

CONTENTS

Title	Page No.
Abbreviations	
Abstract	i
Chapter – 1. General Introduction	1
1.1. Agriculture in India	1
1.2. Cotton	1
1.2.1. Cotton pests	2
1.2.1.1. <i>Dysdercus cingulatus</i> (Fab.) (Hemiptera: Pyrrhocoridae)	3
1.2.1.2. <i>Spodoptera litura</i> (Fabricius) (Lepidoptera: Noctuidae)	4
1.2.1.3. <i>Aphis craccivora</i> Koch (Homoptera: Aphididae)	4
1.2.1.4. <i>Helicoverpa armigera</i> (Hubner) (Lepidoptera: Noctuidae)	5
1.2.1.5. <i>Mylabris pustulata</i> (Thunb.) (Coleoptera: Meloidae)	5
1.2.1.6. <i>Mylabris indica</i> (Faust) (Coleoptera: Meloidae)	5
1.2.1.7. <i>Pericallia ricini</i> Fab. (Lepidoptera: Arctiidae)	5
1.2.1.8. <i>Oxycarenus hyalinipennis</i> (Costa) (Hemiptera: Lygaeidae)	6
1.3. Non-IPM	6
1.4. IPM	6
1.4.1. Entomopathogens in Biological control programme	7
1.4.1.1. Bacteria and virus	8
1.4.1.2. Fungi	9
1.4.1.2.1. <i>Metarhizium anisopliae</i> (Metsch.) Sorokin	10
1.5. Nature of infection	11
1.6. Low cost mass multiplication	12
1.7. DNA Polymorphism necessity	13
1.8. Objectives of the study	14
Chapter – 2. Distribution of <i>Metarhizium anisopliae</i> in Tamil Nadu	15
2.1. Introduction	15
2.1.1. Tamil Nadu	15
2.1.2. Soil type in Tamil Nadu	15
2.1.3. Soil as a habitat of <i>Metarhizium</i>	16

2.1.4. Taxonomy of <i>Metarhizium</i>	16
2.1.5. Growth factors	17
2.2. Materials and methods	17
2.2.1. Study area	17
2.2.2. Collection of samples	17
2.2.3. Preparation of potato dextrose agar media (PDA)	18
2.2.4. Isolation and Enumeration of TFP of soil and insects	18
2.2.5. Subculture and identification of <i>M. anisopliae</i>	19
2.2.6. Storage of <i>M. anisopliae</i> isolates	19
2.3. Results	20
2.3.1. Distribution of <i>Metarhizium anisopliae</i> isolates in soil	20
2.3.2. Distribution in insects	21
2.4. Discussion	22
2.5. Conclusion	25
Chapter – 3: Biological control potential	26
3.1. Introduction	26
3.2. Materials and Methods	31
3.2.1. Sources of pests	31
3.2.2. Cultivation of the isolates and preparation of conidial suspension	32
3.2.3. Laboratory bioassay on <i>D. cingulatus</i> eggs	33
3.2.3.1. Soil preparation	33
3.2.3.2. Bioassay	33
3.2.4. Bioefficacy of selected isolates on <i>D. cingulatus</i> instars	34
3.2.5. Bioassay on <i>D. cingulatus</i> adults	35
3.2.5.1. Contact toxicity	35
3.2.5.2. Direct plate assay	35
3.2.5.3. Sapling bioassay	35
3.2.6. Bioefficacy of selected isolates on 6 cotton pests	36
3.2.7. Statistical analysis	37
3.3. Results	37

3.3.1. Laboratory bioassay on <i>D. cingulatus</i> eggs	37
3.3.2. Bioefficacy of selected isolates on <i>D. cingulatus</i> instars	38
3.3.3. <i>D. cingulatus</i> adults	38
3.3.3.1. Contact bioassay	38
3.3.3.2. Direct plate assay	39
3.3.3.3. Sapling bioassay	40
3.3.4. Bioefficacy of selected isolates on 6 pests	41
3.4. Discussion	41
3.4.1. Bioassay on <i>D. cingulatus</i> eggs	42
3.4.2. Bioefficacy of selected isolates on <i>D. cingulatus</i> instars	44
3.4.3. Contact bioassay on <i>D. cingulatus</i> adults	44
3.4.4. Direct plate assay	47
3.4.5. Sapling Bioassay	49
3.4.6. Bioefficacy of selected isolates on 6 cotton pests	50
3.5. Conclusion	53
Chapter – 4: Route of infection and Haematology	54
4.1. Introduction	54
4.2. Materials and Methods	56
4.2.1. Re-isolation of <i>Metarhizium anisopliae</i>	56
4.2.2. Haemosomic index	56
4.2.3. Total haemocyte count	57
4.2.4. Enumeration of <i>M. anisopliae</i> in haemolymph	58
4.2.5. Cuticle mounting	58
4.2.6. Statistical analysis	59
4.3. Results	59
4.3.1. Re-isolation	59
4.3.2. Haemosomic index	59
4.3.3. THC	60
4.3.4. Enumeration of <i>M. anisopliae</i> in haemolymph	60
4.3.5. Cuticle mounting	61
4.4. Discussion	61

4.5. Conclusion	66
Chapter – 5: Screening of low cost media for mass production of <i>Metarhizium anisopliae</i>	67
5.1. Introduction	67
5.2. Materials and Methods	71
5.2.1. Fungal cultures	71
5.2.2. Conidial inoculum preparation	71
5.2.3. Culture media	72
5.2.3.1. Synthetic and non-synthetic liquid household waste media	72
5.2.3.2. Dry fish waste water	72
5.2.3. Inoculation	72
5.2.4. Growth parameters	73
5.2.5. Colonial radial growth of isolates for three successive generations	74
5.2.6. Statistical analysis	74
5.3. Results	74
5.3.1. Spore production in PDB	74
5.3.2. Media consumption and biomass production	75
5.3.3. Non-synthetic liquid media	75
5.3.3.1. Rice wash water and rice boiled water	75
5.3.3.2. Coconut water	76
5.3.3.3. Dry fish wastewater	77
5.3.4. Colonial generation growth	77
5.3.5. Correlation analysis	79
5.4. Discussion	79
5.5. Conclusion	83
Chapter - 6: Genetic diversity among <i>Metarhizium anisopliae</i> isolates from Tamil Nadu	84
6.1. Introduction	84
6.2. Materials and Methods	87
6.2.1. Fungal strains	87
6.2.2. Mycelial preparation	87

6.2.3. Genomic DNA extraction from <i>M. anisopliae</i>	87
6.2.3.1. Quantity and quality of DNA	89
6.2.4. RAPD analysis	90
6.2.4.1. Random primers used	90
6.2.4.2. PCR conditions	90
6.2.4.2.1. Amplification reactions mixture of the RAPD	90
6.2.4.2.2. Temperature profile	91
6.2.3. Statistical analysis	91
6.3. Results	92
6.4. Discussion	95
6.5. Conclusion	98
7. Summary	100
8. Future area of research	102
9. References	103
10. List of publications	