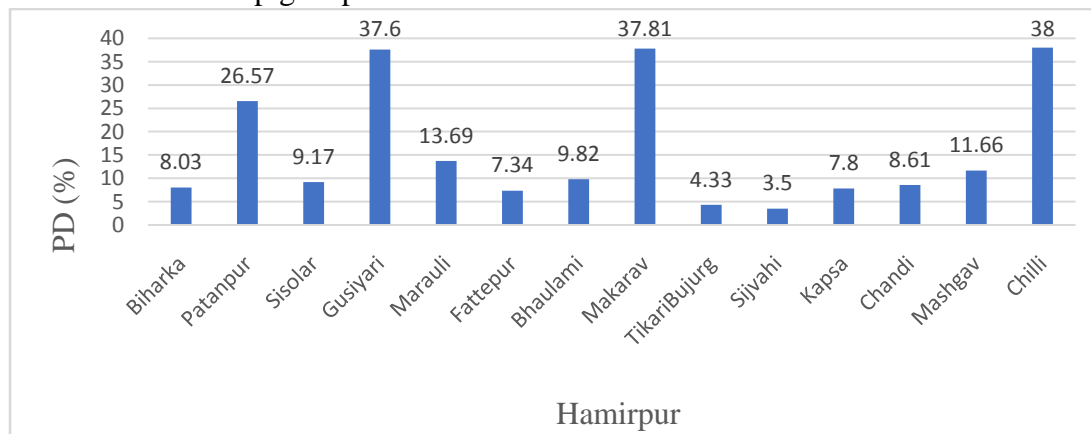
fungus-soil mixture was prepared by mixing 200g of inoculums with 1 kg of autoclaved sand: soil mixture (3:5). 15cm diameter earthen pots were sterilized by formalin (0.1%). These were then filled with fungus-soil mixture for pathogenicity test. Seeds sterilized with mercuric chloride (1%) were sown in each pot. Two actively grown mycelia discs (5mm dia.) from the periphery of 7 days old culture of each isolate were separately inoculated in 500ml conical flasks containing 100g pigeonpea meal medium. The flasks were incubated at 25+₂⁰C for 20 days.

Potato Dextrose Agar (PDA) medium composition: Peeled Potato – 200 gm, Agar-Agar - 20 gm, Dextrose - 20 gm, Distilled water -1000ml, pH - 6-6.5.s

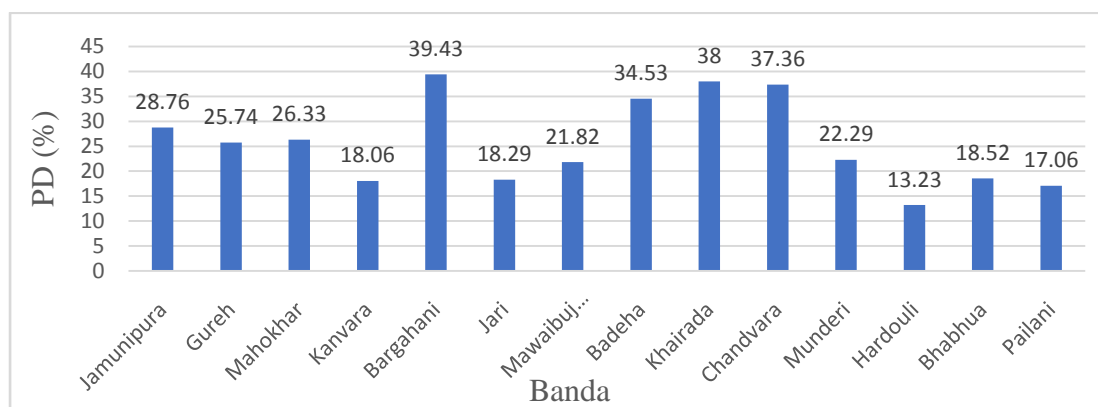
Potato Sucrose Agar (PSA) medium composition: Potato extract – 500 ml, Agar-Agar - 20 gm, Sucrose - 20 gm, Distilled water - 500 ml, P^H - 6- 6.5

3. RESULTS AND DISCUSSION

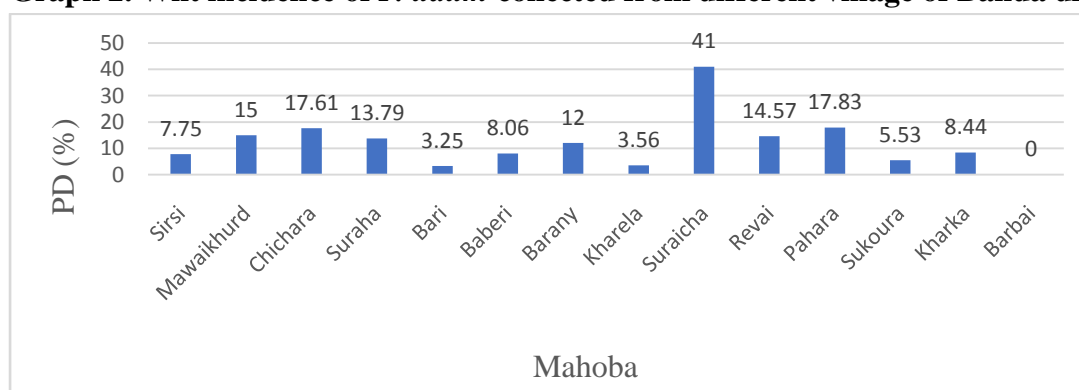
Survey of *Fusarium oxysporum* spp. *udum* on pigeonpea- The average disease incidence ranged between 3.25% (Bari village of Mahoba) to 49.00% (Khoh village of Chitrakoot) from district to districts. The average percentage of wilted plants in Banda district was (25.67%) minimum (13.23 %) being in Hardouli, village of Banda while maximum (39.43%) in Bargahani of village of Banda. The average percentage of wilted plants in Mahoba district was (14.64%) minimum (3.25%) being in Bari village of Mahoba while maximum (41.00%) in Suraicha village of Mahoba. Hamirpur district showed 15.99% average disease incidence, the minimum (3.50%) being in Sijvahi village of Hamirpur, maximum (38.00%) in Chilli village of Hamirpur. The average percentage of wilted plants in Chitrakoot district was 39.06% the minimum (26.00%) being in Kalla village of Chitrakoot, while maximum (49.00%) in Khoh village of Chitrakoot (Table-1). The detailed survey inferred that maximum disease incidence was recorded in Chitrakoot district and followed by Banda, Hamirpur and Mahoba. The similar finding of wilt disease of pigeonpea [17, 23,13]. The detailed survey inferred that maximum disease incidence was recorded in Chitrakoot and followed by Banda, Hamirpur and Mahoba. The incidence of disease has been reported 30-60% at crop maturity and flowering stages by [9]. [15] suggested that this disease depends upon the stage of the crop infection, which approach over 50% and even upto 100% when wilt occurs at the pre pod stage. [8] Reported maximum yield loss up to 90% caused due to *Fusarium udum* in pigeonpea.



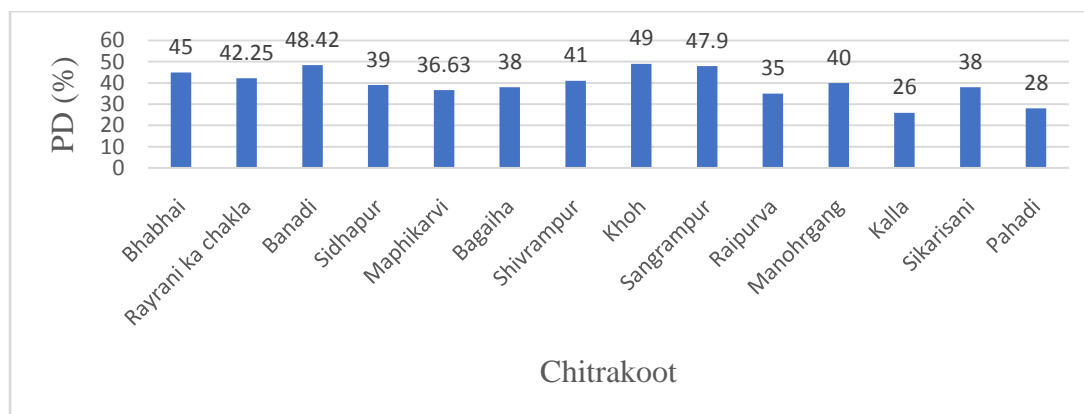
Graph 1. Wilt incidence of *F. udum* collected from different village of Hamirpur district



Graph 2. Wilt incidence of *F. udum* collected from different village of Banda district



Graph 3. Wilt incidence of *F. udum* collected from different village of Mahoba district



Graph 4. Wilt incidence of *F. udum* collected from different village of Chitrakoot district

Colony character- It is clear from the table that isolates differed in their colony characters on different media. Sporulation was moderate to excellent in different isolates. All the isolates were in good growth on both media. There was a considerable variation among all the isolates of *Fusarium udum* (Table-2). PDA and PSA media were best for the growth of *Fusarium udum*. The several workers have been reported that mycelial growth and colony characters on PDA and PSA media [6,11,18,19,20,22].

Table-1. Colony characters of different isolates of *F. udum* on solid media

Media	Isolates	Colour		Aerial mycelium
		Mycelium	Substrate	
	Banda	White	Light Yellow	Fluffy forming concentric rings,
PSA	Hamirpur	White	Light Yellow	Cottony, forming sector
	Mahoba	White	Yellowish	Fluffy, sticky in the middle
	Chitrakoot	White	Light Yellow	Fluffy, sticky
	Banda	White	Yellowish White	Fluffy, Cottony
PDA	Hamirpur	White	Yellowish White	Fluffy, Cottony with sticky mycelium in the middle, forming sectors
	Mahoba	White	Yellowish White	Sticky, Cottony at the point of inoculation
	Chitrakoot	White	Whitish Yellow	Fluffy, sticky



A

B

C

D

Plate 1. Microscopic view of A. micro conidia, B. macro conidia and C & D. onidiophores of *F. udum*

Radial growth- The radial growth of Chitrakoot isolate was maximum on PDA and PSA media consistently on 21 days of inoculation followed by Banda, Hamirpur and Mahoba isolates. Data of the observations were taken after 21 days of inoculation. Chitrakoot isolate

showed the highest radial growth i.e 85.33 mm on PDA followed by banda 84.33 mm, Hamirpur 82.33 mm and Mahoba 81.66 mm. The mean of radial growth on PDA and PSA media in Chitrakoot isolate showed highest 84.99 mm followed by Banda 83.99 mm, Hamirpur 82.16 mm and Mahoba 81.33 mm after 21 days of inoculation. Also, the highest radial growth was observed on PDA in case of Chitrakoot isolate while least radial growth was observed on PDA in Mahoba isolates (Table-3). The maximum radial growth was recorded on PSA in Chitrakoot isolate and minimum in Mahoba isolate. The *Fusarium udum* isolates exhibited high variation in radial mycelial growth and sporulation on PDA. The maximum radial growth was recorded on PSA in Chitrakoot isolate and minimum in Mahoba isolate [12,20,23]

Table 2. Radial growth of different isolates *F. udum* on solid media (mm)

Media	Isolate				Mean
	Banda	Mahoba	Hamirpur	Chitrakoot	
PDA	84.33	81.66	82.33	85.33	83.41
PSA	83.66	81.00	82.00	84.66	82.83
Mean	83.99	81.33	82.16	84.99	

Morphological character- Micro conidia are single or bicelled, hyaline, mostly curved, ovoid to fusoid and scattered and measure 3.80-9.12 x 0.98-2.10 µm. Macro conidia which are hyaline, thin walled, falcate with a distinct foot cell, and an apical cell that decreases in width towards the tip. The most distinguishing characteristic of the macro conidia are their strongly curved or hooked apices. They are mostly 3 septate, less frequently 4-5 septate, very rarely 5 septate and measure 11-21.12 x 1.95 to 3.78 µm.

Pathogenicity test- *Fusarium* wilt symptoms on pigeonpea plants were observed from the ten days of inoculation and the fungus re - isolated from the stems and roots of infected plants on the 16 days. The height and number of leaves was maximum in Mahoba district followed by Hamirpur, Banda and Chitrakoot isolate (Table-3). However, Chitrakoot isolate was found to be highly pathogenic followed by Banda, Mahoba and Hamirpur isolates. The Hamirpur isolate was low pathogenic in compare to Chitrakoot, Banda and Mahoba isolates [12,15,16,23].

Table 3. Pathogenicity test of different isolates of *Fusarium udum* in pot condition

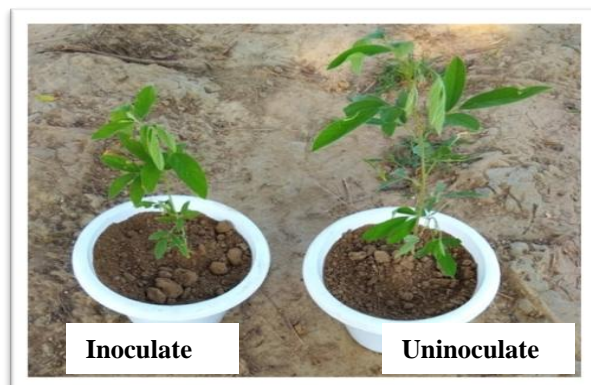
Sr no.	Name of Isolates	Height of uninoculated	Height of inoculated	No of leaves in	No of leaves in	Wilting symptom
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		plant (cm)	plant (cm)	uninoculated plant	inoculated plant	
1.	Banda isolate	39.2	34.2	34.2	23.4	Positive
2.	Mahoba isolate	38.0	35.4	35.0	27.2	Positive
3.	Hamirpur isolate	38.8	35.0	34.5	25.5	Positive
4.	Chitrakoot isolate	39.0	34.0	34.0	20.5	Positive

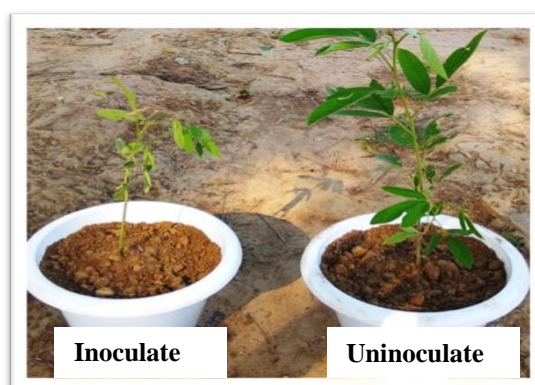
4. CONCLUSION

Pigeonpea is an important pulse crop grown throughout India. The average *Fusarium oxysporum sp. udum* incidence ranged between 3.25% to 49.00% from district to districts. The average incidence percentage of wilted plants in Chitrakoot district was 39.06% followed by Banda district 25.67%, Hamirpur district 15.99% and Mahoba district 14.64% respectively. The height and number of leaves was maximum in Mahoba isolates followed by Hamirpur, Banda and Chitrakoot isolates. The maximum wilting was recorded in Chitrakoot isolate followed by Hamirpur, Banda and Chitrakoot isolates. The highest radial growth was observed in Chitrakoot isolate on PDA while minimum radial growth was observed on PDA in Mahoba isolate. However, the maximum radial growth was a recorded-on PSA in Chitrakoot isolate and minimum radial growth in Mahoba district was isolated.

Banda



Mahoba



Hamirpur



Chitrakoot



Plate 2. Pathogenicity test on pigeonpea

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COMPETING INTERESTS

Authors have declared that no competing interests exist.

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