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RESEARCH ARTICLE

Host selection for the mass production of *Trichogrammatoidea cryptophlebiae* (Nagaraja, 1979) (Hymenoptera: Trichogrammatidae)

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The generalist egg parasitoid *Trichogrammatoidea cryptophlebiae* (Nagaraja, 1979) (Hymenoptera: Trichogrammatidae) is mass-reared in South Africa on *Thaumatotibia leucotreta* (false codling moth). However, alternative tortricid hosts may improve or enhance production. This study assessed the suitability of *Cydia pomonella*, *Cryptophlebia peltastica* and *Lobesia vanillana* as alternative hosts for the parasitoid through no-choice and choice trials. Host acceptance, emergence, superparasitism, unsuccessful emergence, sex ratio, and offspring morphology were measured. In no-choice assays, *T. leucotreta* and *L. vanillana* yielded the highest parasitism, while *C. peltastica* showed significantly less parasitism. Emergence was highest from *C. pomonella* and *C. peltastica*, but high superparasitism in these species resulted in increased unsuccessful development compared to the other species. *Lobesia vanillana*, the smallest host, classified by egg size, produced significantly smaller offspring of the parasitoid. Morphological analyses showed limited differences in forewing length, but significant host- and sex-dependent variation in hind tibia length. Choice trials revealed a consistent preference for *T. leucotreta*, particularly eggs that had been irradiated, suggesting its continued suitability for mass production of *T. cryptophlebiae*. These findings highlight key host-related traits affecting *T. cryptophlebiae* biology and rearing potential and confirm *L. vanillana* and *C. peltastica* as physiological hosts.

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INTRODUCTION

Successful mass-rearing of beneficial insects requires an understanding of species-specific biological and ecological parameters, as well as by the intended application of the organism, including its use for pest suppression through predation or parasitism (Leppla et al. 2023). Globally, more than 350 species of natural enemies have been reared in sufficient quantities for commercial release (van Lenteren et al. 2003; van Lenteren et al. 2018). Ensuring biological integrity and developmental success in mass-reared insects is critical, particularly through careful host selection and quality assurance measures (van Lenteren et al. 2003).

In South Africa, *Trichogrammatoidea cryptophlebiae* (Nagaraja, 1979) (Hymenoptera: Trichogrammatidae), an egg parasitoid, has been successfully mass-reared for decades on false codling moth, *Thaumatotibia leucotreta* (Meyrick, 1913) (Lepidoptera: Tortricidae) eggs for research and commercial application (Newton 1988a,b; Newton 1989; Newton and Odendaal 1990; Moore and Fourie 1999; Moore and Richards 2000, 2001, 2002; Moore et al. 2015). *Trichogrammatoidea cryptophlebiae* is known to parasitise several tortricid species, primarily within the Grapholitini and Olethreutini tribes, including *Cydia pomonella* (Linnaeus, 1758) (codling moth), *Cryptophlebiae peltastica* (Meyrick, 1921) (litchi moth), *Cryptophlebiae ombrodelta* (Lower, 1898) (Australian macadamia nut borer), *Epiblema strenuana* (Walker, 1863) (ragweed borer), *Thaumatotibia batrachopa* (Meyrick, 1908) (South African macadamia nut borer), and *Lobesia botrana* (Denis & Schiffermüller, 1775) (European grapevine moth) (Newton and Crause 1990; Chambers et al. 1995; Kaspi et al. 2020; BioResources 2023). Its ability to successfully parasitise multiple hosts suggests potential for alternative mass-rearing hosts beyond *T. leucotreta*, which could improve the efficiency or scalability of large-scale production systems. However, the suitability of alternative hosts must be carefully evaluated, as host species can influence key parameters including parasitism rate, emergence, sex ratio, and adult fitness.

Egg parasitoid-host interactions occur at two critical stages: initial host acceptance by the adult female and subsequent pre-imaginal development within the egg. Female parasitoids assess host suitability based on egg morphology such as size, chorion thickness, and nutritional content, which in turn influences oviposition decisions and the developmental success of the offspring (Saour 2009; Parra 2010; Pehlivan 2021). Differences in host egg characteristics may lead to varying levels of host acceptance, superparasitism, or sex allocation, all of which can affect rearing output. For instance, smaller host eggs may be more likely to receive unfertilised eggs, producing male offspring, which are smaller, less resource-demanding, and have reduced longevity compared to females (Suzuki et al. 1984; Cherif et al. 2021). Larger or more nutrient-rich eggs may allow for increased female development and better fitness outcomes. However, superparasitism can reduce emergence and skew the sex ratio due to intra-host competition as a single, or multiple females have oviposited more than one egg into a single host (Suzuki et al. 1984; Kaspi et al. 2020).

Trichogrammatoidea cryptophlebiae and other egg parasitoids face nutritional constraints during their pre-imaginal development due to the host egg's characteristics (Martel et al. 2011; Zhang et

al. 2024). The nutritional content, including the volume, size, and age of the host egg, significantly influences the fitness and developmental success of the emergent parasitoid (Pehlivan 2021). Several trials reported that younger eggs yield offspring with higher fecundity, longevity, and flight ability (Carpenter et al. 2004; Tian et al. 2017; Cherif et al. 2021). As host eggs age, the nutritional quality declines, potentially resulting in lower fitness for the parasitoid. Studies on artificial and factitious hosts highlight the impact of nutritional elements on parasitoid development (Nordlund et al. 1997; Cherif et al. 2021). Cherif et al. (2021) and Martel et al. (2011) note that increased host egg size can lead to larger female wasps and higher fecundity, as evidenced by *Trichogramma euproctidis* (Girault, 1991) (Hymenoptera: Trichogrammatidae) having higher fecundity when developing in larger hosts.

Given these interactions of the parasitoid with its host, host selection for mass-rearing should prioritise eggs that are highly accepted by ovipositing females, support successful development, yield a favourable sex ratio, and result in fit, fecund offspring. To date, while *T. leucotreta* has been the primary host for *T. cryptophlebiae* mass-rearing, there has been limited comparative evaluation of alternative host species. Thus, this study aimed to determine which of four host species, *T. leucotreta*, *C. pomonella*, *C. peltastica* (Lepidoptera: Tortricidae), or Vanillana vine moth, *Lobesia vanillana* (Joannis, 1900) (Lepidoptera: Tortricidae), is most suitable for the mass-rearing of *T. cryptophlebiae*.

This study hypothesised that host species significantly influence host acceptance, developmental success, and offspring fitness of the egg parasitoid *T. cryptophlebiae*, resulting in differences in suitability among tortricid hosts for mass-rearing. Specifically, *T. leucotreta*, the current rearing host, would show higher acceptance and preference than alternative hosts, and that host egg size and species identity would affect parasitism rates, emergence, superparasitism, unsuccessful emergence, sex ratio, and offspring morphology. The objectives of this study were to compare parasitism and pre-imaginal development of *T. cryptophlebiae* on *T. leucotreta*, *L. vanillana*, *C. pomonella*, *Helicoverpa armigera* (Hübner, 1809) (Lepidoptera: Noctuidae) and *C. peltastica* using no-choice and choice assays; to evaluate host-mediated effects on offspring quality through measurements of sex ratio and morphological traits; to assess host preference hierarchies, including responses to irradiated versus non-irradiated *T. leucotreta* eggs; and to identify physiologically suitable hosts with potential application in mass-rearing and biological control programmes.

MATERIALS AND METHODS

Source of insects

Trichogrammatoidea cryptophlebiae was sourced from River Bioscience's production culture (Gqeberha, Eastern Cape Province, South Africa), where it was mass-reared at a temperature of 25 °C ± 1, a relative humidity of 60% ± 5, and a photoperiod of 16:8 Light:Dark (L:D), on 180 gamma-ray irradiated *T. leucotreta* eggs, which were sourced from the XSIT *T. leucotreta* mass-rearing facility in Citrusdal, Western Cape, South Africa. *Thaumatotibia leucotreta* eggs are a byproduct from the XSIT mass-rearing facility, where *T. leucotreta* are mass-reared for the sterile insect technique to suppress wild *T. leucotreta* populations. The development of the egg is halted through irradiation of the *T. leucotreta* eggs, thereby sterilising the egg, which allows for *T. cryptophlebiae* to be released in the field and avoiding the risk of *T. leucotreta* larvae hatching in the orchards.

Thaumatotibia leucotreta, L. vanillana, C. peltastica and *H. armigera* eggs were sourced from locally available cultures. *Thaumatotibia leucotreta* species were reared at a temperature of 26 °C ± 1, relative humidity of 50% ± 5, and a photoperiod of 12:12 L:D. *Lobesia vanillana* was reared at a temperature of 24 °C ± 1, relative humidity of 50% ± 5, and a photoperiod of 12:12 L:D. *Cryptophlebiae peltastica* was reared at a temperature of 25 °C ± 1, relative humidity of 70% ± 5, and a photoperiod of 12:12 L:D. *Helicoverpa armigera* was reared at a temperature of 25 °C ± 1, relative humidity of 65% ± 5, and a photoperiod of 12:12 L:D. *Cydia pomonella* eggs were reared at 26 °C, relative humidity of 65% ± 5, and a photoperiod of 16:8 L:D.

For all tests, *T. cryptophlebiae* was provided with a modified version of Perera and Hemachandra's (2014) carbohydrate source, consisting of filter paper soaked in a 20% sugar solution to saturation. The excess sugar solution was allowed to drain off the filter paper, and this was done to prevent *T. cryptophlebiae* adults from becoming stuck in the carbohydrate source.

No-choice assays

Ten sets of 100 eggs from each species, *T. leucotreta, L. vanillana, C. pomonella, C. peltastica*, and *H. armigera,* were counted under a dissecting microscope and placed into glass vials. *Helicoverpa armigera* eggs were used as a negative control to confirm if *T. cryptophlebiae* can parasitise host eggs outside of the Tortricidae family.

A male and female pair of *T. cryptophlebiae* adults were placed into the glass vials with the carbohydrate source and respective host eggs. This was done by placing a petri dish of newly emerged *T. cryptophlebiae* in a fridge at 10 °C for 1 hour to reduce movement. The *T. cryptophlebiae* adults are sexed by the presence of setae on the male antennae and the absence of setae on the female antennae. The *T. cryptophlebiae* adults were carefully picked up using a modified paintbrush and placed into the glass vials. The glass vials were closed using cotton wool to prevent *T. cryptophlebiae* from escaping.

The host eggs were exposed to *T. cryptophlebiae* adults for 48 hours. The host egg cards were removed from the glass vials and placed into Petri dishes, allowing the larvae to hatch. The parasitoids developed in the host eggs for 16 days at a temperature of 25 °C ± 1, a relative humidity of 60% ± 5, and a photoperiod of 16:8 L:D. Hatched larvae were removed from the Petri dishes. The Petri dishes were sealed with parafilm to prevent the escape of emerging *T. cryptophlebiae* offspring. Thirty eggs per host had their lengths (*2a*) and widths (*2b*) measured using a dissecting microscope and ocular micrometre to calculate the defined ellipse (*axbxπ*) (Kaspi et al. 2020).

$$a(\text{mm}) = \frac{\text{Length}(2a)}{2}$$

$$b(\text{mm}) = \frac{\text{Width}(2b)}{2}$$

$$\text{Ellipse}(\text{mm}^2) = axbx\pi$$

The number of parasitised eggs, emergence holes, superparasitism, and sex ratio were determined under a dissecting microscope using a laminate counting grid, tally counter, and soft forceps.

For each individual wasp, measurements of their forewing and hind tibia were taken. This was done to determine the variation in the size of male and female wasps emerging from different hosts. Individual wasps were mounted on a glass slide with a 0.01 mm ruler, with a drop of 70% ethanol and a cover slip placed on top, and images were captured under a compound light microscope (Zeiss Primo Star) with a mounted camera (Zeiss Axiocam ERc 5s). The magnification of the microscope used was set to 4x/0.10, and the images were used for the measurements. Forewing measurements were done by measuring along the central vein on their wing, and hind tibia measurements were done from the femur joint to the tarsal joint. The measurements were then taken using ImageJ 1.54d software, where every sample had the ruler calibrated to ensure measurements were to the nearest 0.01 mm.

Choice assays

Ten sets of 50 eggs from the respective tortricid cultures were paired (Table 1) and placed into a glass vial. A single-mated adult female *T. cryptophlebiae* was placed into the glass vial for 120 minutes to parasitise eggs (Kaspi et al. 2020). The glass vial was closed with cotton wool to prevent the female *T. cryptophlebiae* from escaping. After 120 minutes, the eggs were removed from the vial containing the female and placed into a Petri dish for 16 days, where the eggs were allowed to develop at a temperature of 25 °C ± 1, a relative humidity of 60% ± 5, and a photoperiod of 16:8 L:D. Thirty samples of each tortricid egg pairing were taken.

Helicoverpa armigera was excluded from the choice tests as no parasitism was found in the no-choice trials.

The number of parasitised eggs and emergence holes was counted under a dissecting microscope using a laminate counting grid, tally counter, and soft forceps.

Statistical analysis

No-choice assays

Parasitism, emergence, unsuccessful emergence, and superparasitism rates across different host species were analysed using generalised linear mixed models (GLMM). Host species were treated as a categorical fixed effect with four levels (*T. leucotreta, L. vanillana, C. pomonella, C. peltastica*), and replicate was included as a random effect to account for non-independence among repeated observations. For parasitism, a negative binomial distribution with a log-link function was used to model parasitism counts, with the total number of host eggs included as an offset to model parasitism rate per egg. For emergence, unsuccessful emergence, and superparasitism rates, a negative binomial distribution with a log-link function was used to model emergence counts, incorporating the number of parasitised eggs as an offset (only replicates with successful parasitism events (parasitism > 0) were included to avoid division by zero in the model offset). Two models were fitted, a null model including only the random effect and a full model including host species as a fixed effect. Model comparison was performed using a Wald test. Post hoc pairwise comparisons between host species were performed using estimated marginal means and Bonferroni-adjusted *p*-values.

Sex ratio (calculated as the proportion of male wasps relative to the total number of emerged wasps) was analysed across different host species using a GLMM. Sex ratio data were modelled using a binomial distribution with a logit-link function. The host species was included as a fixed effect, with *T. leucotreta* set as the reference category. Replicate was included as a random effect to account for repeated measures. A null model including only the random effect and a full model including host species as a fixed effect. Model comparison was conducted using a Wald test. Post hoc pairwise comparisons were performed between host species using estimated marginal means with Bonferroni-adjusted *p*-values.

To assess whether the ellipse size differed among host species eggs, a GLMM with a Gamma distribution and log-link was used. The response variable was the ellipse area (mm^2). The host species was included as a fixed effect, and the replicate was included as a random effect to account for variation across experimental units. A Wald test was used to compare the full model (with host species) to the null model. Pairwise comparisons were corrected using the Bonferroni method.

Morphological trait data were analysed to determine whether forewing length and hind tibia length of *T. cryptophlebiae* offspring emerging from different host species, *T. leucotreta, L. vanillana, C. pomonella,* and *C. peltastica*, varied between sexes and host species. A unique replicate identifier was created by combining the replicate number, the sample number, and the sex. For each morphological trait (forewing length and hind tibia length), GLMMs were fitted. Host species and sex were included as fixed effects, and their interaction was incorporated to test whether the effect of host species differed between sexes. Replicate was included as a random effect to account for the non-independence of observations. Two models were compared for each trait: a null model, including only additive effects of host and sex (no interaction), and a full model, including host, sex, and their interaction. Models were compared using a Wald test. Estimated marginal means were calculated, and significant differences among groups were evaluated with Bonferroni-adjusted post hoc tests. All models were fitted in R version 4.5.0 (R Core Team 2025).

Table 1: The pairings of tortricid eggs that were used in choice trials. Abbreviations indicate species names, FCM Non-irradiated – *T. leucotreta* non-irradiated eggs, FCM Irradiated – *T. leucotreta* irradiated eggs, LV – *L. vanillana*, CM – *C. pomonella*, and LM – *C. peltastica*.

Tortricid 1	Tortricid 2
CM	FCM Non-irradiated
CM	FCM Irradiated
CM	LM
CM	LV
LM	FCM Non-irradiated
LM	FCM Irradiated
LM	LV
LV	FCM Non-irradiated
LV	FCM Irradiated
FCM Non-irradiated	FCM Irradiated

Choice assays

To assess whether parasitism preference differed across species combinations and within species pairings, linear mixed models (LMMs) and non-parametric tests were conducted. To do this, the parasitism difference for each replicate was calculated as the number of eggs parasitised by species A minus the number parasitised by species B. Positive values indicated a preference for species A, while negative values indicated a preference for species B. An LMM was fitted to test the effect of species combination on parasitism difference, with species combination as a fixed effect and replicate as a random effect. Model significance was assessed by comparing the full model to a null model containing only the random effect using a Wald test ($p < 0.05$). All models were fitted in R version 4.5.0 (R Core Team, 2025).

RESULTS

No-choice assays

Parasitism differed significantly among host species in the no-choice assay ($\chi^2 = 228.05$, df = 4, $p < 0.001$), with the inclusion of host species significantly improving model fit over the null model. Relative to *T. leucotreta* ($\beta = -1.43$, SE = 0.18, $p < 0.001$), parasitism was significantly lower on *C. peltastica* ($\beta = -1.07$, SE = 0.15, $p < 0.001$) and *C. pomonella* ($\beta = 0.36$, SE = 0.15, $p = 0.013$), but not significantly different on *L. vanillana* ($\beta = -0.25$, SE = 0.15, $p = 0.085$). Parasitism on *H. armigera* was effectively zero, and model estimates for this species were unstable, leading to its exclusion from Figure 1. ($\beta = -23.04$, SE = 3433.62, $p = 0.995$). Post hoc comparisons using Bonferroni correction confirmed that parasitism on *C. peltastica* (8.20 ± 0.19%) and *C. pomonella* (16.70 ± 0.18%) was significantly lower than on *T. leucotreta* (23.92 ± 0.18%) and *L. vanillana* (18.63 ± 0.18%) ($p < 0.001$). No significant difference in parasitism was recorded between *T. leucotreta* and *L. vanillana* ($p = 0.511$) or between *C. pomonella* and *L. vanillana* ($p = 1.000$) (Figure 1).

The proportion of parasitoids emerging differed significantly across host species ($\chi^2 = 76.05$, df = 3, $p < 0.001$), with the inclusion of host species as a fixed effect significantly improving model fit over the null model. Relative to *T. leucotreta* (β = −0.17, SE = 0.05, $p < 0.001$), emergence was significantly higher on *C. pomonella* (β = 0.31, SE = 0.04, $p < 0.001$) and *C. peltastica* (β = 0.32, SE = 0.06, $p < 0.001$) but did not differ significantly on *L. vanillana* (β = 0.03, SE = 0.05, $p = 0.590$). Post hoc comparisons using Bonferroni correction confirmed that emergence on *C. peltastica* (137.3 ± 0.06%) and *C. pomonella* (133.3 ± 0.05%) was significantly higher than on *T. leucotreta* (98.5 ± 0.05%) and *L. vanillana* (95.5 ± 0.05%) ($p < 0.001$). No significant difference in emergence was recorded between *T. leucotreta* and *L. vanillana* ($p = 1.000$) or between *C. pomonella* and *C. peltastica* ($p = 1.000$) (Figure 2).

Superparasitism proportion differed significantly across host species ($\chi^2 = 232.52$, df = 3, $p < 0.001$), with the inclusion of host species as a fixed effect significantly improving model fit over the null model. Relative to *T. leucotreta* (β = −3.39, SE = 0.17, $p < 0.001$), superparasitism was significantly higher on *C. pomonella* (β = 2.34, SE = 0.16, $p < 0.001$) and *C. peltastica* (β = 2.27, SE = 0.18, $p < 0.001$), and significantly lower on *L. vanillana* (β = −0.78, SE = 0.30, $p = 0.009$). Post hoc comparisons using Bonferroni correction confirmed that superparasitism on *C. pomonella* (34.8 ± 0.16%) and *C. peltastica* (32.4 ± 0.18%) was significantly higher than on *T. leucotreta* (3.4 ± 0.17%) and *L. vanillana* (1.5 ± 0.27%) ($p < 0.001$). No significant difference in superparasitism was recorded between *C. pomonella* and *C. peltastica* ($p = 1.000$), while *L. vanillana* differed significantly from all other host species ($p < 0.001$) (Figure 3).

Unsuccessful emergence proportion differed significantly across host species ($\chi^2 = 16.36$, df = 3, $p < 0.001$), with the inclusion of host species as a fixed effect significantly improving model fit over the null model. Relative to *T. leucotreta* (β = −1.71, SE = 0.13, $p < 0.001$), unsuccessful emergence was significantly higher on *C. pomonella* (β = 0.37, SE = 0.11, $p = 0.001$) but did not differ significantly on *C. peltastica* (β = −0.15, SE = 0.16, $p = 0.330$) or *L. vanillana* (β = 0.00, SE = 0.12, $p = 0.979$). Post hoc comparisons with Bonferroni correction confirmed that unsuccessful emergence was significantly higher on *C. pomonella* (26.1 ± 2.9%) than on *T. leucotreta* (18.1 ± 2.4%), *C. peltastica* (15.6 ± 2.6%), and *L. vanillana* (18.2 ± 2.5%) ($p = 0.0072$; $p = 0.0052$; $p = 0.0164$, respectively). No significant differences were detected between *T. leucotreta*, *L. vanillana*, and *C. peltastica* ($p = 1.000$). Only *C. pomonella* differed significantly from all other host species (Figure 4).

Sex ratio (proportion of male offspring) differed significantly across host species ($\chi^2 = 61.25$, df = 3, $p < 0.001$), with the inclusion of host species as a fixed effect significantly improving model fit over the null model. Relative to *T. leucotreta* (β = 0.61, SE = 0.19, $p = 0.002$), sex ratio was significantly lower on *C. pomonella* (β = −0.37, SE = 0.10, $p < 0.001$) and *C. peltastica* (β = −0.85, SE = 0.12, $p < 0.001$), but did not differ significantly on *L. vanillana* (β = 0.00, SE = 0.11, $p = 0.987$). Post hoc comparisons with Bonferroni correction confirmed that sex ratio was significantly lower on *C. peltastica* (0.439 ± 0.028) and *C. pomonella* (0.560 ± 0.026) than on *T. leucotreta* (0.647 ± 0.024) and *L. vanillana* (0.648 ± 0.025) ($p < 0.001$; $p = 0.0013$; $p = 0.0004$; $p = 0.0038$, respectively). No significant difference was detected between *T. leucotreta* and *L. vanillana* ($p = 1.000$). Only *C. peltastica* differed significantly from all other host species (Figure 5).

Ellipse size differed significantly among host species ($\chi^2 = 1585.7$, df = 4, $p < 0.001$). Ellipse size for *C. pomonella* was the largest (mean = 0.66 mm²), followed by *C. peltastica* (0.43 mm²), *T. leucotreta* (0.27 mm²), *L. vanillana* (0.20 mm²), and *H. armigera* (0.19 mm²). *Helicoverpa armigera* and *L. vanillana* showed no significant differences in ellipse size. *Thaumatotibia leucotreta*, *C. pomonella*, and *C. peltastica* differed significantly in ellipse size (Figure 6).

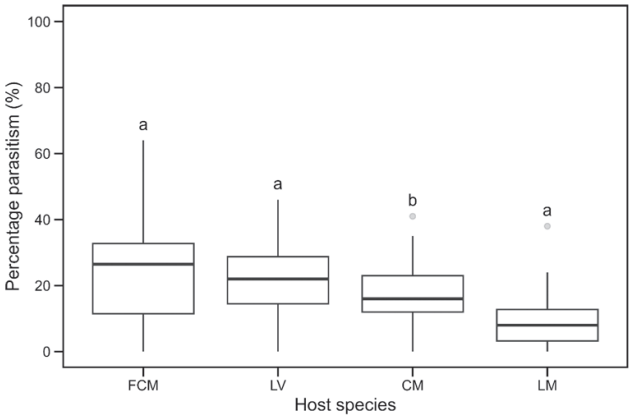

Figure 1: Boxplot of percentage parasitism by *Trichogrammatoidea cryptophlebiae* across host species FCM, LV, CM, and LM. Different letters above the boxes indicate significant differences ($p < 0.05$). Abbreviations indicate species names, FCM – *T. leucotreta*, LV – *L. vanillana*, CM – *C. pomonella*, and LM – *C. peltastica*.

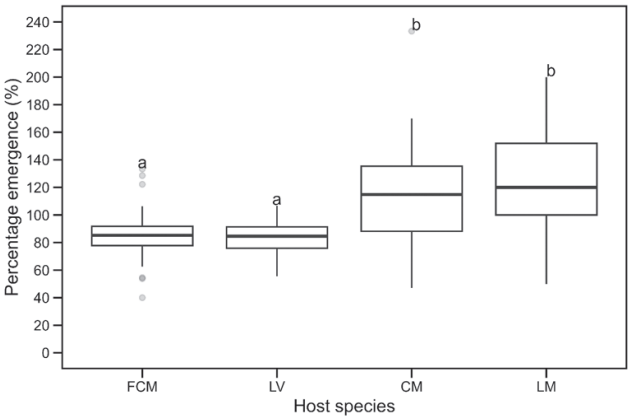

Figure 2: Boxplot of percentage emergence by *Trichogrammatoidea cryptophlebiae* across host species FCM, LV, CM, and LM. Different letters above the boxes indicate significant differences ($p < 0.05$). Abbreviations indicate species names, FCM – *T. leucotreta*, LV – *L. vanillana*, CM – *C. pomonella*, and LM – *C. peltastica*.

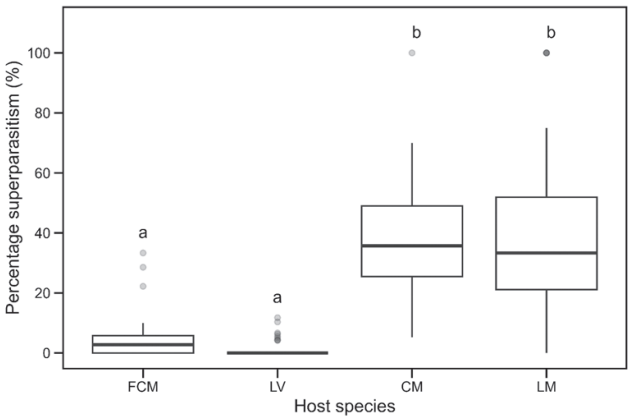

Figure 3: Boxplot of percentage superparasitism by *Trichogrammatoidea cryptophlebiae* across host species FCM, LV, CM, and LM. Different letters above the boxes indicate significant differences ($p < 0.05$). Abbreviations indicate species names, FCM – *T. leucotreta*, LV – *L. vanillana*, CM – *C. pomonella*, and LM – *C. peltastica*.

Hind tibia length also differed significantly across host species and sexes ($\chi^2 = 588.83$, df = 3, $p < 0.001$), with the host-sex interaction again significantly improving model fit. Relative to females reared on *C. pomonella* (β = 0.129, SE = 0.0007), hind tibia length was significantly reduced on *L. vanillana* (β = -0.0154,

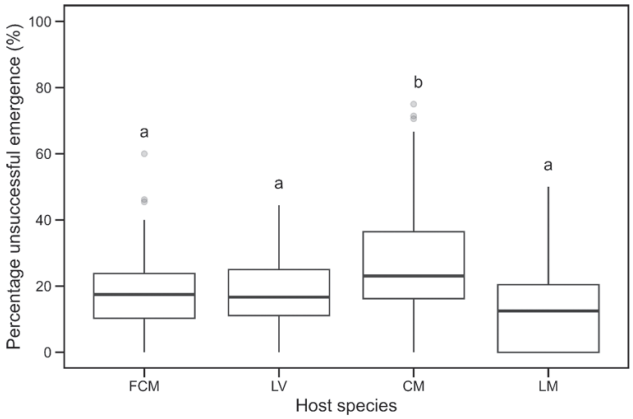

Figure 4: Boxplot of unsuccessful emergence by *Trichogrammatoidea cryptophlebiae* across host species FCM, LV, CM, and LM. Different letters above the boxes indicate significant differences ($p < 0.05$). Abbreviations indicate species names, FCM – *T. leucotreta*, LV – *L. vanillana*, CM – *C. pomonella*, and LM – *C. peltastica*.

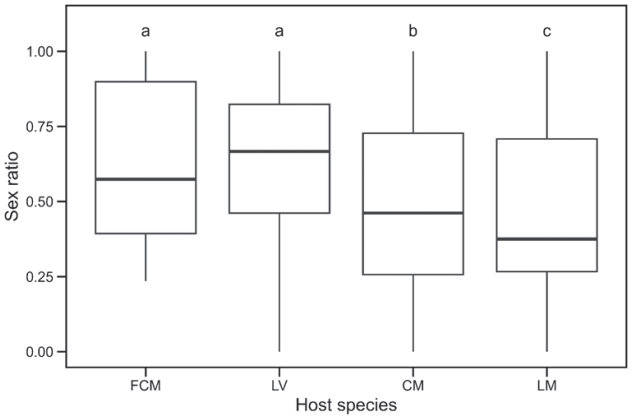

Figure 5: Boxplot of sex ratio by *Trichogrammatoidea cryptophlebiae* across host species FCM, LV, CM, and LM. Different letters above the boxes indicate significant differences ($p < 0.05$). Abbreviations indicate species names, FCM – *T. leucotreta*, LV – *L. vanillana*, CM – *C. pomonella*, and LM – *C. peltastica*.

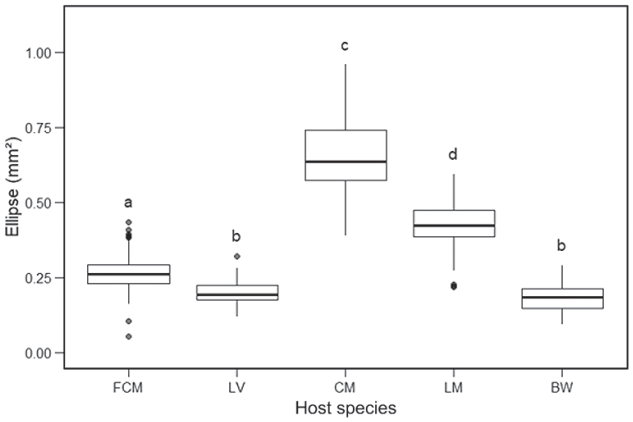

Figure 6: Ellipse size of potential host eggs offered to *Trichogrammatoidea cryptophlebiae* in no-choice testing ($p < 0.05$). Abbreviations indicate species names, FCM – *T. leucotreta*, LV – *L. vanillana*, CM – *C. pomonella*, LM – *C. peltastica*, and BW – *H. armigera*.

SE = 0.0008, $p < 0.001$), slightly increased on *C. peltastica* (β = 0.0017, SE = 0.0008, $p = 0.050$), and showed no difference on *T. leucotreta* (β = -0.0002, SE = 0.0008, $p = 0.793$). The main effect of sex was non-significant (β = –0.0002, SE = 0.0009, $p = 0.805$),

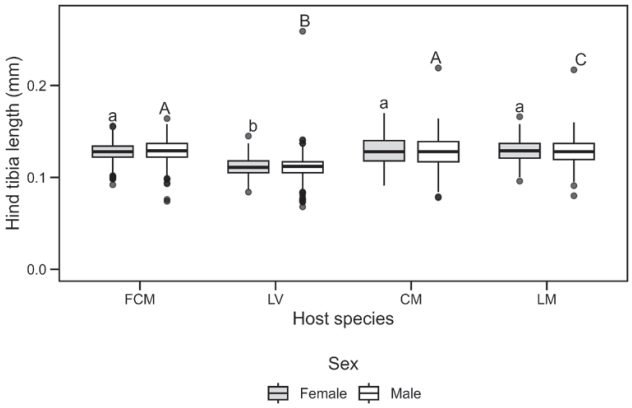

Figure 7: Male and female hind tibial measurements (mm) of *Trichogrammatoidea cryptophlebiae* offspring that emerged from FCM, LV, CM, and LM eggs. Different letters indicate statistically significant differences between sexes emerging from different hosts ($p < 0.05$). Abbreviations indicate species names, FCM – *T. leucotreta*, LV – *L. vanillana*, CM – *C. pomonella*, and LM – *C. peltastica*.

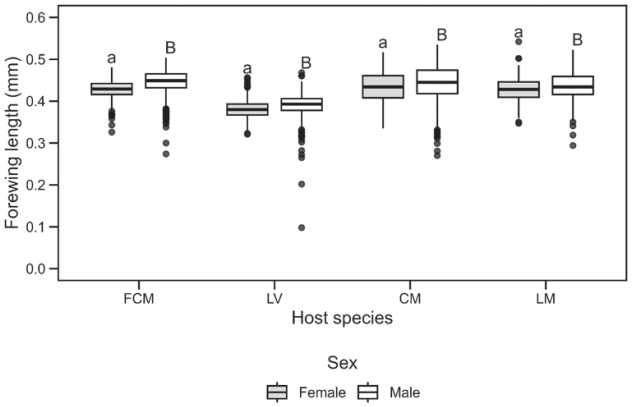

Figure 8: Male and female forewing measurements (mm) of *Trichogrammatoidea cryptophlebiae* offspring that emerged from FCM, LV, CM, and LM eggs. Different letters indicate statistically significant differences between sexes emerging from different hosts ($p < 0.05$). Abbreviations indicate species names, FCM – *T. leucotreta*, LV – *L. vanillana*, CM – *C. pomonella*, and LM – *C. peltastica*.

but a significant host-sex interaction was detected on *C. peltastica* (β = 0.0087, SE = 0.0012, $p < 0.001$), indicating that male tibia length was considerably longer than females only on this host. Post hoc comparisons confirmed that male hind tibia length was longest on *C. peltastica* (0.139 ± 0.0009 mm), while the shortest values were recorded on *L. vanillana* for both sexes (females = 0.113 ± 0.0008 mm; males = 0.114 ± 0.0006 mm) (Figure 7).

Forewing length differed significantly across host species and sexes ($\chi^2 = 128.46$, df = 3, $p < 0.001$), with the inclusion of a host-sex interaction significantly improving model fit over the null model. Relative to females reared on *C. pomonella* (β = 0.434, SE = 0.0018), forewing length was significantly shorter on *T. leucotreta* (β = –0.0063, SE = 0.0023, $p = 0.006$), *C. peltastica* (β = –0.0049, SE = 0.0026, $p = 0.054$), and especially *L. vanillana* (β = –0.0496, SE = 0.0025, $p < 0.001$). Males had significantly longer forewings overall (β = 0.0084, SE = 0.0023, $p < 0.001$), with the greatest sex difference recorded on *C. peltastica* (β = 0.0094, SE = 0.0012, $p < 0.01$). Post hoc comparisons with Bonferroni correction showed that male forewing length was greatest on *C. peltastica* (0.447 ± 0.0024 mm) and *T. leucotreta* (0.445 ± 0.0014 mm), followed by *C. pomonella* (0.443 ± 0.0015 mm), and was smallest on *L. vanillana* (0.392 ± 0.0016 mm). Female forewing length followed a similar pattern, with the longest forewings on *C. pomonella* (0.434 ± 0.0018 mm) and the shortest on *L. vanillana* (0.385 ± 0.0022 mm) (Figure 8).

Choice assays

Parasitism preference differed significantly among host combinations in the choice trial assay (χ^2 = 22.01, df = 9, p = 0.009), with the inclusion of species combination significantly improving model fit over the null model. Relative to the *C. pomonella* vs *T. leucotreta* Non-irradiated (β = −1.36, SE = 1.57, p = 0.395), parasitism differences were significantly more negative in *C. peltastica* vs *T. leucotreta* Irradiated (β = −4.34, SE = 1.78, p = 0.015) and *L. vanillana* vs *T. leucotreta* Irradiated (β = −4.12, SE = 1.78, p = 0.021), indicating stronger preferences for *T. leucotreta* irradiated in those combinations. No significant differences were detected for any other combinations (all $p > 0.05$). Wilcoxon signed-rank tests corroborated these findings. Significant differences in parasitism were recorded for *C. pomonella* vs *T. leucotreta* Irradiated (p = 0.003), *C. peltastica* vs *T. leucotreta* Non-irradiated (p = 0.044), *C. peltastica* vs *T. leucotreta* Irradiated ($p < 0.001$), *L. vanillana* vs *T. leucotreta* Non-irradiated (p = 0.017), and *L. vanillana* vs *T. leucotreta* Irradiated (p = 0.003). In each case, the more heavily parasitised host was *T. leucotreta* Irradiated or *T. leucotreta* Non-irradiated, suggesting consistent preference patterns (Figure 9).

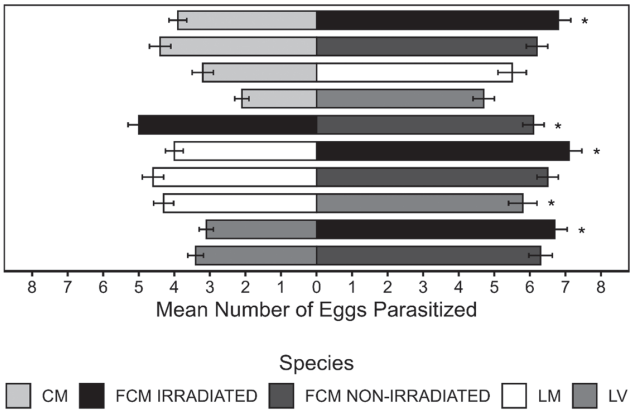

Figure 9: Mean number of eggs parasitised by *Trichogrammatoidea cryptophlebiae* in different paired host choice combinations. The stars indicate a significant preference for one species over the other ($p > 0.05$). Abbreviations indicate species names, FCM Non-irradiated – *T. leucotreta* non-irradiated eggs, FCM Irradiated – *T. leucotreta* irradiated eggs, LV – *L. vanillana*, CM – *C. pomonella*, and LM – *C. peltastica*.

DISCUSSION

Parasitism levels reflect host acceptance for oviposition. In no-choice trials, *T. leucotreta, L. vanillana*, and *C. pomonella* were parasitised at similar rates, consistent with Kaspi et al. (2020), who found comparable parasitism between *T. leucotreta* and *C. pomonella*. In contrast, *C. peltastica* eggs were parasitised at lower rates, suggesting structural or physiological barriers to oviposition.

One possibility is that *C. peltastica* eggs have a thicker or harder chorion, requiring more time for drilling and reducing oviposition success within the 48-hour test period (Pak et al. 1990; Liu et al. 2024; Zhang et al. 2024). Successful parasitism may depend on females having longer and wider ovipositors, which are more capable of penetrating tough chorions (Grenier et al. 2001; Liu et al. 2024; Zhang et al. 2024). Because trichogrammatid sensory receptors are located along the ovipositor, if penetration fails, females may not assess host suitability at all (Le Ralec et al. 1996). It would be valuable to further investigate ovipositor morphology and egg chorion thickness across confirmed hosts (*T. leucotreta, L. vanillana, C. pomonella, C. peltastica*). If ovipositor traits strongly predict success across hosts, rearing programmes can select for females with these traits.

Host egg development time may also play a role, as it alters egg structure and nutrition. Under optimal rearing conditions, *T. leucotreta* eggs hatch in ~3 days, *L. vanillana* and *C. pomonella* in ~6 days, and *C. peltastica* in ~4 days. However, these timelines do not clearly explain parasitism patterns, as *T. leucotreta* did not show the lowest parasitism. Environmental conditions optimised for *T. cryptophlebiae*, especially based on its interaction with *T. leucotreta*, may have unintentionally hindered parasitism on other hosts. Their development rates under trial conditions may have influenced chorion hardening, desiccation, or nutritional changes, resulting in different levels of parasitism.

Host egg size influenced superparasitism. Particularly, larger egg ellipses in *C. pomonella* and *C. peltastica* correlated with higher superparasitism, including emergence rates exceeding 100%. Conversely, *L. vanillana*, which had the smallest eggs, showed almost no superparasitism. This has implications for rearing: high superparasitism can yield more offspring per egg, but it also brings risks. These risks include increased unsuccessful emergence and skewed sex ratios. Male production is typically favoured under crowded pre-imaginal conditions (Suzuki et al. 1984; Parra 2010; Luo et al. 2025). However, in this study, unsuccessful emergence was highest in *C. pomonella*, likely due to poorer egg nutrition linked to adult diet (Martel et al. 2011). This is plausible because *T. leucotreta*, *L. vanillana*, and *C. peltastica* were reared on the same diet, which differs from that of *C. pomonella*. The low emergence levels may be related to this difference. However, further testing, including nutrient analysis of the egg contents of these species, would need to be conducted, among other assessments.

Sex ratios were male-biased across all hosts, which deviates from expectations for trichogrammatids (Luck et al. 2001). As the rearing colony typically produces female-biased broods, this could reflect an artefact of the trial design. Placing a single mating pair with host eggs may have led to repeated copulation, blocking the female's spermatheca and impeding fertilisation (Luck et al. 2001). Given that host egg quality and quantity were sufficient, and crowding was not a factor, this design issue likely explains the male-biased results. Interestingly, *C. pomonella* and *C. peltastica*, which had higher superparasitism, also produced significantly more female offspring. This contradicts prior findings that suggest superparasitism produces more males than females (Suzuki et al. 1984). A possible explanation for this is phenotypic plasticity, which the mass-reared culture used for these trials has possibly undergone, whereby males are larger, and females are smaller, which is the opposite of what Nagaraja (1978) reported for *T. cryptophlebiae*. Thus, as males are larger than females in the culture used for these trials, superparasitism selects for the smaller, more space-efficient females.

Host egg size also affected offspring size. *Lobesia vanillana*, the smallest host, produced the smallest offspring, particularly in hind tibia length, which suggests reduced fertility. This study did not confirm that hind tibial length is a true indicator of fertility for this trichogrammatid species, but literature on other species suggests this to be true when parasitoids are reared on differing hosts. Iranipour et al. (2010) showed that fitness differed in *T. brassicae* Trichogramma brassicae (Bezdenko, 1986) when reared on differing hosts, with the parasitoids having life table parameters that were significantly different when emerging from different hosts. Further studies could look at rearing the F_2 generations of *T. cryptophlebiae* offspring on the various hosts to test if these expected fitness trade-offs are apparent. Despite significant differences in host egg size, offspring size was not dramatically different, likely due to phenological plasticity and spatial limitations imposed by host egg dimensions. Superparasitism may further homogenise offspring size due to competition.

In choice trials, *T. leucotreta* irradiated eggs were typically preferred over other hosts. This, however, is different to what Carpenter et al. (2004) found, where *T. cryptophlebiae* preferred

non-irradiated *T. leucotreta* eggs, on which they had been reared for several generations. These contradictory findings suggest pre-imaginal conditioning from rearing history influences the adult parasitoids choice in which host to oviposit its offspring, although Kaspi et al. (2020) found *T. cryptophlebiae* still favoured *T. leucotreta* even after being reared on *L. botrana* for 10 generations. This suggests a genuine host preference hierarchy when exposed to hosts in a laboratory setting (Bjorksten and Hoffmann 1998). Additionally, Githae et al. (2024) assessed the acceptability of eggs produced by differing ratios of sterile (moths used in sterile insect techniques programmes) and fertile *T. leucotreta* combinations. Githae et al. (2024) demonstrated that *T. cryptophlebiae* were able to develop successfully within sterile eggs, suggesting nutrients and egg structure were not altered significantly to inhibit the successful development of the parasitoid. Parasitism levels did not differ significantly between irradiated and non-irradiated *T. leucotreta* eggs, though irradiated eggs were marginally preferred. Irradiation may soften the chorion, inhibit host defences, and reduce developmental cues, thereby lowering resistance to parasitism (Xu et al. 2016). This suggests that *T. cryptophlebiae* can develop and proliferate from the varying states, irradiated, non-irradiated and sterile, of *T. leucotreta* eggs that they have been exposed to.

From a rearing perspective, *T. leucotreta* remains the most suitable host due to high parasitism, low superparasitism, high emergence, and consistently sized offspring. Future research should assess how offspring from different hosts compare in fitness and whether ovipositor morphology influences parasitism success across hosts. Identifying a cost-effective factitious host with similar benefits remains a worthwhile goal as this could increase production outputs in mass-rearing facilities while ensuring fit wasps are produced in the system. The confirmation of *L. vanillana* and *C. peltastica* as physiological hosts also opens the door to field trials for their biological control. While choice trials suggest clear preferences, it is essential to remember that *T. cryptophlebiae* is a generalist egg parasitoid that may opportunistically exploit multiple tortricid hosts in natural systems. Further work should also be done to assess the sex allocation differences seen in the trials to determine if the rearing system has selected for females to be smaller in comparison to those found in wild populations.

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AUTHOR CONTRIBUTIONS

EJT: conceptualisation, formal analysis, investigation, methodology, project administration, writing – original draft
MM: supervision, conceptualisation, writing – review and editing
ST: supervision, conceptualisation, funding acquisition, resources, writing – review and editing
SM: supervision, writing – review and editing
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REFERENCES

Bioresources (Pty) Ltd. 2023. *MacTrix- Trichogramma Wasps*. Accessed: 24 October 2023. Bioresources (Pty) Ltd. https://bioresources.com.au/good-bugs/mactrix-wasps/

Bjorksten TA, Hoffman AA. 1998. Separating the effects of experience, size, egg load, and genotype on the host response in *Trichogramma* (Hymenoptera: Trichogrammatidae). Journal of Insect Behaviour. 11(1):129–148. https://doi.org/10.1023/A:1020826818884.

Carpenter JE, Bloem S, Hofmeyr JH. 2004. Acceptability and suitability of eggs of false codling moth (Lepidoptera: Tortricidae) from irradiated parents to parasitism by *Trichogrammatoidea cryptophlebiae* (Hymenoptera: Trichogrammatidae). Biological Control. 30:351–359. https://doi.org/10.1016/j.biocontrol.2003.10.006.

Chambers BA, Samways MJ, Ironside DA. 1995. Egg distribution of the fruit borers *Cryptophlebia leucotreta* (Meyrick) and *C. batrochopa* (Meyrick) (Lepidoptera: Tortricidae) and their egg parasitoid *Trichogrammatoidea cryptophlebiae* Nagaraja (Hymenoptera: Trichogrammatidae) in macadamia trees in Malawi. African Entomology. 3(2):181–188.

Cherif A, Mansour R, Grissa-Lebdi K. 2021. The egg parasitoids *Trichogramma*: from laboratory mass rearing to biological control of lepidopteran pests. Biocontrol Science and Technology. 31(7):661–693. https://doi.org/10.1080/09583157.2020.1871469.

Githae MM, Coombes CA, Mutamiswa R, Moore SD, Hill MP. 2024. Suitability of false codling moth eggs from different sterile to fertile moth ratios in the sterile insect technique programme, to parasitism by *Trichogrammatoidea cryptophlebiae*. Crop Protection. 182:106744. https://doi.org/10.1016/j.cropro.2024.106744.

Grenier S, Grille G, Basso C, Pintureau B. 2001. Effects of the host species and the number of parasitoids per host on the size of some *Trichogramma* species (Hymenoptera: Trichogrammatidae). Biocontrol Science and Technology. 11:21–26. https://doi.org/10.1080/09583150020029709.

Iranipour S, Vaez N, Ghanbalani GN, Zakaria RA, Jafarloo MM. 2010. Effect of host change on demographic fitness of the parasitoid, *Trichogramma brassicae*. Journal of Insect Science. 10(78):1–12. https://doi.org/10.1673/031.010.7801.

Kaspi R, Steinitz H, Nemny-Lavi E, Lebedev G, Melamed E, Gazit Y. 2020. Nontarget host risk assessment of the egg parasitoid *Trichogrammatoidea cryptophlebiae* (Hymenoptera: Trichogrammatidae) for classical biological control of the false codling moth (Lepidoptera: Tortricidae) in Israel. Journal of Economic Entomology. 113(2):1023–1027. https://doi.org/10.1093/jee/toz327.

Le Ralec A, Rabasse JM, Wajnberg E. 1996. Comparative morphology of the ovipositor of some parasitic Hymenoptera in relation to characteristics of their hosts. The Canadian Entomologist. 128:413–433. https://doi.org/10.4039/Ent128413-3.

Liu S, Yan Z-C, Zhou F-W, Wang C-X, Li Y-X. 2024. Relationship between parasitism and morphology of *Trichogramma* (Hymenoptera: Trichogrammatidae) on *Spodoptera frugiperda* (Lepidoptera: Noctuidae). Journal of Economic Entomology. 117(1):93–101. https://doi.org/10.1093/jee/toad231.

Luo T-I, Wang Z-Y, Wei X, Jiang Y-T, Xu K, Jiang K, Qui L-F, Hu H-Y, Liu P-C. 2025. Preferentially laying female offspring during self-superparasitism in a quasigregarious parasitoid. Animal Behaviour. 224:123205. https://doi.org/10.1016/j.anbehav.2025.123205.

Luck RF, Janssen JAM, Pinto JD, Oatman ER. 2001. Precise sex allocation, local mate competition, and sex ratio shifts in the parasitoid *Trichogramma pretiosum*. Behavioral Ecology and Sociobiology. 49:311–321. https://doi.org/10.1007/s002650000294.

Martel V, Darrouzet E, Boivin G. 2011. Phenotypic plasticity in the reproductive traits of a parasitoid. Journal of Insect Physiology. 57:682–687. https://doi.org/10.1016/j.jinsphys.2011.01.018.

Moore SD, Fourie J. 1999. Assessment and development of the augmentation technique for FCM control with the parasitoid *Trichogrammatoidea cryptophlebiae*. Experiment 401. 1–9.

Moore SD, Richards GI. 2000. Assessment and development of the augmentation technique for FCM control with the parasitoid *Trichogrammatoidea cryptophlebiae*. Experiment 401. 1–6.

Moore SD, Richards GI. 2001. Assessment and development of the augmentation technique for FCM control with the parasitoid *Trichogrammatoidea cryptophlebiae*. Project: FCM, Experiment 401. 1–13.

Moore SD, Richards GI. 2002. Assessment and development of the augmentation technique for FCM control with the parasitoid *Trichogrammatoidea cryptophlebiae*. Experiment 401. 1–9.

Moore SD, Kirkman W, Mommsen W, Beetge L, Otto H. 2015. Late-season releases of *Trichogrammatoidea cryptophlebiae* for suppression of FCM. Final Report, Project 1021. 1–16.

Nagaraja H. 1978. Studies on *Trichogrammatoidea* (Hymenoptera: Trichogrammatidae). Oriental Insects. 12(4):489–529. https://doi.org/10.1080/00305316.1978.10432534.

Leppla NC, Morales-Ramos JA, Shapiro-Ilan D, Guadalupe Rojas M. 2023. Introduction. In: Morales-Ramos JA, Guadalupe Rojas M, Shapiro-Ilan D, editors. Mass Production of Beneficial Organisms. 2nd edition. London, United Kingdom: Academic Press; pp. 3–12. https://doi.org/10.1016/B978-0-12-822106-8.00007-5.

Newton PJ. 1988a. Movement and impact of *Trichogrammatoidea cryptophlebiae* Nagaraja (Hymenoptera: Trichogrammatidae) in citrus orchards after inundative releases against the false codling moth, *Cryptophlebia leucotreta* (Meyrick) (Lepidoptera: Tortricidae). Bulletin of Entomological Research. 78:85–99. https://doi.org/10.1017/S0007485300016096.

Newton PJ. 1988b. Inversely density-dependent egg parasitism in patchy distributions of the citrus pest *Cryptophlebia leucotreta* (Lepidoptera: Tortricidae) and its agricultural efficiency. Journal of Applied Ecology. 25:145–162. https://doi.org/10.2307/2403615.

Newton PJ. 1989. Combinations of applications of a chitin synthesis inhibitor and inundative releases of egg parasitoids against the false codling moth, *Cryptophlebia leucotreta* (Meyrick) (Lepidoptera: Tortricidae), on citrus. Bulletin of Entomological Research. 79:507–519. https://doi.org/10.1017/S0007485300018472.

Newton PJ, Crause C. 1990. Oviposition on *Litchi chinensis* by *Cryptophlebia* species (Lepidoptera: Tortricidae). Phytophylactica. 22(3):365–367.

Newton PJ, Odendaal WJ. 1990. Commercial inundative releases of *Trichogrammatoidea cryptophlebiae* (Hym: Trichogrammatidae) against *Cryptophlebia leucotreta* (Lepidoptera: Tortricidae) in Citrus. Entomophaga. 35(4):545–556. https://doi.org/10.1007/BF02375089.

Nordlund DA, Wu ZX, Greenberg SM. 1997. *In Vitro* rearing of *Trichogramma minutum* Riley (Hymenoptera: Trichogrammatidae) for ten generations, with quality assessment comparisons of *in Vitro* and *in Vivo* reared adults. Biological Control. 9:201–207. https://doi.org/10.1006/bcon.1997.0534.

Pak GA, Van Dalen A, Kaashoek N, Dijkman H. 1990. Host egg chorion structure influencing host suitability for the egg parasitoid *Trichogramma* Westwood. Journal of Insect Physiology. 36(11):869–875. https://doi.org/10.1016/0022-1910(90)90174-E.

Parra JRP. 2010. Mass rearing of egg parasitoids for biological control programs. In: Cônsoli FL, Parra JRP, Zucchi RA, editors. Egg Parasitoids in Agroecosystems with Emphasis on *Trichogramma*. Dordrecht: Springer; pp. 267–292.

Pehlivan S. 2021. Role of host diet on the fitness of the egg parasitoid species, *Trichogramma evanescens* Westwood (Hymenoptera: Trichogrammatidae). Egyptian Journal of Biological Pest Control. 31:a10. https://doi.org/10.1186/s41938-020-00353-7.

Perera MCD, Hemachandra KS. 2014. Study on longevity, fecundity and oviposition of *Trichogrammatoidea bactrae* Nagaraja (Hymenoptera: Trichogrammatidae) to facilitate mass rearing. Tropical Agricultural Research. 25(4):602–609. https://doi.org/10.4038/tar.v25i4.8065.

R Core Team. (2025). R: A language and environment for statistical computing. R Foundation for Statistical Computing. https://www.R-project.org/

Saour G. 2009. Effect of early oviposition experience on host acceptance in *Trichogramma* (Hymenoptera: Trichogrammatidae) and application of F_1 sterility and *T. principium* to suppress the potato tuber moth (Lepidoptera: Gelechiidae). Biocontrol Science and Technology. 19(sup1):225–234. https://doi.org/10.1080/09583150802522838.

Suzuki Y, Tsuji H, Sasakawa M. 1984. Sex allocation and effects of superparasitism on secondary sex ratios in the gregarious parasitoid, *Trichogramma chilonis* (Hymenoptera: Trichogrammatidae). Animal Behaviour. 32:478–484. https://doi.org/10.1016/S0003-3472(84)80284-5.

Tian JC, Wang ZC, Wang GR, Zhong LQ, Zheng XS, Xu HX, Zang, LS, Lu ZX. 2017. The effects of temperature and host age on the fecundity of four Trichogramma species, Egg parasitoids of the *Cnaphalocrocis medinalis* (Lepidoptera: Pyralidae). Journal of Economic Entomology. 110:949–953. https://doi.org/10.1093/jee/tox108.

Van Lenteren JC, Bolckmans K, Kohl J, Ravensberg WJ, Urbaneja A. 2018. Biological control using invertebrates and microorganisms: plenty of new opportunities. BioControl. 63:39–59. https://doi.org/10.1007/s10526-017-9801-4.

Van Lenteren JC, Hale A, Klapwijk JN, Van Schelt J, Steinberg S. 2003. Guidelines to quality control of commercially produced natural enemies. In: Van Lenteren JC, editor. Quality control and production of biological control agents: theory and testing procedures. Wageningen, The Netherlands: CABI Publishing; pp. 265–303. https://doi.org/10.1079/9780851996882.0265.

Xu J, Yang X, Ying L, Zang L-S, Tian C-Y, Ruan C-C. 2016. Effects of fertilized, unfertilized, and UV-irradiated hosts on parasitism and suitability for *Trichogramma* parasitoids. Entomologia Experimentalis et Applicata. 161:50–56. https://doi.org/10.1111/eea.12485.

Zhang Y-H, Xue,J-Z, Tariq T, Li T-H, Qian H-Y, Cui W-H, Tian H, Monticelli LS, Desneux N, Zang L-S. 2024. Parasitism and suitability of *Trichogramma chilonis* on large eggs of two factitious hosts: *Samia cynthia ricini* and *Antheraea pernyi*. Insects. 15(1):2–11. https://doi.org/10.3390/insects15010002.

Zhang Y, Zhu W, Mei W, Fan R, Wang J, Zhang J. 2024. Parasitoid fitness and quality of *Corcyra cephalonica* (Stainton) eggs after vacuum packaging combined with cold storage. Journal of Asia-Pacific Entomology. 27(1):102207. https://doi.org/10.1016/j.aspen.2024.102207.