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MORPHOLOGY AND EFFECT OF DIFFERENT SOLID MEDIA ON THE GROWTH OF THE *FUSARIUM OXYSPORUM F. SP. CICERI* CAUSES FUSARIUM WILT OF CHICK PEA

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Abstract

In vitro studies were conducted to check the growth of the *Fusarium oxysporum f. sp. ciceri* on different solid medium. The fungal growth was observed best on Potato Agar medium followed by Richard's agar medium. Morphological character of the fungus studies on potato Agar medium.

Key word: *fusarium oxysporum f. sp. ciceri,* Solid medium, Morphological.

Background

Chick pea (*Cicer arietinum* L.), 2n = 16 originated near South west Asia and the Mediterranean region as the primary centres of origin with Ethiopia as the secondary centres Vavilov (1926). Chick pea is the third most important pulse crop in the world. It is the good source of protein and calcium. It is affected by various biotic and abiotic stresses. Biotic stresses like Ascochyta blight, Dry root rot, wet root rot, botrytis grey mold, chick pea stunt. Among them, fusarium is most important disease of chick pea causing about 60 per cent yield loss (Singh *et al*., 2007). *F. oxysporum* f.sp. *ciceri* is a soil borne root pathogen, colonizes xylem vessels, blocking them and causing wilting (Bateman *et al.,*1996). Phytopathogenic *Fusarium* causes several diseases of small-grain cereals, including seedling blight and foot rot, fusarium head blight (FHB, also known as 'scab' or ear blight) and ear rot of maize (Parry et al., 1995). The *Fusarium* species *Fusarium graminearum* (teleomorph *Gibberella zeae*), *F. culmorum, F. poae*, *F. avenaceum* (teleomorph *G avenacea*) and *Microdochium nivale* (formerly known a *Fusarium nivale*, teleomorph *Monographella nivalis*) are common pathogens . Booth (1971) reported that *Fusarium oxysporum* produces three types of asexual spores microconidia, macroconidia and chlamydospores. According to Messiaen and Cassini (1981), there is a good deal of variation in spore morphology within the species, even within a specialised form or race, with respect to shape and size of macroconidia and the proportion of microconidia to macroconidia. The aerial mycelium first appears white and may then change to a variety of colors, ranging from violet to dark purple, according to the strain of *F. oxysporum.* The objective of the work were to better understanding of the different media for the growth of the *fusarium oxysporum*.

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Materials and Method

The present investigation based on laboratory experiment was undertaken at the Department of Plant Pathology, C. S. Azad University of Agriculture & Technology, Kanpur, during the year 2014-15.

Isolation and purification of the pathogen

The isolate of *fusarium oxysporum* F. sp.cerci recovered from the affected chickpea roots were used for isolation. The infected roots were thoroughly washed with sterilized water to remove dust and others surface contaminations. The infected parts of roots just touching the healthy portion of the roots were cut into small pieces by the sterilized scalpel. These pieces were thoroughly washed in 3 to 4 times changes of sterilized water after

that dipping into 0.1 percent Mercuric chloride solution for 20-30 seconds with the help of sterilized forceps, then thoroughly and repeatedly washed in 3-4 changes of sterilized water to remove the last traces of Mercuric chloride solution. The excess moisture was removed by drying the pieces between the folds of sterilized blotting papers. These sterilized pieces were then transferred into the sterilized Petri-dishes containing 2 per cent potato dextrose agar medium, 2 to 3 pieces were placed at an equal distance with the help of sterilized forceps in poured Petri-dishes and then the Petri dishes were incubated at 28±1°C temperature. After 24 hours of incubation period, as soon as white mycelia growth was visible in different Petri-dishes. It was transferred into sterilized culture tubes already containing 2 per cent sterilized potato dextrose agar medium. Isolate was identified as *F. oxysporum* f. sp. *ciceri* by morphological criteria (Leslie and Summerell, 2006).

A single microconidial culture was prepared from isolated pathogen. Isolated pathogen was purified and tested for their virulence against susceptible variety "C-104" by using percent disease index (PDI) in field (Nene *et al.,* 1981).

Morphological studies of the fungus

Two percent potato dextrose agar medium was used for study of the colony and mycelia characters of the fungus. Poured Petri dishes were inoculated with the isolate and the following characters were examined after incubation for 10 day at 28±1^{0}C temperature. The important morphological characters of the fungus taken as Mycelium and colony color,conidia and conidiophores (Micro conidia, Macro conidia, Chlamydospores).

Cultural studies of the pathogen:

Following media were used for the growth and mharphological characteristicsof the pathogen

(a) Potato dextrose agar medium (Pealed potato 200 g, Dextros 20.0g, Agar 20.0g, Distilled water 1000 ml)

(b) Czapek's (Dox) Medium (Magnesium sulphate 0.50 g, Sodium nitrate 2.00 g, Dipotassium hydrogen phosphate 1.00g, Ferrous sulphate 0.50g, Potassium chloride 0.50 g, Sucrose30.0g., Distilled water 1000 ml)

(c) Oat meal agar medium (Oatmeal 50.0g, Agar 20.0 g, Distilled water1000 ml)

(d) Sabouraud's agar medium (Glucose 40.0 g, Peptone 10.0g, Distilled Water 1000 ml)

(e) Richard's agar medium (Potassium nitrate 100 g Potassium dihydrogen phosphate 500g Magnesium sulphate 250g Ferric chloride 020g Sucrose 500g Distilled water 1000 ml)

(f) Brown's starch agar medium (Tripotassium phosphate 1.25 g, Magnesium sulphate 0.75 g, Asparagine 2.00 g, Glucose 2.00g, Distilled water 1000 ml)

(g) Malt Extract agar medium (Malt Extract 25g, Agar-Agar 20g, Distilled water 1000 ml).

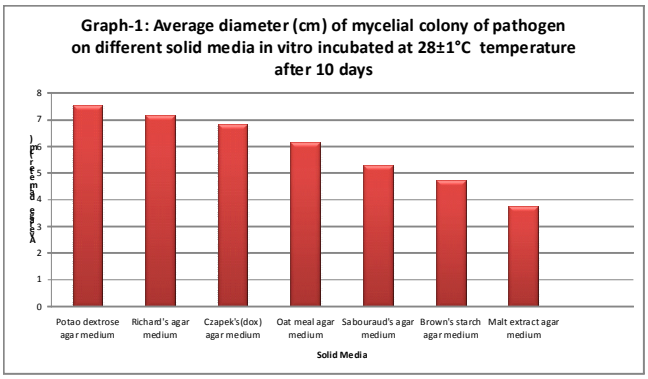
After preparing the solid media, each medium was poured in Petri-dishes. Three replications were kept for each medium in the Petri-dishes. The media were sterilized in usual manner and previously sterilized Petri-dishes containing media were inoculated with culture pieces of equal size, cut by the sterilized cork-borer. After inoculation, the Petri-dishes were incubated at room temperature (28±1^{0}C) and after 10 days of the incubation, the fungal growth was recorded by measuring the diameter of the growth in Petri dishes to know the best medium for the growth of the fungus. Complete randomized design with three replicates was used for the experiment.

Results

The pathogen was allowed to grow on the seven different solid medium for 10 days at 28±1^{0}C temperature. Out of them, potato dextrose agar medium supported the best growth of the fungus followed by Richard's, Czapek's (Dox) agar medium, Richard's agar medium and Oat meal agar medium. Malt extract agar medium supported the lowest growth of the fungus as shown in the data represented in table no.1. Mycelium and Colony of the fungus produce white to deep light pink color, aerial mycelium, hyphae were septate, profusely branched, sporodochia were rarely produced, but if

Table 1: Average diameter (cm) of mycelial colony of pathogen on different solid media in vitro incubated at 28±1^{0}C.

S. No.	Solid media	Average diameter (cm)	Sporulation
1.	Potato dextrose agar medium	7.55	Excellent
2.	Richard's agar medium	7.14	Good
3.	Czapek's (dox) agar medium	6.83	Good
4.	Oat meal agar medium	6.41	Good
5.	Sabouraud's agar medium	5.27	Fair
6.	Brown's starch agar medium	04.69	Fair
7.	Malt extract agar medium	3.71	Fair
	C.D. at 5%	0.1615	



produced these were completely covered with aerial mycelium. Micro conidia were oval ellipsoid, cylindrical, straight or curved, 2.5-3.5 × 5-11 µm in size produced from single short lateral phialides. Macro conidia were ellipsoid oval and cylindrical and posses generally 3- 5 septa and 3.5-4.5 × 25-65µm , Chlamydospores were formed in the old culture at high temperature measuring 7.0-8.0 × 3.5 µm. They were globose, formed singly or impairs, intercalary or on short lateral branches.

Discussion

The pathogen was isolated from the diseased material obtained from wilt sick plot of legume section, C.S. Azad University of Agriculture and Technology, Kanpur during December, 2014. Pathogen was purified by single spore method, pathogenicity of purified pathogen was proved in pot and Koch's postulates were fully satisfied with a view of identifying the pathogen, various morphological, cultural and pathological characters were studied in detail.

The colony of the fungus on potato dextrose agar medium were white radiating with wine-red pigmentation in the later stage which was clearly visible from the bottom side of the Petri-plate. The mycelium of the fungus was properly branched creeping, hyaline, cylindrical, septate and measuring 3.2 to 4.6 µm in width.

The micro conidia of the fungus were hyaline single celled oval to cylindrical, straight on branched conidiophores. They are fusoid with pointed ends, hyaline septate (3-9 septate) and measuring 3.4-45 × 25-65 µm. The chlamydospores were produced in the old culture, which were globose to sub globose, thick walled, smooth surfaced and measuring 7.0-8.0 × 3.5 µm. Morphological characters of the present fungus was observed reported by Pad Wick., (1939 and 1942), Haware *et al.,* (1978), Jalali and Harichand., (1991), Chakravarty and Gupta., (1995).

In order to find out suitable media for the growth of the fungus, the pathogen was grown on seven different natural synthetic and semi synthetic media in solid forms. From the results it was found that potato dextrose agar medium supported the best vegetative growth of the pathogen and gave excellent sporulation (dox) agar medium also solid form followed by Richard's agar medium & Czapek's (dox) agar medium. On the nest of media the growth of pathogen was good but poor in Malt extract agar medium. These findings were more or less similar to the findings of Chakravarty and Gupta (1995).

These studies will help in further studies of pathogen and its identification.

References

Bateman, G. L., E. Ward and H. Kwaœna (1996). Relationships among Fusarium spp. estimated by comparing restriction fragment length polymorphisms in polymerase chain reaction-amplified nuclear rDNA. *Canadian Journal of Microbiology*, **42(12)**: 1232-1240.

Booth, C. (1971). 'The genus *Fusarium.*' (Commonwealth Mycological Institute: Kew).

Chakraborty, A. and P.K.J. Gupta (1995). Factors affecting chlamydospore formation by *Fusarium udum*, the causal organism of wilt of pigeonpea. *J. Mycopath Res.,* **33(2)**: 71-75.

Comparing restriction fragment length polymorphism in polymerase chain reaction amplified nuclear DNA.

Ferreira, D.F. Sisvar (2008): um programa para análises estatísticas e ensino de estatística. *Revista Symposium*, v. 6: 36-41.

Haware, M.P., Y.L. Nene and R. Rejeshwari (1978). *Phytopathology,* **68**: 1364-1367.

Jalali, B.L. and Harichand (1991). Chickpea wilt. In Diseases of International importance, Vol. 1 Eds. Mukhoppadhyay, A.N.; Chaube, H.S. Singh, U.S. and Kumar, *J. Prentice Hall,* New Jersey, U.S.A.

Leslie, J.F. and B.A. Summerell (2006) The *Fusarium*, Laboratory Manual, Blackwell Publishing, pp. 1- 388.

Messiaen, C.M. and R. Cassini (1981). Taxonomy of Fusarium. *Fusarium. Diseases,biology and Taxonomy*, 427-445.

Nene, Y. L., and M.V. Reddy (1987). Chickpea diseases and their control. *Chickpea diseases and their control.*, 233-270.

Padwick, G.W. (1939). The genus *Fusarium* I, own occurrence in India. *Indian Agric. Sci.,* **9**: 17 1-184.

Padwick, G.W. (1942). Some problems on control of soil borne fungal disease in plants. *Anniv. Vol. R. Bot. Gdn*. Calcutta, pp. 213-220.

Singh, Y. P. and Sumbali Gupta (2007). Efficacy of leaf extract and essential oils of some plant species against *Penicillium expansum* rot of apples. *Ann. of Pl. Protec. Sci.,* **15(1)**: 135-139.

Vavilov, N.I. (1926). Studies on the Origin of Cultivated Plants., Leningrad, pp. 129-238.