Official publication of Pakistan Phytopathological Society
Pakistan Journal of Phytopathology
ISSN: 1019-763X (Print), 2305-0284 (Online)
http://www.pakps.com

DETECTION AND CHARACTERIZATION OF FUNGAL WILT OF DIOON PALM (*DIOON SPINULOSUM)* IN PAKISTAN

Muhammad Hassan, Imran U. Haq, Anjum Faraz*, Amer Habib, Muhammad K. Sarwar

Department of Plant Pathology, University of Agriculture, Faisalabad, Pakistan.

A B S T R A C T

Ornamental industry has been flourished quickly in recent years in Pakistan. Fusarium wilts are major threat to ornamental plant industry of the world. In current study major ornamental growing sites were focused for the disease assessment and sample collection. Maximum disease incidence of 50% and minimum disease incidence of 16.67% was observed from District Kasur and Faisalabad respectively. Frequently isolated fungus was characterized as *Fusarium oxysporum* on morphological basis. Confirmation of *Fusarium oxysporum* as causal agent of Dioon wilt was tested by fulfilling the Koch's postulates. Based on morphological characterization and pathogenicity test it is claimed that *F. oxysporum* is the cause of Dioon palm wilt in Pakistan. The basic knowledge generated through this publication could be applied for the prevention of Fusarium wilt of *Dioon spinulosum* in the country.

Keywords: Etiology, *Fusarium oxysporum*, cycads, perennial ornamental

INTRODUCTION

Ornamental industry has been flourished quickly in past few decades in the world. Ornamental include foliage plants, garden plants, flower bulbs, cut flowers, potted garden plants, deciduous and evergreen shrubs and trees (Gullino *et al*., 2015). Ornamental plants can be grown either in indoor or outdoor environment (Daughtrey and Benson, 2005). Fusarium wilt is major disease of ornamental palms such as queen palm, canary island date palm and washingtonia palm (Swart *et al*., 1999). *Dioon spinulosum* native to Mexico is a cycad (Stevenson and Osborne, 1993). Cycads are ornamental plants and also used as food and production of highly nutritious flour as crude material. Approximately 9 genera and 300 different species of cycads have been reported from 1985 to 2010 and about 185 species cultivated as ornamental plants (Haynes, 2011 and Rivadeneyra-Dominguez and Rodriguez-Land, 2014). Fusarium wilts are major hindrance in the cultivation and production of ornamental plants throughout world (Gullino *et al*., 2015). Fusarium species are considered among major mycoflora of Pakistani soils that is responsible of almost 50% field and horticultural crop losses (Akhtar, 2000).

Submitted: November 03, 2019
Revised: December 02, 2019
Accepted for Publication: December 26, 2019
* Corresponding Author:
Email: faraz2537@gmail.com

MATERIALS AND METHODS

Field surveys of major ornamental growing sites were arranged for disease assessment. Different nurseries, parks and lawns of District Faisalabad and Kasur were visited. Disease incidence (DI= Infected plants/total plants observed) of the surveyed sites was calculated. Field samples were collected from the infected plant parts. Samples were submitted to Fungal Molecular Biology Lab, Department of Plant Pathology UAF for isolation of the pathogenic fungi. Samples were surface sterilized in 70% ethyl alcohol followed by 2 washings in sterile distilled water and placed on PDA. Whitish fungal growth was observed on the bated samples after 20-24 hours of incubation at 26^0C. A portion from these mycelia was transferred on PDA for the purification of the fungus. Purified culture was submitted to fungal molecular biology culture collection (FMB-CC-UAF) with accession no FMB 0116. Fungus was characterized morphologically by macro (visualizing colony pattern, color) and microscopic (Shape and size of conidia)

characters. The pathogen was multiplied on liquid broth (LB). For that 3 mm bit from fresh culture was transferred in 100 mL LB. Pathogenicity test was conducted on healthy nursery plants.

Table No. 1 Disease incidence of different nurseries at different locations

Sr. No.	Site		Total plants observed	Infected plants	Disease incidence
	Nursery Name	District			
1	Garden Nursery	Faisalabad	10	3	30%
2	Pakistan Nursery	Faisalabad	13	3	23.07%
3	Neelam Nursery	Faisalabad	12	2	16.67%
4	Baba Abdul-rehman market	Kasur	8	4	50%
5	Kareem nursery farm	Kasur	11	3	27.27%
6	Madina Nursery	Kasur	16	7	43%

Figure 1. Dioon palm showing wilt symptoms

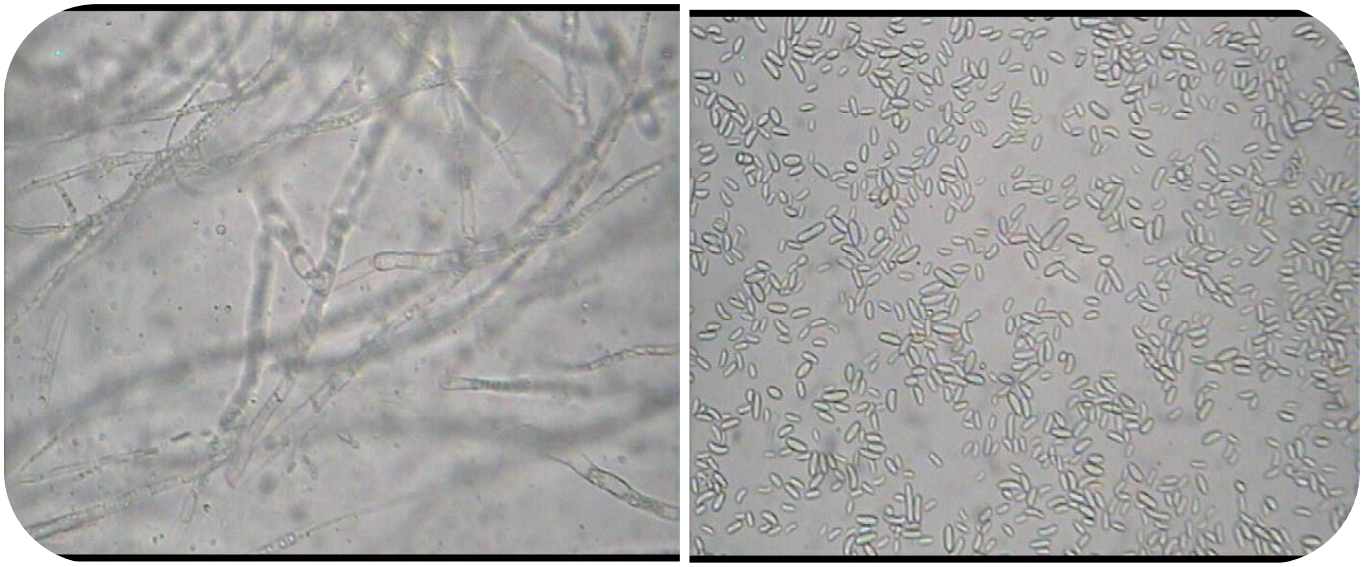

Figure 2. Mycelia and microconidia of *F. oxysporum*

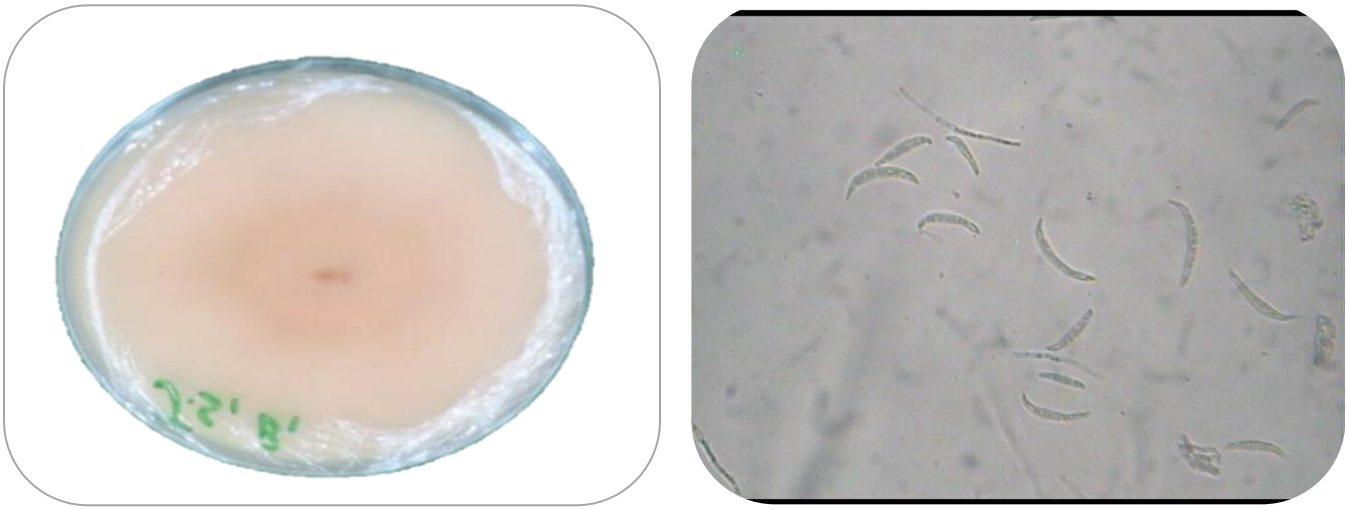

Figure 3. Pure culture of *F. oxysporum* on PDA and Macroconidia of *F. oxysporum*

RESULTS AND DISCUSSION

On site typical wilt symptoms yellowing or necrosis extended along the midrib and partial to complete wilting of leaves were commonly observed on infected plants. Maximum disease incidence of 50% was observed from Baba Abdul-rehman Market District Kasur and minimum disease incidence 16.67% was calculated from Neelum nursery Faisalabad. Fusarium species were isolated from all the collected samples. Fungal mycelia were sparse, abundant with white to pinkish in color. Abundant micro and macro conidia were observed in seven days old culture. *Fusarium oxysporum* produce three different types of asexual spores; micro and macro conidia and chlamydospores (Correll, 1991). Microconidia were aseptate and oval to kidney shaped. Macroconidia were curved with 3 to 5 septations. Colony morphology of *Fusarium oxysporum* varies greatly on PDA. Generally, it gives pale to whitish appearance when cultured on PDA and microconidia have no septations and macroconidia varies from medium to large, slightly curved and possess 3 septations. Spore suspension of $1\times10_6$ of *Fusarium oxysporum* @ 25 mL was applied into the root zone of healthy plants while control plants with 25 mL of distilled water and plants were allowed for initiation of symptoms. Yellowing followed by wilting of leaves was observed after 90 days of inoculation while plants treated with distilled water remained disease free throughout the experiment. *F. oxysporum* was successfully re-isolated from the inoculated plants. *Fusarium oxysporum* is a common wilt pathogen of world (Nelson, 1983 and Gullino *et al*., 2015). Fusarium wilts caused by different forma specialis of *F. oxysporum* are most destructive diseases of many crops including ornamental plants (Elmer, 2008). *F. oxysporum* is capable to affect several herbaceous and some woody ornamentals at any growth stages (Sinclair and Lyon, 2005).

REFERENCES

Akhtar, C. 1977. Biological control of some plant diseases lacking genetic resistance of the host crops in Pakistan. Annals of the New York Academy of Sciences.287:45-56.

Correll, J.C. 1991. The relationship between formae speciales, races, and vegetative compatibility groups in *Fusarium oxysporum*. Phytopathology.81:1061-1064.

Daughtrey, M.L. and D.M. Benson. 2005. Principles of Plant Health Management for Ornamental Plants. Annual Review of Phytopathology.43:141-169.

Elmer, W.H. 2008. Preventing spread of Fusarium wilt of Hiemalis begonias in the greenhouse. Crop Protection.27:1078-1083.

Gullino, M.L., M.L. Daughtrey, A. Garibaldi and W.H. Elmer. 2015. Fusarium wilts of ornamental crops and their management. Crop Protection.73:50-59.

Haynes, J.L. 2011. World list of cycads: a historical review. IUCN/SSC Cycad Specialist Group. :8- 35.

Leslie, J.F. and B.A. Summerell 2008. The Fusarium laboratory manual, John Wiley & Sons.

Nelson, P.E., T.A. Toussoun and W. Marasas. 1983. Fusarium species: an illustrated manual for identification. The Pennsylvania State Univ. Press: 191-193.

Rivadeneyra-Domínguez, E. and J.F. Rodríguez-Landa. 2014. Cycads and their association with certain neurodegenerative diseases. Neurología (English Edition) 29:517-522.

Sinclair, W.A. and H.H. Lyon 2005. Diseases of trees and shrubs, Comstock Publishing Associates.

Swart, L., S. Denman, S.C. Lamprecht and P.W. Crous. 1999. Fusarium wilt: A new disease of cultivated Protea in Southern Africa. Australasian Plant Pathology. 28:156.

Contribution of Authors:

Muhammad Hassan	:	Conducted research and data recording
Imran U. Haq	:	Supervised the research activities being the main Supervisor of Mr. Hassan
Anjum Faraz	:	Principle author
Amer Habib	:	Member Supervisory committee/Co-supervisor
Muhammad K. Sarwar	:	Data analysis and interpretation