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Vegetative compatibility, host range and pathogenicity of *Verticillium dahliae* isolates in Iran

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Abstract

Verticillium wilt is an economically important disease which inflicts serious losses in potato, cotton, alfalfa, some vegetable crops and fruit trees and occasionally ornamentals. *Verticillium dahliae*, infected cultivated species and weeds were collected from several areas in Iran during tewelve years from 1993-2005 and studied for their vegetative compatibility, host range and pathogenecity. The pathogen was isolated from 27 species belonging to 24 genera and 15 families of plants but was most frequently isolated from *Solanaceae, Cucurbitaceae, Oleaceae* and *Rosaceae* hosts. The morphology of *V. dahliae* isolates on Czapecks agar and water agar media were different especially for micosclerotial appearance time (4-19 days), pigmented zone of colony (37.8-48.33 mm) and microsclerotial morphology (abundant, irregular and elongated shaped or more spherical and scattered). The ratio index of length/width of conidia ranged between 2.32 and 2.70 micrometer with an average of 2.43±0.11. Temperature influenced the radial growth ratio of the isolates and the growth response of *V. dahliae* isolates to temperature *in vitro* was quadratic. All isolates were categorized in three groups based on pathogenecity tests on differential test plants (cotton cv. Sahel and eggplant cv. local). 548 *V. dahliae* isolates from different locations and hosts were assigned to vegetative compatibility groups (VCGs) using nitrate-nonutilizing (*nit*) mutants. A higher frequency of nit1/nit3 mutants (93%) were obtained compared to nitM (7%). 51.1% of the isolates were assigned to VCG4B, 25.9% to VCG2A and 23% to VCG1. The results demonstrated that *V. dahliae* isolates assigned to VCGs were closely associated with specific pathogenecity within the group/diverse.

Keywords: Vegetative compatibility; Host range; *Verticillium dahliae*; Pathotypes; Iran

Introduction

The genus *Verticillium* Nees represents one of the world's major pathogens, affecting crop plants mostly in the cool and warm temperate regions, but has also been reported from subtropical and tropical areas. There are some seven major pathogenic species

affecting trees, herbaceous plants, plantation crops and mushrooms: *Verticillium dahliae* Kleb., *Verticillium albo-atrum* Reinke et Berth., *Verticillium nigrescens* Pethybr., *Verticillium nubilum* Pethybr., *Verticillium tricorpus* Isaac., *Verticillium theobromae* (Turc.) Mas. & Hughes and *Verticillium fungicola* (Preuss) Hassebrauk (=*Verticillium malthousei* Ware). Of these the polyphagous wilt pathogens, *V. dahliae* and *V. albo-atrum*, stand out in importance of agriculturally (Devax, 1966; Himelick, 1969; Pegg and Bradly, 2002).

V. dahliae is one of the most important vascular pathogen of plants, especially in tropical and temperate areas of the world (Devax, 1966; Ligoxigakis, 2000; Rudolph, 1931). The pathogen infects many species belonging to several categories of dicotyledons including trees, vegetables, field and forage crops, ornamentals plants and weeds (Levin et al., 2003; Ligoxigakis, 2000; Mamluk and Skaria, 1981; Pegg and Bradly, 2002; Phillips and Burdekin, 1983; Sherf and Macnab, 1986; Slowson, 1987; Smith, 1965; Thanassoulopoulos et al., 1981). However, some records of the pathogen on gymnosperm (Sherf and Macnab, 1986) and cryptogam (Pegg and Bradly, 2002) hosts are also present. *V. dahliae* is a soil-borne pathogen and persists in the soil as microsclerotia that may remain viable for up to 13 years (Schnathorst, 1971). These structures, in the soil, have been considered as the principal source of inoculation in all host plants (Pegg and Bradly, 2002).

Early host list of *V. dahliae* was reported by Van de Meer (Pegg and Bradly, 2002). Since then (not sure) other catalogues of *V. dahliae* hosts were reported by several authors worldwide (Engelhard, 1957; Himelick, 1969; Ligoxigakis, 2000; Pegg and Bradly, 2002; Slowson, 1987; Wolliams, 1966). In Iran, Verticillium wilt was originally noticed in 1952 and 1959 in cotton fields in the eastern (Azerbaijan) and northern (Golestan) provinces, respectively (Ershad, 1974). Later *V. dahliae* was reported on other plants such as almond (Zakii and Pourmansoori, 1995), cotton (Ershad, 1974), cucumber (Ershad, 1974), okra (Fasihiani, 1995), olive (Sanei et al., 1998), peach (Sanei et al., 1998), pistachio (Arab-salmani and Banihashemi, 2000), potato (Ershad, 1974), plum (Abdi et al., 1989), sesame (Fasihiani,1995) and tomato (Ershad, 1974) in different areas of Iran. The establishment of orchards in infested fields where previously cultivated with susceptible hosts is a major contributing factor for the increase of Verticillium wilt of new plants (Sanei et al., 1998).

Wilt diseases are expressed as a complex function of the species and strain of the pathogen as well as degree of host resistance and environmental conditions (Ligoxigakis and Vakalounakis, 1997; Sanei and Nassrollahnejad, 1995). The findings of Schnathorst and Mathre (Schnathorst, 1997; Schnathorst and Mathre, 1966) showed the existence of variation in virulence in the population of *V. dahliae*. The pathotypes of the pathogen include the most virulent, defoliating type and less virulent, nondefoliating type strains based on the inoculation to susceptible cotton cultivars (Schnathorst and Mathre, 1966). Other reports also indicated variation in virulence of nondefoliate pathotypes (Razavi and Sanei, 1997). However, interaction between *V. dahliae* and different hosts are influenced by environmental conditions particularly high temperatures (Sanei and Nassrollahnejad, 1995).

The wide host range of the pathogen and apparently little host specificity makes it impractical to differentiate the large number of *V. dahliae* strains into formae specials or physiological races (Korolev et al., 2000). Vegetative or heterokaryon compatibility is a powerful tool to assign the natural populations of fungi into subgroups based on their

genetic diversity. Studies of vegetative compatibility groups (VCGs) of *V. dahliae* using nitrate-nonutilizing (nit) mutants indicated that *V. dahliae* populations were composed of a limited number of VCGs (Korolev et al., 2000).; Razavi and Sanei, 1997; Sanei et al., 2004). Among isolates collected from a variety of plant species most were classified into three main groups: VCG1, VCG2 (including subgroups 2A and 2B) and VCG4 (including 4A, 4B, 4AB) (Tsror and Levin, 2003).

Although Verticillium wilt disease is a serious disease of field and glasshouse crops, little is known about the host range and strains of the pathogen in Iran. Therefore, study for determination of genetic diversity based on vegetative compatibility; host range of *V. dahliae*, comparison of strains based on morphology and pathogenecity on different hosts was conducted.

Materials and methods

Isolation of V. dahliae

V. dahliae was isolated from cultivated plants and weed species from different provinces during 1993-2005 (Table 1). The diseased plant parts were surface sterilized superficially with alcohol and rinsed thrice with sterile distilled water. Infected roots and stems were cut into small pieces and placed on Czapecks agar medium in 9 cm petri plates. The plates were then incubated in dark at 25±1°C. All plates were evaluated for 15-20 days and fungus was sub-cultured into a fresh Czapecks agar medium.

Morphology of isolates

V. dahliae isolates were cultured in petri plates containing Czapecks agar and/or water agar media. A 0.5cm diameter of agar disc was taken from 4-day old colony of each isolates and sub-cultured into new petri plates (9 cm), with five replicates per isolates. The zone of mycelial growth and colony morphology were evaluated for the isolates.

Pathogenecity tests

Pathogenecity of *V. dahliae* isolates was determined using stem puncture method (Schnathorst and Mathre, 1966). The spores (3×10^6 spores per ml) were obtained from 4-day old mono-conidial culture of isolates on Czapecks agar medium. Pathogenecity of isolates was evaluated on 1-2 years old woody plants and 4 leaf seedlings of other plants. Pathogenecity of isolates was also evaluated on eggplant cv. local and cotton cv. Sahel as differential hosts (Razavi and Sanei, 1997). Each isolates were inoculated on 5 plants of each host. The plants maintained in a glasshouse and control plants received only few drops of sterile distilled water.

Vegetative compatibility

Mono-conidial culture was obtained from each *V. dahliae* isolate and maintained on Czapeck–Dox agar (CDA) at 5°C. Nit mutants from mono-conidial cultures (10 replications for each isolate) were generated on water agar chlorate (WAC) medium (containing 2% agar, 3% potassium chlorate and 0.02% glucose) using previous techniques (Tsror and

Levin, 2003). Cultures on WAC were incubated at 24°C in dark for 20 days and mycelia from growing edges of colonies were transferred onto CDA and grown for 5 days. Partial phenotyping of nit mutants (nit1/nit3, NitM) was carried out by placing two mycelial plugs of each isolate on both CDA and CDA amended with 0.02% hypoxanthin (Korolev et al., 2000). Complementation between nit mutants was tested on CDA. Mycelial blocks (5 mm) of NitM of international *V. dahliae* tester isolates for VCGs (obtained from Z. Banihashemi, Shiraz University) and the tested isolates were placed 1.5 cm apart in a triangular pattern and incubated at 25°C. Complementation was characterized by phototropic growth at the contact zone between the two complementary nit mutants after 14–20 days of incubation.

Data analysis

The results of the tests were analyzed using MSTATC (version 2) statistical software.

Results and discussion

Isolation of *V. dahliae*

Plant species with natural symptoms from which *V. dahliae* was isolated is shown in Table 1. Among 23 plant species tested, 13 species are cultivated and belong to 7 families, while remaining were weeds, ornamental and trees belonging to 6 families (Table 1). For the field crops, although several hosts are known to be susceptible, the frequency of *V.dahliae* isolation from them was variable. Different plant species were known to be hosts of *V. dahliae*, but the frequency of the fungal isolates on plant species belonging to the families' Solanaceae, Cucurbitaceae and Malvaceae were remarkable. Infected plants in the fields were found with various levels of the pathogen, especially in fields previously cropped with *V. dahliae* susceptible crops which gave rise to increasing occurrence of disease (Sanei and Nassrollahnejad, 1995). However, a few number of *V. dahliae* isolates were also obtained from other hosts such as melon, watermelon and pepper.

Woody plants such as maple, pistachio, olive, peach, almond and plum are in the list of Verticillium hosts in Iran. The pathogen was easily isolated from young twigs of infected trees which showed wilt and dieback symptoms. *V. dahliae* was also isolated from symptomless branches of infected plants such as olive trees during each of the stdudy. The pathogen isolated from twigs of all ages but mostly from 2–year old olive brancheswhere the percentage isolation in year 1 to year 3 was 25%, 87% and 63% respectively. Among the weed species, some other cultivated plants have been also reported as host of *V. dahliae* in Greece (Ligoxigakis, 2000) and Canada (Thanassoulopoulos et al., 1981). In this experiment, weeds could grow in infected soil, but visible infection occurred rarely. The pathogen was only isolated from *Aamaranthus retroflexus* L. and *Capsella bursa- pastoris* (L.) Medik among weeds in olive growing gardens. However, in nature, Verticillium was isolated from many plants without showing any symptoms (Pegg and Bradly, 2002; Slowson, 1987).

Table 1. Plant species showing natural wilt symptoms from which *Verticillium dahliae* was isolated in Iran.

Family	Species	Common name
Aceraceae	*Acer platanoides* L.	Maple
Asteraceae	*Helianthus annus* L.	Sunflower
	Lactuca sativa L.	Lettuce
Amaranthaceae	*Amaranthus retroflexus* L.	Pigweed
Anacardiaceae	*Pistachio vera* L.	Pistachio
Asteraceae	*Carthamus tinctorius* L.	Safflower
Brassicaceae (syn.	*Xanthium strumarium* L.	Cocklebur
Cruciferae)	*Capsella bursa-pastoris* (L.) Medik.	Shepherd's purse
Cucurbitaceae	*Citrullus vulgaris* Schrad	Watermelon
	Cucumis melo L.	Muskmelon
	Cucumis sativus L.	Cucumber
Geraniaceae	*Geranium* sp.	Geranium
Fabaceae (syn.	*Pisum sativum* L.	Pea
Leguminosae)	*Vicia faba* L.	Broadbean
	Abutilon theophrasti Medik.	Velvetleaf
Malvaceae	*Gossypium hirsutum* L.	Cotton
	Hibiscus esculentus L.	Okra
	Olea europaea L.	Olive
Oleaceae	*Sesamum indicum* L.	Sesame
Pedaliaceae	*Fragaria ananassa* Duch.	Strawberry
Rosaceae	*Malus sylvestris* Mill.	Apple
	Prunus domestica L.	Plum
	Prunus amygdalus L.	Almond
Solanaceae	*Capsicum annum* L.	Pepper
	Lycoperssicon esculentum Miller.	Tomato
	Solanum melongena L.	Eggplant
	Solanum tuberosum L.	Potato

The results show that a large number of *V. dahliae* can be isolated from greenhouse plants, especially in cucumber greenhouse which may be attributed to the establishment of greenhouse experiments on infected soil that is partially fumigated or not annually fumigated. The general pattern of wilt symptoms found in most herbaceous and woody host is a total or partial loss of torgor originating as flaccidity of the lowest leaf, or a terminal leaflet (in a compound leaf) developing towards the stem. The symptoms spread until the whole plant is affected and in severe cases resulting in death. Associated with these symptoms but depending on the type and age of host, are epilate petioles abscission, occurrence of chlorotic leaves and necrotic symptomsin infected plants. The vascular discoloration is a general internal symptom except for infected olive tree. Cross section of infected olive branches did not show any discoloration but in some cases the cross section of recently infected branches showed a light red color, as reported by other workers (Levin et al., 2003; Sanei et al., 1996).

Morphology of isolates

White colony mycelium growth was observed after 8–16 days of inoculation of plant materials on medium. The color of the colony mycelium turned to black with 5– 7 days based on the isolates. Examined isolates showed diversity in morphology. Some isolates

formed black or dark gray colonies with little or no aerial mycelium. In contrast, most of the isolates formed dark colonies with dense aerial mycelium. Microsclerotia appeared in the mycelial colony after 4-19 days on medium. The characteristics of microsclerotia varied among isolates. Some isolates produced abundant irregular microsclerotia whereas others were more spherical and scattered on water agar medium (Table 2).

The length, width and ratio index of length/width of *V. dahliae* conidia ranged between 3.19-3.74 µm, 1.32-1.42 µm and 2.4-2.7 with average values of 3.56±0.11 µm, 1.43±0.04µm and 2.25±0.23, respectively. The data was clearly different from diploid diameter of *V. dahliae* isolates with a ratio index greater than 2.97 which occasionally was reported from crucifer hosts in other study (Sanei et al., 2004).

Mycelial growth at different temperatures

The results showed that temperature influenced the radial growth rate of isolates. The relation of final growth response (12 days after inoculation, y) of all isolates and temperature (*T*) were quadratic,

$$Y = -57.83 + 11.45T - 0.27T^2 \text{ with } R^2 = 0.941 \text{ and } P<0.01.$$

However, some variations were observed among growth response of *V. dahliae* isolates to different temperatures. From this point all isolates category into two groups based on *T* value (Table 3). These results show the temperature-dependent growth rates of the isolates. With other reports, these data can be related to the pathotypes of the pathogen with 25°C and 27°C optimal temperature (Schnathorst, 1971).

Pathogenecity test

All examined isolates were pathogenic to cotton and eggplant. The pathogenic isolates varied in virulence on different hosts. First group of isolates that incited mild symptoms on cotton were defined as non-defoliating pathotypes (ND). Second group of isolates that caused severe foliar symptoms, defoliating, stunting and often death were defined as defoliating pathotypes. Isolates belonged to cotton that caused mild symptoms on eggplant were defined as ND1, and those that caused moderate to severe symptoms were defined as ND2 (Table 4) and the results were found to be similar to other research findings (Tsror and Levin, 2003). The host specification was not the same in *V. dahliae* isolates. The fungal isolates might exhibit different virulence against different hosts (Sanei and Nassrollahnejad, 1995).

Table 2. Morphological variation of 30 *Verticillium dahliae* isolates collected from different hosts in Iran.

Mean diameter of		Appearance of Microsclerotium on water agar (days)	Microsclerotia morphology on water agar
Growth in culture medium on Czapecks agar (mm/d)	Pigmented zone on Czapecks agar (mm)		
58.83±1.32	48.33±1.856	Mostly 4 –6 exceptionally 9-12	Irregular shaped, elongated (more abundant)
53.2±2.03	39.6±2.768	Mostly 4-9 exceptionally 19	More spherical and scattered
48.6±2.97	37.8±1.068	Mostly 8-9 exceptionally 19	More spherical and scattered

* Observation were made 10-20 days after inoculation for all data five replicates were considered, Value±SE.

Table 3. Effect of temperature on radial growth of *Verticillium dahliae* isolates from Iran.

Groups	No. of isolates	Mean average growth at each temperature (°C)						
		5	10	15	20	25	30	35
I		142a	387a	650a	746a	870a	547a	0a
II		137a	365a	580a	757a	669b	427b	0a
	temperature-dependent growth rates							
	Group I	$Y= -55.21 + 11.23\ T- 0.273\ T^2$				$R^2 = 0.97$		
	Group II	$Y= -59.68 + 11.60T – 0.274\ T^2$				$R^2= 0.93$		
	All isolates	$Y= -57.83 + 11.45\ T – 0.27\ T^2$				$R^2=0.94$		

* In each column, values with different letters are significantly different according to Students t-test ($p<0.05$)

Table 4. Characteristics of *Verticillium dahliae* from several hosts of Iran

Hosts	Symptoms	No. of Isolates	Nit mutants
	Cotton(Sahel) eggplant(Local)		
Acer platanoides		5	
Amaranthus retroflexus		7	
Carthamus thinctorious	Severe	6	
Xanthium strumarium	leaf necrosis wilting defoliate	3	
Capsella bursa-pastoris	(Defoliate pathotype)	9	
Citrullus vulgaris		7	
Cucumis sativus		11	
Gossypium hirsutum		5	
Helianthus annus		20	
Hibiscus esculenthus		18	VCG1
Olea europaea		6	
Malus sylvestris		2	
Prunus domestica		12	
Prunus amygdalus		6	
Pistachio vera		9	
Lycopersicon esculenthum			
Solanum melongena			
Solanum tuberosum			
Cucumis melo	Severe wilting	16	
Cucumis sativus	Leaf Necrosis Non-defoliate	32	
Lactuca sativa	(Non-defoliate Pathotypes, severe	15	
Pisum sativum	ND2)	12	
Vicia faba		9	
Abutilon theophrasti		57	
Gossypium hirsutum		16	
Hibiscus esculentus		52	VCG4B
Malus sylvestris		5	
Olea europaea		8	
*Fragaria * ananassa*		29	
Capsicum annum		12	
Lycopersicon ecsulentum		17	
Solanum tuberosum			
Solanum melongena			
Cucumis melo	Mild symptoms	24	
Gossypium hirsutum	Leaf Necrosis Non-defoliate	32	
Helianthus annus		39	
Olea europaea	(Non-defoliate pathotypes, mild	21	VCG2A
Solanum tuberosum	ND1)	26	
Solanum melongena			

Vegetative compatibility

All isolates produced chlorate–resistant sectors on WAC. Of these mutants 93% were characterized as nit1/nit3 mutants and 7% as NitM mutants. Based on positive complementation reaction between the tested *V. dahliae* isolates originating from Iran and the international tester isolates, 51.09% of the isolates were assigned to VCG4B, 25.9% to VCG2A and 23.01 to VCG1. Seven isolates were not assigned to any specific VCG, because they were self–incompatible did not produce sectors and mutants or produced mutants which showed negative reaction with all of standard tester and the other isolates. The defoliating and non-defoliating isolates were clearly differentiated into different VCGs (Table 4).

The higher frequency of Nit1 and NitM mutants than the Nit 3 mutants in this experiment has also been reported by several authors (Elena and Paplomatas, 1998; Korolov et al., 2000, Sanei et al., 2005). The comparison of selected Nit mutants produced only three VCGs. This shows that *V. dahliae* population is homogeneous and that the isolates are genetically closely related. The presence of low vegetative compatibility groups in *V. dahliae* has been reported by different authors (Joaquim and Rowe, 1991; Joaquim and Rowe, 1990). Even though the *V. dahliae* tested strains come from different geographical sites, they all belong to the same VCG suggesting the absence of relation between VCG and their geographic origin. This indicates that the studied population of *V. dahliae* isolates is homogenous and the strains are genetically related.

References

Abdi, N., Rahimian, H., Hamdollahzadeh, A., Ebrahimi, A.G., 1989. Verticillium wilt of plum .Proc. 9th Iran Plant Protec. Cong. p. 117.

Alizadeh, A., Agharafee, S., 1998. The causes of stone fruit trees decline in Tehran province. Proc. 13th Iran Plant. Cong. p. 232.

Arab–Salmani, M., Banihashemi, I., 2000. Vegetative compatibility groups among isolates of *Verticillium dahliae* from cotton and other sources in Iran. Proc. 14th Iran Plant Protec. Cong. p. 54.

Devax, A.L., Sackston, W.B., 1966. Taxonomy of *Verticillium* species causing wilt of horticultural crops in Quebec. Can. J. Bot. 44, 803–811.

Elena, K., Paplomatas, E.J., 1998. Vegetative compatibility groups within *Verticillium dahliae* isolates from different hosts in Greece. Plant Pathol. 47, 635-640.

Engelhard, A.W., 1957. Host index of *Verticillium albo–atrum* (including *Verticillium dahliae* kleb.). Plant Dis. ep. Suppl. 244, 23 – 49.

Ershad, D., 1974 . Fungi of Iran. Ministry Agriculture Agricultural and Natural Resources Research Organization, Tehran .

Ebrahimi, A.G., Sanei, S., 1995. Classification of isolates of *Verticillium dahliae* from Iran based on heterokaryon compatibility. Proc. 12th Iran Protec. Cong. p. 353.

Fasihiani, A., 1995. Reaction of some cotton cultivars to *Verticillium dahliae*. Proc. 12th Iranian Plant Protec. Cong. p. 114.

Himelick, E. B., 1969. Tree and shrub hosts of *Verticillium albo–atrum*. Biol. Nat. Illin. Nat. Hist. Surv. 66, 3–6.

Joaquim, T.R., Rowe, R.C., 1990. Reassessment of vegetative compatibility relationships among strains of *Verticillium dahliae* using nitrate- nonutilizing mutants. Phytopathology 80, 1160-1166.

Joaquim, T.R., Rowe, R.C., 1991. Vegetative compatibility and virulence of strains of *Verticillium dahliae* from soil and potato plants. Phytopathology 81, 552-558.

Korolev, N., Katan, J., Katan, T., 2000. Vegetative compatibility groups of *Verticillium dahliae* in Israel : Their distribution and association with pathogenecity . Phytopathology 90, 529 – 536.

Levin, A. G., Lavee, S., Tsror, L., 2003 .Epidemiology of *Verticillium dahliae* on Olive and its effect on yield under saline conditions. Plant Pathol. 52, 212–218 .

Ligoxigakis, E. K., 2000. Hosts of *Verticillium dahliae* in Kriti (Greece). Bulletin EPPO 30, 235–238.
Ligoxigakis, E.K., Vakalounakis, D.J., 1997. Hosts of *Verticillium dahliae* race 2 in Greece proc. Proc. 7th International *Verticillium* Symposium, p. 49.
Mamluk, O. F., Skaria, M., 1981.Verticillium wilt on vegetables in Jordan. FAO Plantec. Bull 27, 125–129.
Pegg, G.F., Bradly, G.F., 2002. Verticillium Wilts . CABI Pub.
Phillips, D.H., Burdekin, D.A., 1983. Disease of Forest and ornamental trees. Macmillan Press, London.
Razavi, S.I., Sanei, S.J., 1997. Vegetative compatibility of some isolates of *Verticillium dahliae* based on nit mutants . Proc. 1st Verticillium Symp. In Iran, p. 1.
Rudolph, B.A., 1931. Verticillium hardomycosis. Hillgardia 5, 201–361.
Sanei, S.J., Okhovvat, S.M., Javan-Nikkhah. M., Saremi, H. 2005. Vegatative compatibility and pathogenecity of *Verticillium dahliae* from Olive in Iran. Comm. Appl. Biol. Sci. (Ghent Univ.) 70, 323-326.
Sanei, S.J., Okhovvat, S.M., Hedjaroude, G.A., Saremi, H., Javan-Nikkhah, M., 2004. Olive Verticillium wilt or dieback of olive in Iran. Comm. Appl. Biol. Sci. (Ghent Univ.) 69, 433-442.
Sanei, S.J., Ebrahimi, A.G., Razavi, S.I., 1998. Hosts of *Verticillium dahliae* in Golestan and Mazandaran Provinces. Proc. 2nd Verticillium Symp. in Iran. p. 15.
Sanei, S.J, Razavi, S.I., Azad, G., 1998. Isolation of *Verticillium dahliae* from peach trees in Golestan Province. Proc. 2nd Verticillium Symp. in Iran. p. 17.
Sanei, S.J., Razavi, S.I., Zarei, H., Latifi, N., 1996. Verticillium wilt of Olive. Islamic Azad university, Gorgan Branch (in persian).
Sanei, S.J., Nassrollahnejad, S., 1995. Aggresiveness of *Verticillium dahliae* isolates to susceptible and resistant cultvans of cotton. J. Agric. Sci. 1, 67–77 (in Persian with English summary).
Schnatherst, W.C., 1971. Some biological aspect of *Verticillium albo–atrum* Cotton Disease Research Aspect of *Verticillium albo–atrum*. Cotton Division of Agriculture Science, University of California. Res. 1963–1969, 37–41.
Schnathorst, W.C., Mather, D.E., 1966. Host range and differentiation of a severe form of *Verticillium albo–atrum* in cotton. Phytopathology 56, 1155–1161.
Sherf, A.F., Macnab, A.A., 1986. Vegetable Diseases and thein control. Vol. B. 2nd ed. John Wiley and Sons, New York.
Slowson, D.D., 1987. Occurrence of *Verticillium dahliae* on various woods ornamental hosts. Plant Pathol. 36, 606–607.
Smith, H.C., 1965. The morphology of *Verticillium albo–atrum, V. dahliae, and V. tricorpus*. J. Agric. Res 8, 450–478.
Thanassoulopoulos, C.C., Biris, D.A., Tjamos, E.C., 1981. Weed as inoculum source of Verticillium in olive orchards. Phytopathol. Meditterr. 20, 164–168.
Tsror, L., Levin, E.G., 2003. Vegatative compatibility and pathogenecity of *Verticillium dahliae* Kleb. Isolation from Israel. J. Phytopathology 151, 451–455 .
Wolliams, G.E., 1966. Host range and symptomology of *Verticillium dahliae* in economic, weed and native plants in interior British Columbia. Can. J. Plant Sci., 46, 661–669.
Zakii, Z., Pourmansoori, T., 1995. Isolation of *Verticillium dahliae* from almond trees in Charmahal and Bokhtiari. Proc. 12th Iran Plant Protec. Cong. p. 132.