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# Occurrence and pathogenicity of various pathogenic fungi on cucurbits from Kermanshah province, Iran

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Cucurbits ( Cucurbitaceae) are among the important plant families providing edible products to humans worldwide. It is known fact that various pathogenic fungi can attack the plants and cause diseases and resultantly yield losses. The aim of this study was to isolate and identify the plant pathogenic fungi from cucurbit plants and to evaluate their pathogenicity on cucurbits. A total of 101 cucurbit samples infected with diseases were collected from different places in Kermanshah Province, Iran. From these samples 101 strains of various pathogenic fungi were isolated and identified through morphological characters. All strains were belongs to Fusarium oxysporum, Macrophomina phaseolina, Phytophthora melonis, Phytophthora drechsleri, Pythium aphanidermatum and two morphotypes of Fusarium solani species complex (FSSC). P. aphanidermatum and Phytophthora sp. strains were recovered from the samples collected from irrigated field, while Fusarium and M. phaseolina strains from rain fed field. All these pathogenic fungi were evaluated to study their pathogenicity on cucumber (Cucumis sativum) and honeydew melon (Cucumis melo) . P. melonis, P. drechsleri, F. oxysporum and P. aphanidermatum caused the damping off within 10 - 20 days on both plants tested. The stem rot symptoms were observed on the 7th day after inoculation of F. solani and M. phaseolina on both plants tested. The inoculated fungi were re-isolated from the diseased plants to prove the Koch's postulates. This is the first comprehensive report on identity and distribution of major plant pathogenic fungi causing root and stem rots on cucurbits in west of Iran.

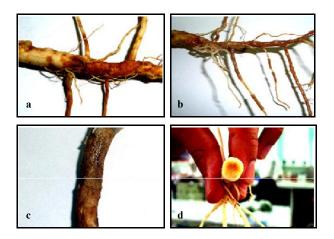
Key words: Root and stem rot, pathogenicity, cucurbits, Iran.

## INTRODUCTION

Annually it is estimated that over 3,000 ha of agricultural area in Kermanshah province are under cucurbits cultivation. Major diseases of cucurbits in Kermanshah province especially in cucumber and melon is sudden death and complete destruction of these economic plants. Various plant pathogenic fungi namely, *Fusarium oxysporum*, *Macrophomina phaseolina*, *Phytophthora melonis*, *Phytophthora drechsleri*, *Pythium aphanidermatum* and two morphotypes of *Fusarium solani* species complex (FSSC) are the major disease causing fungi on cucurbits in Kermanshah province, Iran (Figure 1). The most important pathogens that cause sudden death in

in cucurbits in the entire world are Phytophthora (Erwin and Ribeiro, 1996) and Fusarium sp. (Armstrong and Armstrong, 1981). The genus *Phytophthora* is a serious threat to production of susceptible crops worldwide, particularly cucurbits and solanaceous plants. Phytophthora species are known to infect many species of pepper, tomato and other agronomic and ornamental crops of Solanaceae and Cucurbitaceae families (Zhang et al., 2006). All the cucurbits are susceptible to Phytophthora rot, but squash and pumpkin are the most commonly affected. Cucumber and melon are considered to be somewhat tolerant for Phytophthora (Erwin and Ribeiro, 1996) . Banihashemi (1969) and Sharif and Ershad (1966) first reported the major causal agent of honeydew melon sudden death due to Phytophthora sp. in Iran. P. drechsleri also has been known as a dominant species in root and foot rot in Iran (Ershad and

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**Figure 1.** Diseases caused by pathogenic fungi on cucurbits; a = root rot on watermelon by *Pythium aphanidermatum*, b = root rot on melon by *Phytophthora* sp., c = Stem infection of honeydew melon by *M. phaseolina*, d = brown discoloration of the root phloem in melon caused by *F. oxysporum*.

Mostowfipoor, 1969).

Diseases caused by Fusarium sp. in cucurbits are fast spreading, aggressive and capable of complete crop failures. The most common species of Fusarium that cause vascular wilts in cucurbits in different regions of world are F. oxysporum, F. solani and F. proliferatum (Namiki et al., 1994). Rhizoctonia solani, M. phaseolina, P. aphanidermatum and Olpidium sp. are other pathogens most frequently isolated from the root systems of cucurbit plants (Pivonia, et al., 1997) . F. oxysporum f. sp. melonis, P. drechsleri and Pythium aphanidermatum have been reported as causal agents of melon root and foot rot in Khorasan province (Vahid, 1998) and in Sistan and Baluchistan province, Macrophomina phaseolina as a causal agent of root rot in cucurbits (Safarnezhad, 2004). Our objectives in this research are: (i) To isolate and identify different disease causing pathogenic fungifrom infected cucurbit plants; (ii) To determine the pathogenicity of identified fungi on cucurbits in Kermanshah province, Iran.

#### MATERIALS AND METHODS

#### Sample collection

Infected cucurbit plants were collected from different regions of Kermanshah province, Iran (Table 1). Each sample were stored in a paper envelope and kept in a cool box with dry ice. In the lab, roots and stems of diseased samples were washed and cut into small blocks (1.5 cm) for further analysis.

## Isolation and identification of *Phytophthora* sp. and *P. aphanidermatum*

For isolation of *Phytophthora* spp. and *P. aphanidermatum*, the blocks were rinsed with several changes of sterile distilled water and after desiccation by filter paper (Esmaili-Shirazifard and

Banihashemi, 2008) were placed onto selective medium of Corn Meal Agar-PARP (CMA- PARP) (Kannwischer and Mitchell, 1981) and incubated at 25°C in dark. Identification of *Phytophthora* strains were based on species description of Erwin and Ribeiro (1996) and identification of *Pythium* strains were based on Waterhouse (1967, 1968).

#### Isolation and identification of Fusarium sp. and M. phaseolina

For isolation of Fusarium spp. and M. phaseolina, the blocks were surface sterilized with 1% sodium hypochlorite for 3 min and rinsed in several changes of sterile distilled water. All the sterilized samples were placed onto general medium (water agar) (Burgess et al., 1994) and Pentachloronitrobenzene agar (PPA) plates, a selective medium for Fusarium (Nash and Snyder, 1962). The plates were incubated at 25°C for 24 h. The resulting single-spore Fusarium colonies were transferred to fresh Potato dextrose agar plates and maintained at 4°C for further studies. To study the growth rates and pigment production of Fusarium sp., all strains were transferred onto potato dextrose agar (PDA) plates and incubated at 25°C. Ten replications were maintained for each Fusarium strain. For microscopic observations, all strains of Fusarium were transferred to Carnation leaf agar (CLA) (Fisher et al., 1982), Spezieller nahrstoffarmer agar (SNA) (Nirenberg, 1976), and Potassium chloride agar (KCIA) (Fisher et al., 1983) medium. The species were identified on the basis of macroscopic characteristics. Identification of species was based on species description of Gerlach and Nirenberg (1982) and Leslie and Summerell (2006).

## Pathogenicity test on potato for differentiation of *P. melonis* and *P. drechsleri*

Based on the morphological characters, the two species of *P. melonis* and *P. drechsleri* cannot be easily identified (Ho, 1986). These two species were discriminated by studying their pathogenicity on potato. In this study, the ability of isolates for causing pink rot on potato was considered (Mostowfizadeh-Ghalamfarsa, 2005). Potato (*Solanum tuberosum*) of alpha cultivar was used in this experiment. Initially, tubers were sterilized with 1% sodium hypochlorite (NaClO) (20 min) and then air dried. Then the tubers were inoculated with fresh *Phytopthora* mycelium by putting CMA blocks (7 mm). Inoculated tubers were incubated at ambient temperature for 5 days. For control we used CMA blocks without fungi. After five (5) days of incubation, tubers were cut from inoculation regions and incubated at ambient temperature for 30 min. In this condition infected tubers to *P. drechsleri* would be pink color.

## Pathogenicity assay of *Phytophthora* sp. and *P. aphanidermatum* on cucurbits

The pathogenicity of *Phytophthora* and *Pythium* species isolates obtained from cucurbits were tested on seedlings of healthy cucumber (*Cucumis sativum*) and honeydew melon (*Cucumis melon*) through pot experiment studies. Each pot was filled with 400 g of autoclaved soil mixed with 10 ml of 6 days old cultures of each strain grown in V-8 juice agar at 25°C. The seedlings were planted (one seedling in one pot) in infested soils and the experiments were carried out in the plant house with day and night temperatures of 30 - 35 and 23 - 30°C, respectively. Sterile soils without inoculum served as control. Three replications were maintained for each strain and the experiment was repeated twice. The inoculated fungi were re-isolated from the infected plants to prove the Koch's postulates.

Table 1. Place of sample collection, host and name of the pathogen identified from each sample.

Place of sample collection	Host	Source	Irrigation type	Pathogen identified
DoroodFaraman - MaoquFeh	Cucumber	Root and Crown	Flooding irrigation	P. drechsleri
DoroodFaraman - MaoquFeh	Cucumber	Root	Flooding irrigation	P. drechsleri
DoroodFaraman -MaoquFeh	Cucumber	Root and Crown	Flooding irrigation	P. drechsleri
DoroodFaraman -MaoquFeh	Cucumber	Root and Crown	Flooding irrigation	P. melonis P.
DoroodFaraman -MaoquFeh	Cucumber	Root	Flooding irrigation	melonis P.
DoroodFaraman - MaoquFeh	Cucumber	Crown	Flooding irrigation	melonis P.
DoroodFaraman - MaoquFeh	Cucumber	Root and Crown	Flooding irrigation	melonis P.
DoroodFaraman - MaoquFeh	Cucumber	Crown	Flooding irrigation	melonis P.
DoroodFaraman -MaoquFeh	Cucumber	Crown	Flooding irrigation	melonis P.
Road DoroodFaraman	Cucumber	Root and Crown	Flooding irrigation	melonis P.
Road DoroodFaraman	Cucumber	Crown	Flooding irrigation	melonis P.
Road DoroodFaraman	Cucumber	Root	Flooding irrigation	drechsleri P.
Road DoroodFaraman	Cucumber	Root and Crown	Flooding irrigation	drechsleri P.
Road DoroodFaraman	Cucumber	Root and Crown	Flooding irrigation	drechsleri P.
Road DoroodFaraman	Cucumber	Root	Flooding irrigation	drechsleri P.
Road DoroodFaraman	Cucumber	Root and Crown	Flooding irrigation	melonis F.
Road Kamyaran - Varmele	Honeydew melon	Root and Crown	Rain-Fed	oxysporum F.
Road Kamyaran -Varmele	Honeydew melon	Crown	Rain-Fed	oxysporum F.
Road Kamyaran - Varmele	Honeydew melon	Root and Crown	Rain-Fed	oxysporum F.
Road Kamyaran-Varmele	Honeydew melon	Root	Rain-Fed	oxysporum
Road Kamyaran - Varmele	Honeydew melon	Crown	Furrow irrigation (Gholam-gardeshy)	F. oxysporum
Miandarband -JaFar Abad	Honeydew melon	Root	Furrow irrigation (Gholam-gardeshy)	F. oxysporum
Miandarband - JaFar Abad	Honeydew melon	Root	Furrow irrigation (Gholam-gardeshy)	F. oxysporum
Miandarband - JaFar Abad	Honeydew melon	Root	Furrow irrigation (Gholam-gardeshy)	F. oxysporum
Miandarband -JaFar Abad	Honeydew melon	Root	Furrow irrigation (Gholam-gardeshy)	F. oxysporum
Miandarband -JaFar Abad	Honeydew melon	Root	Furrow irrigation (Gholam-gardeshy)	F. oxysporum
Miandarband -Varmenjeh	Cucumber	Root and Crown	Furrow irrigation (Gholam-gardeshy)	P. drechsleri
Miandarband -Varmenjeh	Cucumber	Crown and Stem	Furrow irrigation (Gholam-gardeshy)	F. solani
Miandarband -Varmenjeh	Cucumber	Root and Crown	Furrow irrigation (Gholam-gardeshy)	P. melonis
Miandarband - Varmenjeh	Cucumber	Crown	Furrow irrigation (Gholam-gardeshy)	P. melonis
Miandarband - Varmenjeh	Cucumber	Root and Crown	Furrow irrigation (Gholam-gardeshy)	P. melonis
Miandarband - Varmenjeh	Cucumber	Root and Crown	Furrow irrigation (Gholam-gardeshy)	P. melonis

#### Table 1. Contd.

Miandarband - Varmenjeh	Cucumber	Root	Furrow irrigation (Gholam-gardeshy)	P. melonis
Miandarband - Varmenjeh	Cucumber	Root and Crown	Furrow irrigation (Gholam-gardeshy)	P. drechsleri
Miandarband - Varmenjeh	Cucumber	Root and Crown	Furrow irrigation (Gholam-gardeshy)	P. drechsleri
Miandarband - Varmenjeh	Cucumber	Crown	Furrow irrigation (Gholam-gardeshy)	P. melonis
Qazanchi - Ahmad Abad	Watermelon	Root and Crown	Furrow irrigation (Gholam-gardeshy)	P. aphanidermatum
Qazanchi - Ahmad Abad	Watermelon	Root	Furrow irrigation (Gholam-gardeshy)	P. aphanidermatum
Qazanchi - Ahmad Abad	Watermelon	Stem	Furrow irrigation (Gholam-gardeshy)	M. phaseolina
Qazanchi - Tazeh Abad	Honeydew melon	Stem	Rain-Fed	M. phaseolina
Qazanchi -Sarablah	Watermelon	Root	Furrow irrigation (Gholam-gardeshy)	P. aphanidermatum
Qazanchi -Ahmad Abad	Watermelon	Root and Crown	Furrow irrigation (Gholam-gardeshy)	P. drechsleri
Qazanchi -Ahmad Abad	Watermelon	Root and Crown	Furrow irrigation (Gholam-gardeshy)	P. aphanidermatum
Qazanchi -Ahmad Abad	Watermelon	Root	Furrow irrigation (Gholam-gardeshy)	P. aphanidermatum
Qazanchi -Docheshmeh	Watermelon	Root and Crown	Furrow irrigation (Gholam-gardeshy)	P. melonis
Road Allah-yari	Honeydew melon	Stem	Rain-Fed	M. phaseolina
Road Allah-yari	Honeydew melon	Root	Rain-Fed	F. oxysporum
Road Allah-yari	Honeydew melon	Root	Rain-Fed	F. oxysporum
Road Allah-yari	Honeydew melon	Root	Rain-Fed	F. oxysporum
Road Allah-yari	Honeydew melon	Root	Rain-Fed	F. oxysporum
Road Allah-yari	Honeydew melon	Root	Rain-Fed	F. oxysporum
Road Allah-yari	Honeydew melon	Root	Rain-Fed	F. oxysporum
Road Allah-yari	Honeydew melon	Root	Rain-Fed	F. oxysporum
Ravansar	Honeydew melon	Root	Rain-Fed	F. oxysporum
Road Ravansar - Kamyaran	Pumpkin	Root	Furrow irrigation (Gholam-gardeshy)	F. oxysporum
Road Ravansar - Kamyaran	Pumpkin	Root	Furrow irrigation (Gholam-gardeshy)	F. oxysporum
Road Ravansar -Kamyaran	Pumpkin	Root	Furrow irrigation (Gholam-gardeshy)	F. oxysporum
Road Javanrood - Salas-Kanigohar-	Honeydew melon	Root	Furrow irrigation (Gholam-gardeshy) irrigation	F. oxysporum
Road Javanrood - Salas-Kanigohar -	Honeydew melon	Stem	Furrow irrigation (Gholam-gardeshy)	F. solani
Kangavar -Pol Shekasteh	Honeydew melon	Stem and Crown	Furrow irrigation (Gholam-gardeshy)	F. solani

### Table 1. Contd.

Kangavar -Pol Shekasteh	Honeydew melon	Stem	Furrow irrigation (Gholam-gardeshy)	F. solani
Kangavar -Rahmat Abad	Cucumber	Root	Flooding irrigation	F. oxysporum
Kangavar -Gaodin	Cucumber	Root	Flooding irrigation	P. melonis
Kangavar	Pumpkin	Root	Furrow irrigation (Gholam-gardeshy)	P. melonis
Kangavar	Honeydew melon	Root	Furrow irrigation (Gholam-gardeshy)	P. melonis
Gaodin	Pumpkin	Root	Furrow irrigation (Gholam-gardeshy)	P. drechsleri
Gaodin				
Continued	Honeydew melon	Root	Furrow irrigation (Gholam-gardeshy)	F. oxysporum
Gaodin	Cucumber	Stem	Furrow irrigation (Gholam-gardeshy)	P. aphanidermatum
Sonqor	Pumpkin	Root and Crown	Furrow irrigation (Gholam-gardeshy)	P. drechsleri
Sonqor	Cucumber	Root and Crown	Furrow irrigation (Gholam-gardeshy)	P. melonis
Road Sonqor -Asad Abad	melon	Root and Crown	Furrow irrigation (Gholam-gardeshy)	P. drechsleri
Road Sonqor -Asad Abad	Cucumber	Root and Crown	Furrow irrigation (Gholam-gardeshy)	P. drechsleri
Road Sonqor -Asad Abad	Cucumber	Stem	Furrow irrigation (Gholam-gardeshy)	M. phaseolina
Harsin	Pumpkin	Root and Crown	Flooding irrigation	P. melonis
Dinavar -Shirkhan	Cucumber	Root and Crown	Flooding irrigation	P. melonis
Dinavar -Shirkhan	Melon	Root	Flooding irrigation	P. melonis
Biotins -Barnaj	Cucumber	Root and Crown	Furrow irrigation (Gholam-gardeshy)	P. melonis
Bisotun -Barnaj	Pumpkin	Crown	Furrow irrigation (Gholam-gardeshy)	P. melonis
Bisotun -Barnaj	Cucumber	Root and Crown	Furrow irrigation (Gholam-gardeshy)	P. melonis
Bisotun -Barnaj	Cucumber	Root and Crown	Furrow irrigation (Gholam-gardeshy)	P. melonis
Bisotun -Hosein Abad	Cucumber	Root and Crow	Furrow irrigation (Gholam-gardeshy)	P. melonis
Bisotun -Hosein Abad	Pumpkin	Root and Crown	Furrow irrigation (Gholam-gardeshy)	M. phaseolina
Bisotun - Barnaj	Cucumber	Crown	Furrow irrigation (Gholam-gardeshy)	M. phaseolina
Kermanshah – Faculty agriculture– Razi university	Cucumber	Root and Crown	Furrow irrigation	P. melonis
Road Gilan Garb – Sarpol Zohab	Melon	Root and Crown	Flooding irrigation	P. melonis
Gahvareh	Cucumber	Crown	Furrow irrigation (Gholam-gardeshy)	P. aphanidermatum
Road Mahi Dasht - Chehar Zabar	Cucumber	Root and Crown	Flooding irrigation	P. melonis

Road Gilan Garb-Cheshmeh Nezami	Cucumber	Root and Crown	Flooding irrigation	P. drechsleri
Road Kermanshah -Sarab NilooFar	Cucumber	Root and Crown	Rain-Fed	P. drechsleri
Road Kermanshah -Sarab NilooFar	Honeydew melon	Root	Rain-Fed	F. oxysporum
Sarab NilooFar	Cucumber	Stem	Rain-Fed	M. phaseolina
Road Koozaran -Boor Boor	Cucumber	Stem and Crown	Flooding irrigation	M. phaseolina
Road Koozaran - Chehar Zabar	Honeydew melon	Root and Crown	Furrow irrigation	P. melonis
Road Paveh -Qeshlaq	Cucumber	Crown	Flooding irrigation	P. melonis
Road Paveh - Qeshlaq	Cucumber	Crown	Flooding irrigation	P. drechsleri
Road Paveh -Ravansar	Cucumber	Root and Crown	Flooding irrigation	P. drechsleri
Paveh - Shemshir	Cucumber	Root and Crown	Flooding irrigation	P. melonis
Paveh - Shemshir	Cucumber	Stem	Flooding irrigation	F. solani
Miandarband -JaFar Abad	Honeydew melon	Stem	Rain-Fed	F. solani
Bisotun - Barnaj	Cucumber	Stemand Crown	Rain-Fed	F. solani
Qazanchi - Ahmad Abad	Watermelon	Stem	Rain-Fed	F. solani

# Pathogenicity assay of *Fusarium* sp. and *M. phaseolina* on cucurbits

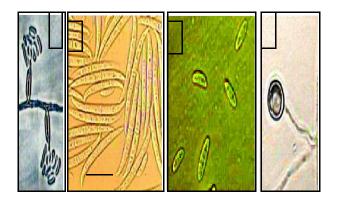
All the identified Fusarium and Macrophomina species were tested for their pathogenicity on apparently healthy cucumber (C. sativum) and honeydew melon (C. melon) plants. For pathogenicity tests the root and stems of the cucumber and honeydew melon were washed and surface sterilized before inoculation. For inoculation, each strain of Fusarium spp. were grown on PDA plates as described by Salleh and Sulaiman (1984). Conidial suspension of each individual strain was prepared by scrapping the mycelium with sterile distilled water onto 7 day-old cultures, shaken thoroughly, and the concentration was adjusted to 2 × 10<sup>6</sup> conidia/ml using haemocytometer. Twenty (20) days old seedlings of cucumber and honeydew melon were brought to the laboratory and their roots were soaked in 20 ml conidial suspension for 20 min for root experiments. For stem experiments twenty milliliter of the conidial suspension of each Fusarium species and M. phaseolina were sprayed on the stems. The control plants were inoculated with 20 ml of sterile distilled water. Three replications were maintained for each strain and the experiment was repeated twice. All the inoculated and controls seedlings of different cucumber plants were placed in the plant house with day and night temperatures of 30 -35 and 23 - 30°C, respectively. Development of symptoms on plants inoculated by fungi and controls were observed continuously every 2 days for 4 weeks. The inoculated fungi were re-isolated from the infected plants to prove the Koch's postulates.

#### RESULTS

In this study, a total of 101 fungal strains were isolated from 101 diseased cucurbit plants. Most of the *Phytophthora* and *Pythium* strains were recovered from irrigated fields and *Fusarium* and *Macrophomina* strains from rain-fed fields. Based on their morphological

characteristics, these strains were identified as P. melonis, P. drechsleri, Pythium aphanidermatum, F. oxysporum, F. solani and M. phaseolina. Differentiation of P. melonis and P. drechsleri based on morphological characterization (homothallism, amphigynous antheridia, the semipapillate sporangia and production of chlamydospores) is not easy. These two species were discriminated by use of the reaction on potato. P. aphanidermatum were identified based on morphological characterization by daily growth rate on V-8 juice agar at 30°C. Main hyphae up to 7.5 µm wide and oospores aplerotic, spherical, smooth, average 41 µm in diameter. Sporangia consisting of terminal complexes of swollen hyphal branches of various length and germinated by extension of long exit tube and vesicle formation and zoospore discharge. Vesicles average 41 µm in diameter. Oogonia globose, terminal, smooth, average 28 µm in diameter, with straight oogonial stalks. Antheridia typically intercalary, usually diclinous and average 14 µm long and 11 µm wide, commonly 1 per oogonium. F. oxysporum strains were identified based on their morphological characterization. This species showed floccose growth, abundant and pale violet aerial mycelia. Pigmentation of reverse colony is pale violet, short monophialides conidiogenous cells, macroconidia usually 3-septate and thin walled, the apical cell is short and basal cell is notch to foot shape, microconidia usually are no septate oval to elliptical, chlamydospores usually singly or in pair (Figure 2).

In morphotype I of *F. solani,* pigmentation of reverse colony is red, macroconidia mostly 5 septate and thick walls. Apical cell is tapered and basal cell foot shape.



**Figure 2.** Morphological characters of *F. oxysporum.* a= Conidiophores, b= Macroconidia, c= Microconidia, d= Chlamydospore (scale bar= 20 µm).

Microconidia are ellipsoid to truncate and clavate 0 - 2 septate. Chlamydospores are with smooth outer walls (Figure 3). In morphotype II of *F. solani*, pigmentations of reverse colony are white to yellow, macroconidia 3 - 4 septate but mostly 3 septate. Apical cell is round curve and short, basal cell is notch to foot shape. Microconidia are mostly 0 - 1 septate and rainy form shape. Chlamydospores usually rough outer walls (Figure 4). In *M. phaseolina* isolates, shape of colony varied from radial to irregular, mycelium production was intermediate in the most of isolates and colour of mycelium is white to dark brown. Pycnidium is in black colour and pycnospores are no septate.

The results of the pathogenicity test revealed that 34 strains of P. melonis, 19 strains of P. drechsleri, 25 strains of F. oxysporum and seven strains of P. aphanidermatum were the major causal agents of cucumber and honeydew melon root rot. Eight strains of each M. phaseolina and F. solani species complex (FSSC) were the main causal agents of cucurbit crown and stem rot, respectively. P. melonis, P. drechsleri, F. oxysporum and P. aphanidermatum strains were shown that cucumber and honeydew melon seedlings were damped-off in the infested soil within 10 - 20 days. This survey revealed that the mentioned strains caused root rots, water- soaked lesions on stems, and damping-off of both plants tested. Their initial symptoms were observed on the 4th day after inoculation. F. solani and M. phaseolina caused stem rot on both plants tested. Their initial symptoms were observed on seven (7) days after inoculation on water-soaked lesions on stems. The inoculated fungi were consistently isolated from the diseased plants again to prove Koch's postulates, but not from control plants.

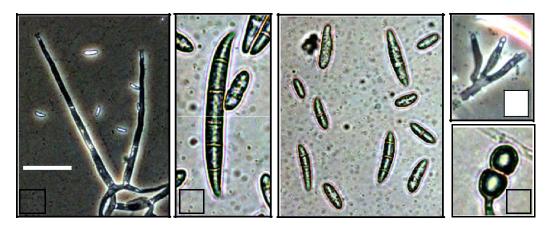
#### DISCUSSION

Soil borne plant pathogens such as Phytophthora and

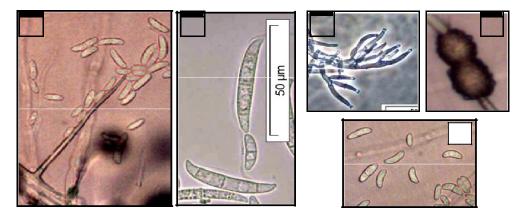
Fusarium sp. can significantly reduce yield and quality in cucurbit crops (Matuo and Snyder, 1973; Ho, 1986; Esmaili-Shirazifard and Banihashemi, 2008). Most of the pathogens identified in this study have ability to grow on a wide range of substrates and have efficient mecha-nisms for dispersal. Also some of the Phytophthora and Fusarium sp. often survive in soil in the form of chlamydospores for many years and may cause diseases to cucurbit plants when the environmental conditions favors fro pathogen (Burgess, 1981; Zheng, 1997). Therefore, accurate identification of pathogenic fungi is very important to develop proper management practices. The results of this revealed that Phytophthora, Pythium and F. oxysporum species are pathogenic to cucurbits. Two other species, *F. solani* and *M. phaseolina* were also pathogenic on stem rot of cucurbits.

In this investigation we observed that rain- fed cucurbit fields in Kermanshah province showed that severity of the disease is high under certain environmental conditions in median of summer due to moisture stress, and also in irrigated field, the severity of the disease is high due to flooding, and furrow irrigation (Gholam-gardeshy). Because many species of Fusarium can act as opportunistic or weak pathogens that are capable of attack plants only when they are weakened previously by some other stress. Certainly, stresses such as those induced by drought, hail and insects are known to affect the disease (Palmer and Kommedahl, 1969). Generally, Phytophthora and Pythium species produces sporangia on leaves and these sporangia may be fallen on surface of irrigation water especially under flooding and furrow irrigation (Duniway, 1974; Zentmayer and Erwin, 1970) and may attack other plants.

The plant pathogenic fungi can survive in the soil and in host debris for months to years (Zheng, 1997). So the control of *Phytophthora* sp. is often very difficult and the best means of control is to prevent the occurrence of the disease in the initial stages. It is thus essential to know the distribution of mentioned genus and their species on the each region such as Kermanshah province. Also using of cultivation controls means following a proper irrigation schedule, on the other hand, prevalence of dry stress to host and improvement methods of irrigation can controlled the components favorable environment. Phytophthora has been recorded from several parts of the world and pathogenic to many plants, especially to seedlings. In Iran, this fungus has been reported to cause root rot on cucurbits but not in Kermanshah province (Esmaili-Shirazifard and Banihashemi, 2008). Our pathogenicity assay also revealed that Phytophthora can cause root rot to cucurbit plants. This study demonstrates the occurrence of pathogenic fungi in cucurbit fields and can cause diseases to cucurbit plants at any stages. As far as we know, this is the first comprehensive report on identification of several pathogenic species and their pathogenicity on cucurbits from Kermanshah province, Iran.



**Figure 3.** Morphological characters of *F. solani* (morphotype I); a = Conidiophores, b = Macroconidia, c = Microconidia, d = Sporodochia conidiophores, e = Chlamydospore (scale bar = 25 µm).



**Figure 4.** Morphological characters of *F. solani* (morphotype II); a = Conidiophores, b= Macroconidia, c= Sporodochia conidiophores, d= Chlamydospores, e= Microconidia.

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#### REFERENCES

- Armstrong GM, Armstrong JK (1981). Formae specials and races of *Fusarium oxysporum* causing wilt disease. In P. E. Nelson, T. A. Toussoun, and R. J. Cook (ed.), *Fusarium*: disease, biology, and and taxonomy The Pennsylvania State University disease, biology, and taxonomy. The Pennsylvania State University Press, University Park, pp. 391-399.
- Banihashemi Z (1969). Cucurbit wilt and root rot diseases in Iran. Proc. 2nd. Plant. Prot. Cong. Iran. pp. 97-98.
- Burgess LW (1981). General ecology of the fusaria. In P. E. Nelson, T. A. Toussoun, and R. J. Cook (ed.). *Fusarium*: diseases, biology, and taxonomy. Pennsylvania State University Press, University Park. pp. 225-235.
- Burgess LW, Summerell BA, Bullock P, Backhouse D (1994). Laboratory manual for *Fusarium* research, Department of crop science. University of Sydney, Sydney, Australia. 3rd ed. p. 133.

- Duniway JM (1974). Formation of sporangia by *Phytophthora drechsleri* in soil at high matric potentials. Can. J. Bot. 53: 1270-1275.
- Ershad D, Mostowfipoor P (1969). The die-back or root rot of cucurbits in Iran. J. Plant Pathol. 5: 35-48.
- Erwin DC, Ribeiro OK (1996). Phytophthora Diseases Worldwide. American Phytopathological Society, St. Paul, MN.
- Esmaili-Shirazifard E, Banihashemi Z (2008). The role of *phytophthora melonis* and p. drechsleri in cucurbit root rot in Iran. Iranian J. Plant Pathol. 44: 54-72.
- Fisher NL, Burgess LW, Toussoun TA, Nelson PE (1982). Carnation leaves as a substrate and for preserving *Fusarium* species. Phytopathol. 72: 151-153.
- Fisher NL, Marasas WFO, Toussoun TA (1983). Taxonomic importance of microconidial chains in *Fusarium* section *Liseola* and effects of water potential on their formation. Mycologia. 75: 693-698.
- Gerlach W, Nirenberg H (1982). The genus *Fusarium* a pictorial atlas. Mitt. Biol. Bundesanst. Land Forstwirtsch. Berlin Dahlem 209:1-406.
- Ho HH (1986). *Phytophthora melonis* and *P. sinensis* synonymous with *P. drechsleri*. Mycologia. 78: 907-912.
- Kannwischer ME, Mitchell DJ (1981). Relationships of numbers of spores of *Phytophthora parasitica* var. *nicotianae* to infection and mortality of tobacco. Phytopathol. 71: 69-73.
- Leslie JF, Summerell BA (2006). The *Fusarium* Laboratory Manual. UK: Blackwell Publish Ltd. p. 388.
- Matuo T, Snyder WC (1973). Use of morphology and mating populations in the identification of formae speciales in *Fusarium*

solani. Phytopathol., 63: 562-565.

- Mostowfizadeh-Ghalamfarsa R (2005). Phylogeny, Taxonomy and Genetic Diversity of *Phytophthora cryptogea* and *P. drechsleri*. Ph. D. Thesis Shiraz. Univ. Shiraz. Iran. p. 172.
- Namiki F, Shiomi T, Kayamura T, Tsuge T (1994). Characterization of the formae specials of *Fusarium oxysporum* causing wilts of cucurbits by DNA fingerprinting with nuclear repetitive DNA sequences. Appl. Environ. Microbiol. 60: 2684-2691.
- Nash SM, Snyder WC (1962). Quantitative and estimations by plat counts of propagules of the bean rot *Fusarium* in field soils. Phytopathol. 73: 458-462
- Nirenberg HL (1976). Untersuchungen uber die morphologische und biologische differenzierung in der *Fusarium* section *Liseola*. Mitteilungen aus der biologischen bundesanstalt fur land–und forstwirtschaft (Berlin-Dahlem) 169: 1-117.
- Palmer LT, Kommedahl T (1969). Root-infecting *Fusarium* species in relation to rootworm infestations in corn. Phytopathol. 59: 1613-1617.
- Pivonia S, Cohen R, Kafkafi U, Ben Zeev IS, Katan J (1997). Sudden wilt of melons in southern Israel: Fungal agents and relationship with plant development. Plant Dis. 81: 1264-1268.
- Safarnezhad MR (2004). Investigation root rot of cucurbits in Sistan region. Proceedings of 16th Iranian Plant Protection Congress, Tabriz, Iran: 262.
- Salleh B, Sulaiman B (1984). *Fusarium* associated with naturally diseases plants in Penang. J. Plant Protec. Tropics 1: 47-53.

- Sharif G, Ershad D (1966). A List of Fungi on Cultivated Plants. Shrubs and Trees of Iran. Plant Pest and Disease Research Instute Evin, Tehran, Iran p. 89.
- Vahid J (1998). Investigation foot and root rot of honeydew melon in Khorasan provinces. M. Sc. Thesis submitted to the College of Agriculture, University of Firdausi Mashhad p. 95.
- Waterhouse GM (1967). Key to *Pythium* pringsheim. C.M.I. Mycol. Pap. 109: 1-15.
- Waterhouse GM (1968). The genus *Pythium* pringsheim diagnoses (or descriptions) and figures from original papers. C.M.I. Mycol. Pap. 110: 1-71.
- Zentmayer GA, Erwin DC (1970). Development and reproduction of *Phytophthora* spp. Phytopathol. 60: 1120-1127.
- Zhang ZG, Li YQ, Fan H, Wang YC, Zheng XB (2006). Molecular detection of *Phytophthora capsici* in infected plant tissues, soil and water. Plant Pathol. 55: 770-775.
- Zheng XB (1997). *Phytophthora* and Methods in *Phytophthora*. Beijing, China: Agricultural Press.