

# Index

1. General introduction .....	1
2. Origin and distribution .....	3
3. Taxonomic framework and synonymous terms.....	6
Identification .....	7
4. Biology and life cycle .....	8
5. <i>Tecia solanivora</i> developement.....	10
6. Damage.....	13
7. Monitoring and decision-making .....	15
8. Control measures.....	16
8.1. Farming control measures.....	16
8.2 Biotechnical control measures .....	17
8.2.1 Pheromones .....	17
8.2.2 Plant extracts .....	19
8.2.3 Varietal resistance .....	20
8.2.4 Attract and kill .....	20
8.2.5 Sunlight treatment.....	21
8.2.6 Modified atmosphere .....	21
8.3 Biological control measures.....	22
8.3.1 Entomopathogenic organisms.....	23
▪ <i>Bacillus thuringensis</i> .....	23
▪ Entomopathogenic fungi .....	24
▪ Baculovirus .....	24
8.3.2 Parasitoids and predators .....	28
▪ <i>Copidosoma koehleri</i> .....	28
▪ <i>Trichogramma</i> spp.....	30
▪ Ichneumonidae.....	32
▪ Predatory mites .....	33
9. Chemical control.....	34
Objectives .....	35
<u>Chapter I</u> .....	36
Abstract .....	37
Resum.....	37
Resumen.....	38
1. Introduction .....	38

<b>2. Materials and Methods</b> .....	40
2.1. Insects Rearing .....	40
2.2. Molecular Identification .....	40
2.3. Host Acceptance (No-Choice).....	41
2.4. Host Preference (Choice).....	42
2.5. Functional Response and Parasitism .....	42
2.5. Illumination Assays.....	43
2.7. Search Ability under Semi-Field Conditions .....	44
<b>3. Results</b> .....	45
3.1. Host Acceptance (No-Choice).....	45
3.2. Host Preference (Choice).....	45
3.3. Functional Response .....	46
3.3.1. <i>Trichogramma euproctidis</i> .....	46
3.3.2. <i>Trichogramma achaeae</i> .....	47
3.4. Parasitism .....	49
3.5. Sex Ratio and Fecundity .....	49
3.6. Illumination Acceptance Assays .....	51
3.7. Search Ability under Semi-Field Conditions .....	51
<b>4. Discussion</b> .....	52
<b>5. Conclusions</b> .....	55
<b>Chapter II</b> .....	56
<b>Chapter II A</b> .....	56
<b>Abstract</b> .....	56
<b>Resum</b> .....	56
<b>Resumen</b> .....	58
<b>1. Introduction</b> .....	58
<b>2. Materials and Methods</b> .....	60
2.1. Biological Materials and Experimental Conditions.....	60
2.2. Prey Acceptance Test and Evaluation of the Predatory Potential at Different Temperatures .....	60
2.3. Microcosm .....	61
2.4. Host Choice: <i>Tecia solanivora</i> (Tcs)- <i>Phthorimaea Operculella</i> (PTH) .....	62
2.5. Functional Response: Predatory Behaviour at Different Prey Densities .....	62

<b>3. Results</b> .....	64
3.1. Evaluation of the Predatory Potential at Different Temperatures.....	64
3.2. Microcosm.....	65
3.3. Host Choice.....	66
3.4. Functional Response: Predatory Behaviour at Different Prey Densities.....	66
<b>4. Discussion</b> .....	68
<b>5. Conclusions</b> .....	70
<b>Chapter II B.</b> .....	71
<b>Abstract</b> .....	71
<b>Resum</b> .....	71
<b>Resumen</b> .....	72
<b>1. Introduction</b> .....	72
<b>2. Materials and Methods</b> .....	74
2.1. <i>Insect Rearing</i> .....	74
2.2. <i>Microcosm 15 °C</i> .....	74
2.3. <i>Semi-Storage</i> .....	76
2.3.1. Experimental Procedure.....	76
2.3.2. Release Preparation of Natural Enemies.....	77
2.3.3. Statistical Analysis.....	77
2.4. <i>Storage</i> .....	78
<b>3. Results</b> .....	79
3.1. <i>Microcosm 15 °C</i> .....	79
3.2. <i>Semi-Storage</i> .....	80
3.2.1. Survival of <i>T. solanivora</i> .....	80
3.2.2. Tuber Damage.....	80
3.3. <i>Storage</i> .....	81
<b>4. Discussion</b> .....	81
<b>5. Conclusions</b> .....	83
<b>General discussion</b> .....	84
<b>General conclusions</b> .....	88
<b>Bibliography</b> .....	89
<b>Legislation</b> .....	104

# Figures index

## General introduction

- Figure 1.** Percentage of cultivated potato area (white numbers) and production (blue numbers) per island respect to the whole Canary Islands. Source: own work, based on ISTAC (2024) data. 1
- Figure 2.** Origin and spreading of *T. solanivora*. The area of origin of the Guatemalan potato moth was limited to Guatemala and the Isthmus of Tehuantepec. The arrows indicate where the pest spread and the year it was reported. .... 3
- Figure 3.** Distribution of *T. solanivora* in the Canary Islands..... 4
- Figure 4.** Distribution of *T. solanivora* in Galicia according to official declarations in the DiarioGalicia (DOG). Source: author's own work. DiarioGalicia (DOG).....5
- Figure 5.** Distribution of *T. solanivora* in the Iberian Peninsula. Red indicates areas declared as infested, orange indicates buffer zones, and blue indicates free areas subject to the declaration of planted crops.....5
- Figure 6.** Chaetotaxy for the identification of *T. solanivora* larvae. Source: Germain and Povolný 2006..... 7
- Figure 7.** Genitalia of *T. solanivora*. Male genitalia (left): V = valva; U = uncus; G =gnathos; S = saccus. Female genitalia (right): Aa = anterior apophysis; Ap = posterior apophysis; A = antrum; Si = signum..... 8
- Figure 8.** Diagram of the life cycle of *T. solanivora*. The minimum values for the duration of each stage are those corresponding to 25 °C and the maximum values are those corresponding to 15 °C. .... 9
- Figure 9.** Couple of *T. solanivora* adults copulating.....9
- Figure 10.** . A) Freshly laid 24-hour eggs. B) Egg about to hatch, the arrow indicates the darkened cephalic case of the larva visible through the chorion. C) Larva in the process of emergence after breaking the chorion with the mandible ..... 10
- Figure 11.** Differences in color between larvae of *T. solanivora* (A and C) and *P. operculella* (B and D) at two different developmental stages. Note the paler pink color in *P. operculella* and the black cephalic case, versus the darker pink and brown cephalic case of *T. solanivora*..... 10
- Figure 12.** Difference in size of *T. solanivora* pupae between sexes. Left: female, right: male. Note that the lengths do not correspond to those established by Ricón (2002), which denotes the variability of sizes depending on specific conditions.. .... 11
- Figure 13.** Genital pore position. Left: female pupa with pore in the fourth segment, right: male pupa with pore in the fifth segment..... 12
- Figure 14.** A) Female *T. solanivora* with the three characteristic rounded markings. B) Female *P. operculella* C) Spicules of the adult female of *T. solanivora*..... **Error! Marcador no definido.**
- Figure 15.** Different wing patterns of moths of the potato complex: A) *T. solanivora*. B) *P. operculella*. C) *S. tangolias* ..... **Error! Marcador no definido.**
- Figure 16.** A) Infested potatoes with short galleries typical of the Guatemalan potato moth. B) Detail of clean exit holes. C) Detail of damage in cross-section, note that the moth damage is concentrated on the edges near the surface..... 14
- Figure 17.** Potato infested by *P. operculella* with soiled exit holes with traces of faeces..... 14

<b>Figure 18.</b> Average annual population trend of <i>T. solanivora</i> .....	15
<b>Figure 19.</b> Container for infested potatoes. ....	17
<b>Figure 20.</b> A) Top: water trap schematic. Bottom: water trap and adult <i>T. solanivora</i> collected from the trap. B) Funnel trap.....	18
<b>Figure 21.</b> Infection process of <i>B. thurigiensis</i> in insect larvae. ....	23
<b>Figure 22.</b> Signs of the development of granulovirus infection in the larval stage of <i>T. solanivora</i> . Diseased larvae: A) day 7, B) day 12, C) day 17, D) day 20, 5) detail of agglomerations of granulovirus inclusion bodies over the 5th, 6th and 7th abdominal segment. Healthy larvae: 1) day 7, 2) day 12, 3) day 17, 4) day 20 .....	24
<b>Figure 23.</b> Symptomatology of baculovirus-infected larvae. A) Healthy larva with normal pink coloration, versus milky white larvae due to infection. B) Larva in an intermediate stage of infection with a pale pink color. C) and D) Larva in an intermediate stage of infection bleached with some flecks of natural pink on the abdomen and bleached in the dorsoventral area. E) Larva necrosed by infection. ....	25
<b>Figure 24.</b> <i>Steinernema carpocapsae</i> .....	27
<b>Figure 25.</b> A) Last instar larva of <i>Tecia solanivora</i> affected by the entomopathogenic nematode <i>Steinernema feltiae</i> . B) Mass of infective juvenile <i>S. feltiae</i> in the intersegmental spaces of a final instar larva of <i>Tecia solanivora</i> . C) Rubbery appearance and disintegration of tissues under dissection of a final instar larva of <i>T. solanivora</i> .....	27
<b>Figure 26.</b> <i>Copidosoma koehleri</i> stages inside the mummy: A) Eggs. B) Larva.C) Pupa. D) Adult emerged. ....	29
<b>Figure 27.</b> Evolution of the external appearance of the mummy of <i>T. solanivora</i> produced by parasitoidism of <i>C. koehleri</i> : A) Mummy with newly formed pupae. B) Mummy with pupae with black melanized structures. C) Empty mummy, when the adults have already emerged. ....	29
<b>Figure 28.</b> Sexual dimorphism in antennae in <i>Trichogramma</i> sp. Left: female with smooth-looking antennae. Right: male with feathery antennae .....	31
<b>Figure 29.</b> A) Healthy egg of <i>T. solanivora</i> . B) <i>Trichogramma</i> sp. ovipositing in a healthy egg. C) Parasitized egg with black coloration due to the growth of parasitoid larvae inside. D) Empty parasitized egg with exit hole due to the emergence of <i>Trichogramma</i> sp. ....	32
<b>Figure 30.</b> Ichneumonid found on <i>T. solanivora</i> . Left: male. Right: female just emerged from pupa.....	32
<b>Figure 31.</b> Left: longitudinal section of <i>T. solanivora</i> , right: typical cross-section of an ichneumonid.....	33
<b>Figure 32.</b> <i>Blattisocius tarsalis</i> preying on an <i>T. solanivora</i> egg.....	33

## Chapter I

- Figure 1.** Summary of laboratory assay methods (no-choice, choice, functional response and illumination acceptance). TCS = *T. solanivora*; PTH = *P. operculella*. ..... 41
- Figure 2.** Summary of the searching semi-field assay. .... 44
- Figure 3.** Mean number of parasitized *T. solanivora* (TCS) and *P. operculella* (PTH) eggs ( $\pm$ SE) when which are exposed to *T. euproctidis* and *T. achaeae* females at 25 °C under 70% RH with 16L:8D. Different letters denote significant differences in the number of parasitized eggs between treatments with different combinations of parasitoids (*T. euproctidis* or *T. achaeae*) and egg hosts ( $p = 0.05$ ). ..... 45
- Figure 4.** Number of parasitized *T. solanivora* (TCS) and *P. operculella* (PTH) eggs ( $\pm$ SE) and obtained Manly Index (MI) when which are exposed to *T. euproctidis* and *T. achaeae* females the same time at 25 °C under 70% RH with 16L:8D. Different lowercase letters represent significant differences in the number of parasitized eggs (one-way ANOVA and Tukey's test;  $p = 0.05$ ), and "\*" indicates significant preference (*t*-test;  $p < 0.05$ ) of each parasitic wasp..... 45
- Figure 5.** Functional response of *T. euproctidis* female parasitizing *T. solanivora* eggs at four temperature levels (15 °C, 20 °C, 25 °C and 27 °C) under laboratory conditions (70% RH and 16L:8D). Vertical bars indicate 95% confidence intervals. Note: For a correct interpretation of the figures it should be taken into account that the y-axis scales may be different. .... 47
- Figure 6.** Functional response of *T. achaeae* female parasitizing *T. solanivora* eggs at four temperature levels (15 °C, 20 °C, 25 °C and 27 °C) under laboratory conditions (70% RH and 16L:8D). Vertical bars indicate 95% confidence intervals. Note: For a correct interpretation of the figures, it should be taken into account that the y-axis scales may be different..... 48
- Figure 7.** Mean number of eggs parasitized by *T. solanivora* ( $\pm$ SE) for *T. euproctidis* compared to *T. achaeae* at a host density of 90 eggs in the temperature range of 15–27 °C. Different uppercase letters represent differences in the number of parasitized eggs among the tested temperatures in each species. Different lowercase letters indicate significant differences between the two species (two-way ANOVA and Tukey's test,  $p = 0.05$ )..... 49
- Figure 8.** Mean of sex ratio (F:M) ( $\pm$ SE) obtained for *T. achaeae* and *T. euproctidis* with temperature over *T. solanivora* eggs. Different lowercase letters indicate significant differences between temperatures (two-way ANOVA and Tukey's test,  $p = 0.05$ ) in each species. "\*" indicates significant differences between the two species at the same temperature (*t*-test,  $p = 0.05$ )..... 50
- Figure 9.** Mean number of *T. solanivora* eggs parasitized ( $\pm$ SE) by *T. euproctidis* and *T. achaeae* under light conditions compared with darkness conditions at 25 °C and 70% RH. Values with different letters indicated significant differences (Kruskal-Wallis test,  $p = 0.05$ )..... 51
- Figure 10.** Mean ( $\pm$ SE) values of the number of surviving *T. solanivora*. Values with different letters indicated significant differences (one-way ANOVA and Tukey's test,  $p = 0.05$ ). ..... 52
- Figure 11.** Mean values of the number of mines per tuber ( $\pm$ SE) and the percentage of undamaged tubers ( $\pm$ SE) obtained in each treatment. Different letters indicate significant differences (omnibus test,  $p = 0.05$ )..... 52

## Chapter II A

- Figure 1.** (A) Healthy egg. (B) Partially consumed egg. (C) Totally consumed egg. .... 61
- Figure 2.** Mean percentage values of partial killed eggs ( $\pm$ SE) and number killed eggs ( $\pm$ SE) in the predatory potential bioassays. Values with different letters mean significant differences at  $p = 0.05$ , using the Wald test in the case of number of killed eggs and Kruskal–Wallis test for partially consumed eggs. .... 65
- Figure 3.** Mean values of surviving eggs ( $\pm$ SE) and mortality rate ( $\pm$ SE) at the infestation levels of 10 and 50 *Tecia solanivora* eggs and doses of 0, 5, 10, and 20. Values with different letters denote significant differences at  $p = 0.5$ , using Wald test. .... 65
- Figure 4.** Functional response (number of pest eggs killed), type II, of the adult female of *B. tarsalis* tarsalis when different densities of *Tecia solanivora* eggs were offered as prey for 24 h under laboratory conditions (whisker plot: 95% confidence intervals). .... 67

## Chapter II B

- Figure 1.** Summary steps of microcosm assay. .... 75
- Figure 2.** Summary steps of semi-storage assay. .... 76
- Figure 3.** Experimental design of treatments in the storage assay: In 3, “\*” indicates the location of the ten infested tubers in the fruit plastic boxes (up, middle or bottom) .... 78
- Figure 4.** Mean values of surviving eggs ( $\pm$ SE) and mortality rate ( $\pm$ SE) at the infestation levels of 10 (A) and 50 (B) *T. solanivora* eggs, and doses of 0, 5, 10 and 20 mites. Values with different letters denote significant differences (Omnibus test,  $p = 0.05$  for low density (A); one-way ANOVA, Tukey test,  $p = 0.05$  for high density (B)). .... 79
- Figure 5.** Mean number of surviving *T. solanivora* ( $\pm$ SE). Different letters indicate significant differences (one-way ANOVA, Tukey test,  $p = 0.05$ ). .... 80
- Figure 6.** Mean number of mines per tuber ( $\pm$ SE) (A) and mean percentage of undamaged tubers ( $\pm$ SE) (B). Different letters indicate significant differences (one-way ANOVA, Tukey test,  $p = 0.05$ ). .... 80
- Figure 7.** Mean number of mines per tuber ( $\pm$ SE) (A) and mean percentage of undamaged tubers ( $\pm$ SE) (B) of Control and Mite (*B. tarsalis*) treatment at storage conditions. Different letters denote significant differences (one-way ANOVA, Turkey-test,  $p = 0.05$  and Omnibus test,  $p = 0.05$ ; respectively). .... 81

# Table index

## General introduction

<b>Table 1:</b> List of the main varieties and species of Ancient Potatoes of the Canary Islands from Tenerife. ....	2
<b>Table 2.</b> Taxonomic classification of <i>T. solanivora</i> .....	6
<b>Table 3.</b> Duration in days of each phase of the <i>T. solanivora</i> lifecycle and number of generations at 15, 20 and 25°C. Table based on Martinelli et al. (2004) and own data. Authors: A) Nortz 1996. B) Torres et al. 1997, 1998. C) Carnero et al. 2008.....	8
<b>Table 4.</b> Natural enemies associated with <i>T. solanivora</i> are documented in the different countries where it is present (Carrillo and Torrado-León 2013).....	22
<b>Table 5.</b> Taxonomic classification of <i>Copidosoma koehleri</i> . ....	28

## Chapter I.

<b>Table 1.</b> Parameters and statistical significance for the type I, II and III functional response equations when different densities of <i>T. solanivora</i> eggs were exposed to the adult female of <i>T. euproctidis</i> for 24 h under laboratory conditions. “*” indicates the lowest $AIC_c$ value at each temperature. ....	46
<b>Table 2.</b> Parameters and statistical significance for the type I, II and III functional response equations when different densities of <i>T. solanivora</i> eggs were exposed to the adult female of <i>T. achaeae</i> for 24 h under laboratory conditions. “*” indicates the lowest $AIC_c$ value at each temperature. ....	48
<b>Table 3.</b> Mean number ( $\pm$ SE) of emerged adults per parasitized egg obtained in the functional response assays at each temperature for <i>T. euproctidis</i> and <i>T. achaeae</i> . ....	50

## Chapter II A.

<b>Table 1.</b> Average values ( $\pm$ SE) of the predatory potential evaluated at 10, 20, 25, and 27 °C.....	64
<b>Table 2.</b> Mean number ( $\pm$ SE) of killed eggs in the “choice” trials. ....	66
<b>Table 3.</b> Results of logistic regression analyses of the proportion of <i>Tecia solanivora</i> eggs killed by the adult female of <i>B. tarsalis tarsalis</i> in the bioassay carried out under laboratory conditions.. <b>Error! Marcador no definido.</b>	
<b>Table 4.</b> Parameters and statistical significance for the functional response equations type I, II, and III when different densities of <i>Tecia solanivora</i> eggs were exposed to the adult female of <i>B. tarsalis tarsalis</i> , during 24 h, under laboratory conditions.....	67