



شبكة المعلومات الجامعية
التوثيق الإلكتروني والميكرو فيلم

بسم الله الرحمن الرحيم



MONA MAGHRABY



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جامعة عين شمس

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MONA MAGHRABY

**STUDIES ON GENE REGULATION OF SOME
GENES THAT RELATED TO *BT*. TOXINS
RESISTANCE IN THE COTTON WORM,
*Spodoptera littoralis***

By

HAGER MOHSEN HASSAN ALI KHALIL
B.Sc. Agric. Sci. (Biotechnology), Fac. Agric., Cairo Univ., 2013

THESIS

Submitted in Partial Fulfillment of the Requirements for the Degree of

MASTER OF SCIENCE

In

**Agricultural Sciences
(Genetics)**

**Department of Genetics
Faculty of Agriculture
Cairo University
EGYPT**

2021

Format Reviewer

Vice Dean of Graduate Studies

SUPERVISION SHEET

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Title of Thesis: Studies on Gene Regulation of Some Genes that Related to
Bt. Toxins Resistance in the Cotton Worm, Spodoptera littoralis.
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Approval: 27/3/2021

ABSTRACT

A better understanding of the processing of *Bacillus thuringiensis* (*Bt*) Cry1C toxin in the midgut of *Spodoptera littoralis* larvae is very important to characterize the main regulatory elements of *Bt* tolerance. The current study aimed to evaluate the mRNA level of trypsin (Try), aminopeptidase N (APN), alkaline phosphatase (ALP), cadherin (Cad), and cytochrome P450 (CYP) in both susceptible and *Cry1C*- tolerant strains of *S. littoralis*. Total RNA was extracted from susceptible and tolerant strains to construct cDNA. Quantitative real-time PCR (qPCR) showed a significant upregulation of CYP gene in tolerant strain. In contrast, the levels of expression of Try, ALP and Cad were significantly downregulated in tolerant strain. APN relative mRNA expression did not show significant differences between susceptible and tolerant strains. Histologically, the midgut of late third-instar larvae of tolerant population *S. littoralis* showed vacuolization of the epithelium and disruption of both the peritrophic membrane and the striated boarder compared to the susceptible strain. Our data indicate the important roles of CYP, Try, ALP and Cad in the resistance development and toxicity to *Bt* Cry1C. The obtained results are useful for further illustrating of *Bt* Cry1C processing and *S. littoralis* tolerance.

Key words: *Spodoptera littoralis*, *Bacillus thuringiensis*, *Bt* Cry1C toxin, Tolerance, qPCR

DEDICATION

I dedicate this work to whom my heartfelt thanks: my parents and brother for all the support they lovely offered during my graduate studies.

ACKNOWLEDGEMENT

*First of all, I would like to thank **ALLAH** almighty, the most merciful and compassionate, for his support, help and generosity.*

*My deepest thanks for **Dr. Haggag Salah Zein**, Professor of Genetics, for his appreciated, continued guidance and help in practical this thesis.*

*I wish to express my sincere thanks, deepest gratitude and appreciation to **Dr. Nagwa Ibrahim Abdelfattah Elarabi**, Associated Professor of Genetics, Faculty of Agriculture, Cairo University, for supervision, continued assistance, and guidance through the course of my study and for revision of the manuscript of this thesis.*

*Sincere thanks and deepest gratitude are also due to **Dr. Saad Moussa**, Head Researcher of Insect Biotechnology, Plant Protection Research Institute, Agricultural Research Center, for sharing in supervision, guiding me and facilitating difficulties.*

*Special thanks and sincere appreciation to **Dr. Dalia Sayed Ahmed Hasaneen**, lectural of Genetics, Faculty of Agriculture, Cairo University, for supervision, continued assistance, and guidance through the course of my study and for revision of the manuscript of this thesis.*

Grateful appreciation is also extended to all staff members of Genetics department, Faculty of Agriculture, Cairo University and all staff members of Insect Biotechnology and Molecular Biology unit, Plant Protection Research Institute, Agricultural Research Center.

Special deep appreciation is given to my father, my mother, my brother and my friends for their support and encouragement.

LIST OF ABBREVIATIONS

ABC	: ATP-Binding Cassette
AGERI	: Agricultural Genetic Engineering Research Institute
ALP	: Alkaline phosphatase
APN	: Aminopeptidases N
APS	: Ammonium persulfate
ARC	: Agricultural Research Center
BBMVs	: Brush Border Membrane Vesicles
BLAST	: Basic Local Alignment Search Tool
bp	: Basepair
BSA	: Bovine Serum Albumin
Bt	: <i>Bacillus thuringiensis</i>
Bti	: <i>B. thuringiensis subsp. israelensis</i>
Cad	: Cadherin
cAMP	: Cyclic Adenosine MonoPhosphate
cDNA	: Complementary DNA
Cry	: Crystal
Cry1AMod	: Cry Modified toxins
Ct	: Threshold Cycle
CTP	: Chymotrypsin
CYP	: Cytochrome P450
Cyt	: Cytolytic
DEPC	: Diethyl Pyrocarbonate
2D-DIGE	: Two-Dimensional Differential In-Gel Electrophoresis
DNA	: Deoxyribonucleic acid
dsRNA	: Double-stranded RNA
DTT	: Dithiothreitol
EDTA	: Ethylenediaminetetraacetic acid
EtBr	: Ethidium Bromide
FL	: Fiducial Limits
GST	: Glutathione S-Transferases
ICPs	: Insecticidal Crystal Proteins
Kb	: Kilo basepair
KDa	: Kilodalton
LC₅₀	: 50% Lethal Concentration
mALP	: Membrane-bound Alkaline phosphatase

