

**In The Name Of God**



**Shiraz University**

**Faculty of Agriculture**

**Ph.D Thesis in Plant Pathology**

**Evolutionary origin and phylogenetic relationships  
among  
*Fusarium oxysporum* f. sp. *melonis* isolates in Iran and  
their relationship with nonpathogenic isolates**

**BY**

**Maryam Mirtalebi**

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**May 2013**



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پایان نامه‌ی دوره دکتری رشته بیماری‌شناسی گیاهی

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*Fusarium oxysporum* f. sp. *melonis*

در ایران و ارتباط آن‌ها با جدایه‌های غیربیمارگر

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خرداد ۱۳۹۲

## **Declaration**

I - Maryam Mirtalebi- a student of Plant Pathology at faculty of Agriculture, hereby declare that the work presented in this thesis is the result of my own research. I have fully cited all materials and results that are not original to my work. Shiraz University holds the copyright on this research.

MARYAM MIRTALEBI

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RELATIONSHIP WITH NONPATHOGENIC ISOLATES**

**BY**

**MARYAM MIRTALEBI**

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**May 2013**

**Dedication To**

**My Beloved Husband**

**Ali**

**and**

**My Dear Daughter**

**Zahra**

**For**

**Their Love, Immense Patience and Understanding**

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branches of the same tree. All these aspirations are directed toward ennobling man's life, lifting it from the sphere of mere physical existence and leading the individual towards freedom". It has been significant challenge but a great pleasure to complete my thesis. I am thankful to all that have assisted in the process of "ennobling" my life through the process of my Ph. D.



## **Abstract**

### **Evolutionary origin and phylogenetic relationships among *Fusarium oxysporum* f. sp. *melonis* isolates in Iran and their relationship with nonpathogenic isolates**

**By**

**Maryam Mirtalebi**

*Fusarium* wilt of melon caused by *Fusarium oxysporum* f. sp. *melonis* is a destructive fungal disease in melon growing regions. Isolates of *F. oxysporum* obtained from five major melon producing provinces in Iran, from *Cucumis melo* L. cultivars and rhizosphere soil, were characterized based on pathogenicity on differential melon cultivars, vegetative compatibility groups (VCGs), nuclear ribosomal DNA intergenic spacer (IGS) region, translation elongation factor-1 $\alpha$  (TEF-1 $\alpha$ ) gene sequencing and rep-PCR analyses using Rep-primer. Ninety of 100 isolates from Iran in this study were identified as race 1,2 which most belonged to VCG 0134 and few to 'unassigned VCG', which based on IGS and TEF-1 $\alpha$  sequencing grouped with the VCG 0135 tester isolate. The remaining ten isolates were identified as nonpathogenic to melon belonging to two undescribed VCGs. Based on sequence analyses of the IGS region of Iranian and foreign isolates, nine lineages were identified, each including one VCG. The separation of VCGs into distinct lineages based on IGS sequences is mostly consistent with TEF-1 $\alpha$  sequencing and repetitive extragenic

palindromic PCR (Rep-PCR) results. Exceptions are VCGs 0130 and 0131, which could be differentiated with IGS sequences, but not with TEF-1 $\alpha$  and Rep-PCR. Different races from the USA, France and Iran associated with VCG 0134 grouped into one TEF-1 $\alpha$  and IGS lineage but could be differentiated with Rep-PCR into three closely related haplotypes, suggesting that this VCG is more diverse than previously thought. Given the long history of melon cultivation in Iran and the Rep-PCR diversity of isolates belonging to this VCG, it could be speculated that VCG 0134 perhaps evolved in Iran.

In this study, the evolutionary relationships between isolates of different *formae speciales* of *F. oxysporum* were also examined based on TEF-1 $\alpha$  sequences with a special emphasis on the *forma specialis melonis*. The tree inferred from bootstrapped maximum likelihood analysis of the dataset showed specific associations between *F. o. melonis* VCGs and the other *formae speciales* whose sequences were obtained from GenBank. Taken together, these results support a polyphyletic origin for *F. o. melonis*, meaning that the ability of this *forma specialis* to cause disease on melon has independently emerged multiple times.

**Key words:** Phylogenetic relationship, *Fusarium oxysporum* f. sp. *melonis*, Vegetative compatibility group, Race, Molecular markers.

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# **Chapter 1**

## **Introduction**