

**SOME BIOCHEMICAL CHANGES IN THE LARVA  
OF THE PINK BOLLWORM PECTINOPHORA  
GOSYPIELLA SAUNDERS IN DIAPAUSE AND  
DURING TERMINATION OF DIAPAUSE**

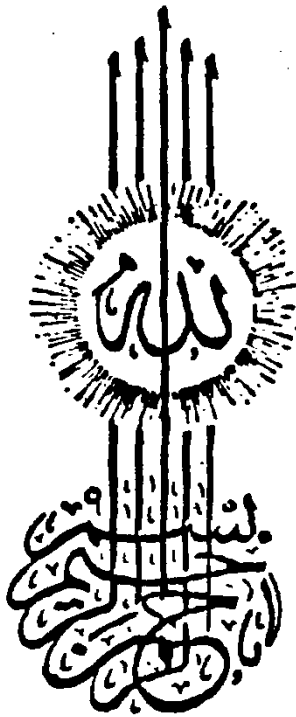
**A THESIS**

Presented to the Faculty of Science  
Ain Shams University  
For the Award of the  
M. Sc. Degree

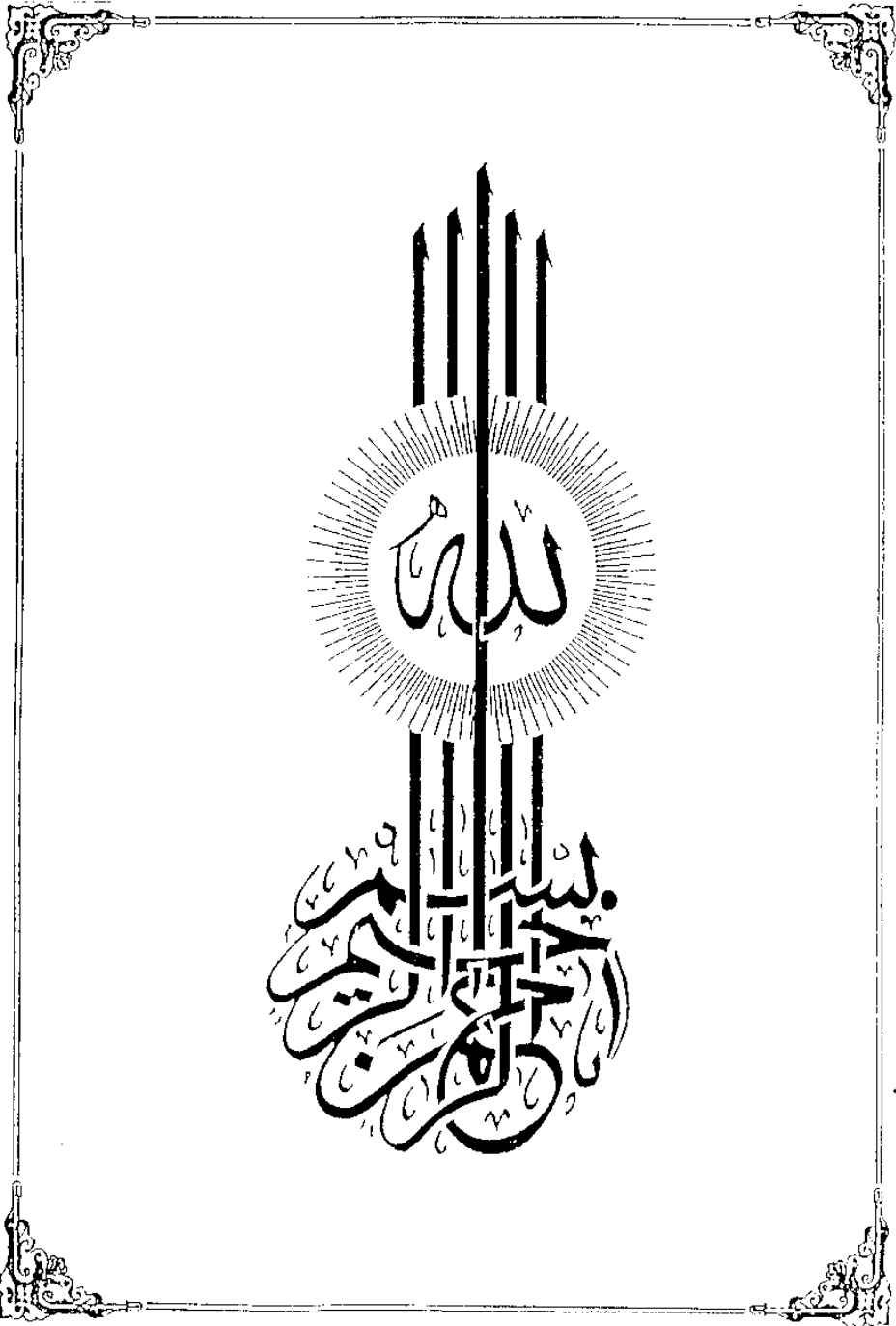
By  
**MOHAMED SAYED SALAMA**  
( B. Sc. )

Department of Entomology  
Faculty of Science  
Ain Shams University  
Cairo

**1985**



وَقُلْ اعْمَلُوا فَسَيَرَى اللهُ عَمَلَكُمْ وَرَسُولُهُ وَالْمُؤْمِنُونَ  
وَصَدَقَ اللهُ أَكْبَرًا



3

THESIS EXAMINATION COMMITTEE

NAME	TITLE	SIGNATURE
.....	.....	.....
.....	.....	.....
.....	.....	.....

## ACKNOWLEDGMENT


The author wishes to express his gratitude to Dr.Z.M.F. Rostom, Professor of Entomology, Faculty of Science , Ain Shams University, for suggesting the problem, supervising the work, criticizing the results and reading the manuscript.

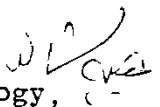
The author is also indebted with thanks to Dr.A.M. El-Shafei, Lecturer of Entomology, Faculty of Science, Ain Shams University, for her help in guiding the work and invaluable facilities and encouragement.

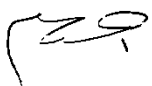
Sincerest gratitude is due to Dr,A.M. Salem, Lecturer of Biochemistry, Faculty of Science Ain Shams University, for his direct guidance of the work and reading the manuscript.

Thanks are also due to Dr.A.A. Salem, Director of the Scientific Computing Center, Ain Shams University, Cairo and to members of this Center who offered the facilities for analysis of the results statistically.

Supervisors

Prof. Dr. Zaky Mohammed Fathy Rostom, Professor of  
Entomology, Faculty of Science, Ain Shams  
University. 

Dr. Akila Mohammed El Shafei, Lecturer of Entomology,  
Faculty of Science, Ain Shams University. 

Dr. Ahmed Mohammed Salem, Lecturer of Biochemistry,  
Faculty of Science, Ain Shams University. 

## BIOGRAPHY

Date and place of birth : 1 October 1955, Kena.

Date of graduation : June 1979.

Degree awarded : B.Sc. (Entomology).

Grade : Excellent.

Occupation : Demonstrator in Entomology Department  
Faculty of Science, Ain Shams University.

Date of appointment: November, 1979.

Date of Registration: 14 June 1982.

for the M.Sc. Degree.

Courses Studied by the Candidate in  
Partial Fulfilments for Degree of  
Master of Science

Language : Germany: M.Sc. Course.

Examination passed on March, 1982.

Entomology Courses :

1. Environmental Pollution.
2. New Approached to Insect Control.
3. Insect Problems in Egypt.
4. Microbial Control.
5. Population Dynamics.
6. Insect Taxonomy.
7. Chemistry of Pesticides.
8. Radiobiology and applied Entomology.
9. Insect Hormones and Pheromones.
10. Research subject.

Examination passed on Feb. 1982.

Statistical Course: Biostatistics

Examination passed on February 1982.



## ABBREVIATIONS.

In the present work the following abbreviations were used:

A = active phase of larvae of Pectinophora gossypiella

Bis = N-N' methylene bis acrylamide.

COBB = coomassie brilliant blue stain.

D<sub>1</sub> = early diapause phase of Pectinophora gossypiella larvae 40 days old.

D<sub>2</sub> = late diapause phase of Pectinophora gossypiella larvae 80 days old.

Dist. = distilled.

FAA = free amino acids.

FB = fat body.

H = haemolymph.

JH = juvenile hormone.

MW = molecular weight.

PAS = periodic acid Schiff's stain.

R<sub>f</sub> = relative mobility.

SBB = Sudan black B stain.

SDS = sodium dodecyl sulphate.

T<sub>2</sub> = Pectinophora gossypiella larvae at the second day during diapause termination at 34°C.

T<sub>4</sub> = Pectinophora gossypiella larvae at the fourth day during diapause termination at 34°C.

T<sub>6</sub> = Pectinophora gossypiella larvae at the sixth day during diapause termination at 34°C.

T<sub>8</sub> = Pectinophora gossypiella larvae at the eighth day during diapause termination at 34°C.

T<sub>10</sub> = Pectinophora gossypiella larvae at the tenth day during diapause termination at 34°C.

T<sub>12</sub> = Pectinophora gossypiella larvae at the twelveth day during diapause termination at 34°C.

T<sub>14</sub> = Pectinophora gossypiella larvae at the fourteenth day during diapause termination at 34°C.

TCA = trichloroacetic acid.

TEMED = N,N,N',N'-tetramethylene diamine.

TRIS = Tris (hydroxymethyl) amino methane.

## CONTENTS

	Page
I. Introduction .....	1
II. Literature review .....	3
III. Materials and methods .....	32
III.1. Sampling of the different phases of larvae .....	32
III.2. Collection of samples of the haemolymph and fat body .....	33
III.3. Biochemical analyses .....	34
III.3.1. Total protein .....	34
III.3.2. Separation of protein bands by electrophoresis .....	36
III.3.3. Refractionation of diapause protein bands by SDS electrophoresis.....	44
III.3.4. Determination of molecular weights of proteins.....	49
III.3.5. Calculation of $R_f$ of protein bands..	49
III.3.6. Free amino acid determination .....	50
III.3.7. Inorganic fractions.....	51
III.3.7.1. Estimation of sodium.....	51
III.3.7.2. Estimation of potassium....	52
III.3.7.3. Estimation of calcium and magnesium .....	54
III.4. Analysis of results .....	58

	Page
IV. Results .....	60
IV.1. Total protein .....	60
IV.2. Proteins, glycoproteins and lipoproteins.....	66
IV.3. Diapause proteins .....	73
IV.4. Free amino acids .....	83
IV.5. Inorganic ions .....	95
V. Discussion and conclusion .....	101
V.1. Total protein .....	101
V.2. Proteins, glycoproteins and lipoproteins.....	107
V.3. Free amino acids. ....	118
V.4. Inorganic ions.....	126
VI. Summary .....	132
VII. Literature Cited .....	137
Arabic summary .	

# ***I-INTRODUCTION***

## I. INTRODUCTION

The pink bollworm Pectinophora gossypiella Saunders, is one of the most destructive world wide distributed pests. It has been introduced into Egypt between 1903-1910 from India (Willcocks, 1916). In 1913 this pest spread to such a degree that it became a real menace to the cotton crop in Egypt. This pest is characterized by a facultative diapause in the last fourth instar larva that is passed usually inside double seeds of cotton. Such diapause larvae represent a very serious source of infestation to the new cotton crop.

The biochemical changes that take place during induction, maintenance and termination of diapause of the pink boll worm have not yet been satisfactorily studied. A promising approach to the understanding of the problem of diapause may be achieved from studying changes of biochemical compounds like simple and conjugated proteins, amino acids, inorganic ions ....etc. Knowledge of these chemical fractions are fundamental in determining metabolic weak points through which the sequence of the physiological events leading to induction, maintenance and termination of diapause may be upset. This might inhibit, minimize or terminate diapause which is a final glorious target.

Most of the available information on changes of chemical constituents during periods of arrested development was obtained from extracts of whole body. Few data, however, are available on either separate haemolymph or fat body fraction.

#### Aim of the present work

In the present work the haemolymph and fat body of the fourth instar larvae of P. gossypiella were analysed for estimating total proteins, glycoproteins, lipoproteins, free amino acids and the inorganic elements  $\text{Na}^+$ ,  $\text{K}^+$ ,  $\text{Ca}^{++}$  and  $\text{Mg}^{++}$ . Larvae used represented the mature active non-diapause phase, an early and late age of the diapause phase and at seven intervals during termination of diapause. Results on these 10 phases of the larvae were compared together in an attempt to clarify the characteristic changes of the above mentioned biochemical compounds during induction, maintenance and termination of diapause.