

# **STUDIES ON ALTERNARIA ROT IN CITRUS FRUITS AND ITS CONTROL**

By

**ABOU-GHANIMA SAAD FATH EL-BAB SHEHATA**

B.Sc. Agric. Sci. (Plant Pathology), Fac. Agric., Al-Azhar Univ., 2003

M.Sc. Agric. Sci. (Plant Pathology), Fac. Agric., Cairo Univ., 2010

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## **Approval Sheet**

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**This thesis for Ph.D. degree has been approved by:**

**Dr. Ahmed Zaky Aly**

.....

Prof. Emeritus of Plant Pathology, Faculty of Agriculture, Zagazig  
University

**Dr. Magdy Gad El-Rab Mohamed El-Samman**

.....

Prof. of Plant Pathology, Faculty of Agriculture, Ain Shams  
University

**Dr. Medhat Kamel Ali El-Sayed**

.....

Prof. Emeritus of Plant Pathology, Faculty of Agriculture, Ain Shams  
University

**Date of examination: / / 2018**

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**Under the supervision of:**

**Dr. Ahmed Ahmed Ahmed Mosa**

Prof. Emeritus of Plant Pathology, Dept. of Plant Pathology, Faculty  
of Agriculture, Ain Shams University (Principle supervisor)

**Dr. Medhat Kamel Ali El-Sayed**

Prof. Emeritus of Plant Pathology, Dept. of Plant Pathology Faculty  
of Agriculture, Ain Shams University

**Dr. Ahmed Korra Mohammed Korra (Late)**

Senior Research of Plant Pathology, Fruit and Wood Trees Disease  
Research Dep., Plant Pathol. Res. Inst., ARC, Giza

## ABSTRACT

**Abou-Ghanima Saad Fath El-Bab Shehata. Studies on Alternaria Rot in Citrus Fruits and its Control. Unpublished Ph.D. Thesis, Department of Plant Pathology, Faculty of Agriculture, Ain Shams University, 2017.**

In this study data indicated that, the disease survey in the nurseries and orchards of citrus during 2011 till 2012 revealed that, citrus fruit-rot disease was presented in all the observed Governorates, *i.e.* Beheira, Kalyobia, Menofia, Ismaelia Gharbia, Dakahlia, Giza and Beni Suief Governorates. Also, the highest percentage of disease survey was obtained in Beheira, followed by Kalyobia while the lowest percentage in Dakahlia Governorate.

Also, the results showed that, the highest percentage of isolated fungus was *Alternaria alternata* (Fries) Keissler followed by *A. citri* Ellis and Pierce. The tested isolates were significantly various in their responsibility to cause the disease.

Pathogenicity tests showed that, the tested isolates of *A. citri* can be arranged led to their pathogenic capabilities such as, *A. citri* isolates (code S24), (S4) and (S26) whilst, *A. citri* (S68) was the lowest pathogenic isolate. Whilst, *A. citri* isolate code (S71) not gave disease symptoms when Minneola detached leaves were inoculated with *A. citri*. On the other side, results of varietal reaction indicated that, the tested citrus cultivars cv., *i.e.* Minneola was more susceptible (indicator host), Navel orange was highly susceptible to infect by *A. citri* compared with minneola but, valencia was more tolerant to infect by tested pathogen.

Also, data of polymerase chain reaction (PCR) detection (molecular biology techniques) revealed that, 16 isolates of *A. citri* were identified by using 5 specific primers (ITS, OPA10-2, OPA2-1, OPA1-3

and Endo-PG) but, analysis technique showed that, the diversity between *A. citri* isolates based on two primers (OPA1-3 and Endo-PG) two both primers showed the diversity between tested *Alternaria* isolates.

Therefore results of *Alternaria* toxin bioassay revealed that, induce and produce necrotic lesions on *Minneola* citrus detached leaves similar to those produced by the pathogen. Moreover, the disease symptom was appeared after 48-72hr.

The effect of different solid cultures media was studied on the growth, sporulation and conidiospore measures of *A. citri* resulted that, potato dextrose agar (PDA) medium gave the maximum growth, the highest spore production (sporulation) and the best spore size with all tested isolates of *Alternaria*.

This study was extended to include the biological control of causal pathogen of citrus fruit-rot disease. Five bioagents were used, *i.e.* *Trichoderma harzianum*, *Bacillus subtilis*, *T. album* (Bio-Zeid), *B. megaterium* (Bio-ARC) and *Bacillus* spp. (Omega). All bioagents were the most antagonistic action effect against the causal pathogenic isolates of *A. citri* *in vitro* and *in vivo* trials but, with different degrees. Chemical control results indicated that, the seven tested fungicides namely, Score, Montoro, Iprodione, Pyraclostrobin, Coprax, Coprareekh and Azoxystrobin were evaluated for their effect on the linear growth of *A. citri* isolates (code K2) causing citrus fruit-rot disease *in vitro* trials. Also, the same fungicides were evaluated for their effect on disease incidence *in vivo* trials. Score, Montoro, Iprodione, were the most effective fungicides against causal pathogen.

**Key words:** Citrus, Fruit-rot, *Alternaria* toxin bioassay, Biological control, chemical control.

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## LIST OF ABBREVIATION

<b>A</b>	: Alternaria
<b>ABS</b>	: Alternaria brown spot
<b>ABR</b>	: Alternaria black rot
<b>ACT</b>	: <i>Alternaria citri</i> toxin
<b>Agric.</b>	: Agriculture
<b>a.i.</b>	: Active ingredient
<b>AKT</b>	: <i>Alternaria kikuchiana</i> toxin
<b>AMT</b>	: <i>Alternaria mali</i> toxin
<b>ANOVA</b>	: Analysis of variance
<b>CFU</b>	: Colony forming unit
<b>CLM</b>	: Czapek's liquid medium
<b>CV</b>	: Cultivar
<b>DNA</b>	: Deoxyribonucleic acid
<b>Endo-PG</b>	: Endo-polygalacturonase
<b>EC</b>	: Emulsifiable concentrate
<b>Fac.</b>	: Faculty
<b>Fig.</b>	: Figure
<b>f.sp.</b>	: Form specials
<b>G</b>	: Granules
<b>g</b>	: Gram(s)
<b>GFP</b>	: Green fluorescent protein
<b>HSTs</b>	: Host-selective toxins
<b>ITS</b>	: Internal transcript spacer
<b>KG</b>	: Kilogram
<b>L</b>	: Liter
<b>LSD</b>	: Least significant differences

## IX

<b>L.W.</b>	: Losses of weight
<b>M. Sc.</b>	: Master Science
<b>NA</b>	: Nutrient agar medium
<b>NB:</b>	: Nutrient broth medium
<b>PCR</b>	: Polymerase chain reaction
<b>Ph.D.</b>	: Doctor of philosophy
<b>PPM</b>	: Part per million
<b>PPO</b>	: Polyphenol oxidase
<b>pv</b>	: Pathovar
<b>SC</b>	: Soluble concentrate
<b>T</b>	: Trichoderma
<b>TBZ</b>	: Thiabendazole
<b>Univ.</b>	: University
<b>UV</b>	: Ultraviolet
<b>WA</b>	: Water agar
<b>WP</b>	: Watible powder