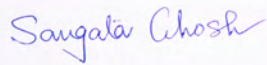


*Dedicated to
all my respected teachers*

DECLARATION

I declare that the thesis entitled 'A STUDY ON THE DEFENSE SYSTEM OF THE TEA PEST *Hyposidra talaca* (WALKER, 1860) (LEPIDOPTERA: GEOMETRIDAE) AGAINST CHEMICAL PESTICIDES AND ENTOMOPATHOGENS' has been prepared by me under the supervision of Professor (Dr.) Min Bahadur, Department of Zoology, University of North Bengal. No part of this thesis has formed the basis for the award of any degree or fellowship previously.



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CERTIFICATE

This is to certify that Mr. Saugata Ghosh has prepared the thesis entitled '**A STUDY ON THE DEFENSE SYSTEM OF THE TEA PEST *Hyposidra talaca* (Walker, 1860) (Lepidoptera: Geometridae) AGAINST CHEMICAL PESTICIDES AND ENTOMOPATHOGENS**' for the award of Ph.D. degree of the University of North Bengal, under my guidance. He has carried out the work at the Department of Zoology, University of North Bengal. The content of the thesis has not been submitted elsewhere for any degree.

Supervisor

Prof. (Dr.) Min Bahadur





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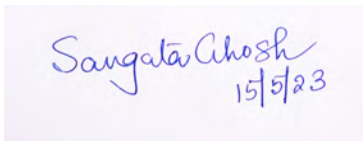
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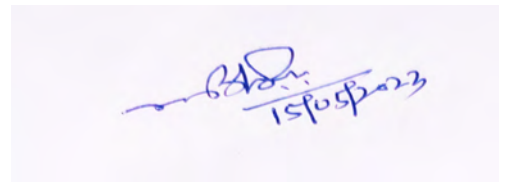
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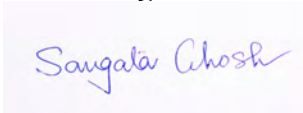
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Preface

Insects, the most successful group of animals colonize almost all ecological niches and are responsible for massive crop-damage leading to huge loss in world economy and human livelihood. To manage the variety of insect pests, an array of chemical pesticides has long been used worldwide. To cope with these chemicals, as well as naturally occurring entomopathogens, insects have evolved an effective defense system. Indiscriminate use of the synthetic pesticides facilitates the development of higher tolerance or resistance against these chemicals in many insect pests leading to frequent failure in the pest control strategies. Pesticide resistance is a most serious problem throughout the world including India. In this situation, the in-depth knowledge about the defense strategy of various insect pests makes it possible to develop a target specific control management against the pests in a sustainable way.

The lepidopteran geometrid looper pest, *Hyposidra talaca* causes immense defoliation in the tea plantations of Darjeeling Terai region resulting in huge economic loss and so to manage this major tea pest, several chemical pesticides are regularly being used indiscriminately leading to development of the resistance of the pests against the pesticides as well as causing environmental and health problems. Therefore, the present study has been planned to investigate the cellular and humoral components of the defense system and the defense enzymes of *Hyposidra talaca* in the Darjeeling Terai region of West Bengal, India to assess the effects of the selected chemical pesticides and entomopathogens on *H. talaca*. This is a pioneer study which has focused on some aspects of the defense system of this geometrid tea pest for the first time. In this context, the knowledge of defense system of this pest along with the immune response against the regularly used chemical pesticides will help the plant growers for selective spraying of the pesticide instead of 'no threshold' spraying throughout the calendar year. Moreover, the information on the effect of the potential entomopathogens on the host's defense system and the subsequent immune response will help in the development of efficient bio-pesticides using those entomopathogens.

The **first part** of this thesis consists of an **introduction** of the Indian tea in global perspective, *Hyposidra talaca* as one of the major tea pests in Darjeeling Terai region of northern part of West Bengal, management strategy against *H. talaca* and the

defense system of insects in general. The **second part** is focused on the **review of literature**, from the multidimensional research areas on the insect defense system and immune responses to chemical pesticides and entomopathogens and different aspects of *H. talaca* and related species including, taxonomy, ecology, pest status, control strategies. The **third and fourth sections** contain the major **objectives** of this research work and detailed **materials and methods**, respectively for fulfilling the objectives of this study. An elaborate **results and discussions** have been included in the **fifth section** comprising development of *Hyposidra talaca*, haemocyte count in respect to diet regimes, chemical pesticides (emamectin benzoate and cypermethrin) and entomopathogens (*Bacillus thuringiensis* and nucleopolyhedrovirus) at different developmental stages (ages), detoxifying enzyme assays for general esterase, cytochrome P450 and glutathione S-transferase and electrophoretic analysis of haemolymph proteins/peptides. Finally, a **conclusion** has been added in the thesis mentioning future prospects of this study and a **summary** for quick understanding. An elaborate **bibliography** has been included with all the references cited in the text. There is also an **appendix** consisting of index, publications and list of seminars/symposia attended and presented the papers.

The outcome of this dissertation is expected to be helpful to the tea growers in optimizing the pest management strategy to control *Hyposidra talaca*, the major tea pest in the Darjeeling Terai plantations in the sub-Himalayan region of West Bengal, India without any adverse effect to the non-target populations and environment and opens up a multidisciplinary arena for future research.



SAUGATA GHOSH

15th May, 2023

ABBREVIATION

°C	Degree Centigrade	DDT	Dichloro-diphenyl-trichloroethane
µl	Micro litre	DHC	Differential haemocyte count
µmol	Micro mole	DPX	Distyrene, plasticizer and xylene
20-E	20-hydroxyecdysone	DOPA	3,4-dihydroxyphenyl alanine
AChE	Acetylcholine Esterase	DOPAC	3,4-dihydroxyphenyl acetic acid
ACP	Acid phosphatase	EB	Emamectin benzoate
AD	Artificial diet	EC	Emulsifiable Concentrate
AH	Adipohaemocyte	EcobNPV	<i>Ectropis obliqua</i> nucleopolyhedro virus
ALP	Alkaline phosphatase	F	F-distribution value used in ANOVA
AMP	Anti-microbial peptide	FBs	Fibrillar Bodies
ANOVA	Analysis of variance	GABA	γ-Aminobutyric Acid
ApoIII	Apolipoprotein III	GE	General Esterase
ApoLp	Apolipoprotein	GI	Geographical indicator
BOD	Biological oxygen demand	GluCl	Glutamate-gated chloride channel
BSA	Bovine serum albumin	GR	Granulocyte
Bt	<i>Bacillus thuringiensis</i>	GSH	Reduced glutathione
Btk	<i>Bacillus thuringiensis</i> var. <i>kurstaki</i>	GST	Glutathione S-Transferase
BV	Budded Virion	H	Test statistic for the Kruskal-Wallis test
C	Control	hrs	Hours
CDNB	1-chloro-2, 4-dinitrobenzene	HytaNPV	<i>Hyposidra talaca</i> nucleopolyhedrovirus
CO	Coagulocyte	HzNPV	<i>Heliothis zea</i> single capsid nucleopolyhedrovirus
COVID 19	Corona Virus Disease 2019	IRAC	Insecticide Resistance Action Committee
CP	Cypermethrin	JH	Juvenile hormone
Cry	Crystal	kDa	Kilo Dalton
CYP	Cytochrome P450	Kg	Kilogram
Cyt	Cytolytic		
D	Day		

L	Litre	PAGE	Polyacrylamide gel electrophoresis
LC₅₀	Mid-lethal concentration	PAMP	Pathogen-associated molecular pattern
L:D	Light: Dark	PBS	Phosphate buffered saline
Lp	Lipophorin	PC	Podocyte
LPS	Lipopolysaccharide	PL	Plasmatocyte
LTA	Lipoteichoic acid	PO	Phenoloxidase
max.	Maximum	PPO	Pro-phenoloxidase
min	Minute	PR	Prohaemocyte
ml	Mili litre	RH	Relative humidity
mM	Mili mole	rpm	Rotation per minute
MNPV	Multiple Nucleopolyhedrovirus	RNA	Ribonucleic acid
MOA	Mode of action	SD	Standard deviation
MW	Molecular weight	SDS	Sodium dodecyl sulphate
MW U	Value used in Mann-Whitney U statistics	SG	Water soluble granules
n	Number of individuals	SP	Storage protein
NADA	N-acetyl dopamine	SpliMNPV	<i>Spodoptera littoralis</i> multiple nucleopolyhedrovirus
NBAD	N-β-alanyl dopamine	spp.	Species in plural
NBU	North Bengal University	t	Value used in a t-test to determine whether to support or reject the null hypothesis
ND	Natural diet	THC	Total haemocyte count
nm	Nano meter	TMBZ	3,3', 5, 5' Tetra methyl benzidine
nmol	Nano mole	TRA	Tea research association
NPV	Nucleopolyhedrovirus	TV	Tocklai variant
OB	Occlusion body	UV/VIS	Ultra violet/ visible
ODV	Occlusion-derived virion	var.	varietas (Latin)
OE	Oenocytoid	VE	Vermicyte
OP	Organophosphate	VS	Virogenic Stroma
p	Probability that the null hypothesis is true		

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