Variation in levels of acceptance, developmental success, and abortion of Halyomorpha halys eggs by native North American parasitoids / Costi, Elena; Wong, Warren; Cossentine, Joan; Acheampong, Susanna; Maistrello, Lara; Haye, Tim; Talamas, Elijah J.; Abram, Paul K.. - In: BIOLOGICAL CONTROL. - ISSN 1049-9644. - 151:(2020), pp. 104396-104396. [10.1016/j.biocontrol.2020.104396]

Journal Pre-proofs

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PII: S1049-9644(20)30623-X
DOI: https://doi.org/10.1016/j.biocontrol.2020.104396
Reference: YBCON 104396

To appear in: *Biological Control*

Received Date: 28 February 2020
Revised Date: 17 June 2020
Accepted Date: 29 July 2020

Please cite this article as: Costi, E., Wong, W., Cossentine, J., Acheampong, S., Maistrello, L., Haye, T., Talamas, E.J., Abram, P.K., Variation in levels of acceptance, developmental success, and abortion of *Halyomorpha halys* eggs by native North American parasitoids, *Biological Control* (2020), doi: https://doi.org/10.1016/j.biocontrol.2020.104396

Variation in levels of acceptance, developmental success, and abortion of *Halyomorpha halys* eggs by native North American parasitoids

Elena Costi[1*], Warren Wong[2,3], Joan Cossentine[4], Susanna Acheampong[5], Lara Maistrello[1], Tim Haye[6], Elijah J. Talamas[7], and Paul K. Abram[3]

[1] Dipartimento di Scienze della Vita, Università di Modena e Reggio Emilia, Via G. Amendola 2, I-42122 Reggio-Emilia, Italy
[2] Department of Biological Sciences, Simon Fraser University, 8888 University Drive, Burnaby, B.C, V5A 1S6, Canada
[3] Agriculture and Agri-Food Canada, Agassiz Research and Development Centre, 6947 Highway 7, P.O. Box 1000, Agassiz, BC V0M 1A0, Canada
[4] Agriculture and Agri-Food Canada, 4200 Highway 97 Box 5000, Summerland, British Columbia, V0H 1Z0, Canada
[5] British Columbia Ministry of Agriculture, Plant Health Unit, Kelowna, BC, V1X 7G5, Canada
[6] CABI Switzerland, Rue des Grillons 1, 2800 Delémont, Switzerland
[7] Florida Department of Agriculture and Consumer Services Division of Plant Industry, 1911 SW 34th St. Gainesville, FL 32608, USA
***Corresponding Author**
E- mail: elena.costi@unimore.it, Phone +390522523544, Fax+390522522027
ORCID: 0000-0003-4925-7133

Abstract

~~In order for native parasitoids to adapt to an unsuitable invasive host, there must be intraspecific variation in traits that could be selected upon to enable exploitation or avoidance.~~ Using a native North American parasitoid species (Hymenoptera: Scelionidae) that often unsuccessfully attack~~ing~~ the eggs of the invasive brown marmorated stink bug *Halyomorpha halys* (Hemiptera: Pentatomidae) ~~as a case study~~, we assessed ~~the~~ variation in traits that may determine the parasitoids' ~~future~~ ability to adapt to the invasive host by either exploiting or avoiding *H. halys* eggs (acceptance, developmental success). ~~The aim of surveying these~~We also assessed variation in the parasitoids' ability to induce *H. halys* host egg abortion, which may contribute to biological control of *H. halys* in invaded areas. ~~parasitoid's variations is to assess their, and their current contribution to theits biological control (ability to induce host egg abortion) of *Halyomorpha halys*.~~ The first set of experiments evaluated intra- and interspecific variation using standardized laboratory tests with iso-female lines of *Telenomus podisi* and *Trissolcus euschisti* that included matching of detailed behavioural observations of acceptance with developmental outcomes. In a second set of experiments, we assessed how variation in developmental ability and abortion induction may affect levels of biological control by indigenous parasitoid species. We examined a broader sample of parasitoids that emerged from field collections of egg masses of an indigenous north American stink bug *Podisus maculiventris* in a region newly invaded by *H. halys*. Results from the first set of experiments showed high levels of acceptance of *H. halys* eggs among iso-female lines of parasitoids, but offspring development success was almost zero. *H. halys* egg abortion due to unsuccessful parasitism was often very low and varied among iso-female lines only for *T. podisi*. In the second set of experiments we never observed increases in abortion levels of *Halyomorpha halys* eggs above natural levels, even for the two species (*T. euschisti* and *T. podisi*) that were observed to oviposit in and abort *H. halys* eggs in the first set of experiments. We conclude that while there may be some variation in behavioural and physiological parameters mediating acceptance and abortion of *H. halys* eggs by native North American egg parasitoids, there does not appear to be significant variation in developmental success. Moreover, ~~and that their~~ current biological control impact of *H. halys* eggs *via* host egg abortion is likely very low.

Keywords: Intraspecific variation, biological control, acceptance behaviour, evolutionary trap, Scelionidae, *Telenomus podisi*, *Trissolcus euschisti*

1.1 **Introduction**

The introduction of invasive species exposes native species to novel biotic selective pressures. The establishment, spread, and proliferation of invasive species in new environments can have important ecological consequences such as niche displacement, competition and in some cases, extinction of native species (Mooney and Cleland, 2001; Sax and Gaines 2008; Ekesi et al., 2009; Wilson and Holway, 2010; San Sebastián et al., 2015; Kenis et al., 2017). However, new and frequent interactions with invasive species may lead to novel behavioural responses and trophic interactions. For example, native natural enemies may begin to exploit the invasive species as a prey resource (Caroll and Dingle, 1996, Mooney and Cleland, 2001). In some cases, invasive species may initially be unsuitable resources for native natural enemies that commonly attempt to exploit them, resulting in evolutionary traps (Schlaepfer et al., 2005). Nevertheless, the adaptation of native species to successfully exploit invasive species can occur over a range of timescales. Native species can adapt either by avoiding the unsuitable invasive species or by evolving traits that increase the suitability of the invasive species as prey (reviewed in Berthon, 2015).

For ~~insect~~ egg parasitoids to adapt to an initially unsuitable invasive host (i.e., an escape from the evolutionary trap), there needs to be genetic variation in traits that mediate the parasitoid's levels of host acceptance (oviposition inside the eggs of the invasive host) or the ability of the parasitoid's offspring to develop once accepted. Selection on variation in behaviours that mediate acceptance could lead to avoidance of the unsuitable host, while selection on variation in developmental success could lead to increased host suitability (Keeler and Chew, 2008; Nelson et al., 2011; Abram et al., 2014). In addition, when an invasive species that acts as an evolutionary trap for native parasitoids is the target of biological control programs, it may be important to consider~~,~~ variation in the ability of parasitoids to kill the invasive host when they unsuccessfully attack it ("non-reproductive effects"; reviewed in Abram et al., 2019a)~~ may be important to consider~~. The mortality of the invasive pest resulting from non-reproductive effects could theoretically contribute to biological control of the invasive pest via both direct and indirect ecological effects (Kaser et al., 2018; Abram et al., 2019a; Stahl et al., 2019a). Assessing intraspecific variation in native parasitoid acceptance, developmental success, and the ability to induce host mortality can provide insight into the potential for the adaptation of native species to new invasive hosts, while also forming ~~and form~~ the basis for genetic improvement through breeding programs for native biological control agents (Lommen et al., 2017; Kruitwagen et al., 2018).

The brown marmorated stink bug, *Halyomorpha halys* (Stål) (Hemiptera: Pentatomidae), is a polyphagous insect pest native to Asia that has rapidly spread throughout North America and

Europe (Haye et al., 2015a; Leskey and Nielsen, 2018, Maistrello et al., 2018), causing economic losses in a number of different crops (Leskey et al., 2012; Maistrello et al., 2017; Bosco et al., 2018). Many studies have been conducted in invaded areas to survey for native parasitoid and predator species with the potential ~~potentially able~~ to contribute to the biological control of *H. halys* ~~biological control~~ (e.g. Ogburn et al., 2016; Dieckhoff et al., 2017; Haye et al., 2015b; Herlihy et al., 2016; Roversi et al., 2016; Cornelius et al., 2016a, b; Abram et al., 2017a; Costi et al., 2019~~8~~). Overall, egg parasitoids in the family Scelionidae (mostly species of *Telenomus* Haliday and *Trissolcus* Ashmead) are the most common group of natural enemies attacking *H. halys* in invaded areas. Several studies have found that these parasitoids readily accept *H. halys* eggs for oviposition, but their offspring rarely develop successfully (Abram et al., 2014; Haye et al., 2015b; Konopka et al., 2018, 2019, 2020). This combination of high levels of acceptance and low developmental suitability suggests that *H. halys* represents an evolutionary trap for these native scelionid parasitoids (Abram et al., 2014; Schlaepfer et al., 2005). However, a few studies have shown that between 15–25% of attacked *H. halys* eggs abort development as a result of unsuccessful ~~parasitoid~~ attack by parasitoids in invaded areas, potentially as a result of a lack of shared evolutionary history (Abram et al., 2014, 2016; Haye et al., 2015b). Recent studies using molecular diagnostic markers have confirmed that the unsuccessful attack of *H. halys* eggs by native scelionid egg parasitoids is common in nature and have shown that ~~observed~~ *H. halys* egg mortality that could be due to parasitoid-induced host egg abortion (Konopka et al., 2018; Gariepy et al., 2019). Thus, aborted host egg development induced by native parasitoids may contribute to biological control of *H. halys* in invaded areas, although the magnitude of the effect is unclear.

Some variation in the ability of indigenous parasitoids to develop in and abort *H. halys* eggs has previously been observed in a number of field and laboratory studies (Abram et al., 2014; Cornelius et al., 2016a, b; Herlihy et al., 2016; Ogburn et al., 2016; Dieckhoff et al., 2017; Konopka et al., 2019; Tognon et al., 2017, 2019; Abram et al., 2014; Haye et al., 2015b; Abram et al., 2016; Abram et al., 2017b). However, laboratory studies have either been restricted in taxonomic breadth, and within-species genetic variation (e.g., Abram et al., 2014, 2016; Konopka et al., 2018, 2020; Haye et al., 2015b), or have lacked direct behavioural observations of parasitoids and unexposed control egg masses to relate levels of acceptance to developmental success and egg abortion (e.g., Tognon et al., 2017, 2019). In field studies, because most ovipositions by native parasitoids are unsuccessful (Abram et al., 2014, 2016, 2017b), the true incidence of *H. halys* egg attack by genetically variable pools of native parasitoids and how it might explain variation in *H. halys* egg mortality is still mostly unclear (Haye et al., 2015b). Previous studies have suggested that native parasitoids might eventually adapt to be able to develop in *H. halys* eggs (Abram et al., 2014, 2016;

Tognon et al., 2017) but none have assessed whether there is~~ enough~~ intraspecific variation in relevant traits that would be needed for natural selection to proceed. Standardized laboratory studies that consider the role of intra- and inter-specific variation in acceptance, offspring development, and host egg abortion induction in field populations of parasitoids could improve understanding of the range of impacts that indigenous parasitoids may have on *H. halys* in different invaded regions, both in the short and long terms.

The goal of this study is to understand intra and interspecific variation in egg parasitoid traits that may determine their ability to adapt to exploiting or avoiding *H. halys* eggs (acceptance, developmental success), and their current contribution to its biological control (ability to induce host egg abortion). We took two approaches. First, we conducted standardized tests ~~with narrow inter-lines genetic samples~~comparing among ~~(~~iso-female lines~~)~~ of two common native North American parasitoid species, ~~-~~attacking *H. halys* that included matching of detailed behavioural observations of acceptance with developmental outcomes. Second, we screened a broader taxonomic and genetic sample of parasitoids that emerged from field collections of egg masses of an indigenous stink bug to assess how variation in developmental ability and abortion induction may affect levels of biological control by indigenous parasitoid species in a region newly invaded by *H. halys*.

1.2 Material & Methods

1.2.1 Insect colonies

The spined soldier bug, *Podisus maculiventris* (Say) (Hemiptera: Pentatomidae), a predatory species native to North America, was used to produce egg masses for rearing native parasitoids and "sentinel" egg masses to collect parasitoids in the field. The *P. maculiventris* colony, established from individuals collected in Ontario and Quebec, Canada in 2015, was reared continuously in mesh cages with access to potted fava bean plants (*Vicia faba* L.) and mealworms (*Tenebrio molitor* L.). Egg masses laid on the sides of cages and on plants were collected daily and glued to strips of filter paper with white non-toxic glue (Elmer's school glue) and offered to parasitoid females to maintain parasitoid colonies. Egg masses laid on strips of Reemay fabric (Avintiv, USA), which were provided as additional oviposition substrate, were stored for up to 4 days at 8°C before deployment as sentinel egg masses. This short period of cold storage does not affect parasitism or parasitoid offspring emergence (PKA, unpublished observations).

Trissolcus euschisti (Ashmead) (Hymenoptera: Scelionidae) ~~(native to North America)~~ and *Telenomus podisi* Ashmead (Hymenoptera: Scelionidae) ~~(native to Southeastern USA)~~ are solitary, sexually reproducing idiobiont ~~and generalist parasitoids of stink bugs species of economic~~

~~importance and~~egg parasitoids distributed throughout North America that both have broad host ranges within the Pentatomidae. T~~t~~hey are also the most common scelionids attacking eggs of the brown marmorated stink bug in North America (Abram et al., 2017b)~~.~~. We established five strains of *Tr. euschisti* (provided by Tara Gariepy, London Research and Development Centre, Agriculture and Agri-Food Canada; Jacques Brodeur and Mathilde Gaudreau, Université de Montréal) and five strains of *T. podisi* (provided by Jacques Brodeur and Mathilde Gaudreau, Université de Montréal; James Walgenbach and Emily Ogburn, North Carolina State University). Strains tested in our study ~~likely~~ represent a small sample of the range present in North America, as our collections are based on a small number of localities. However, our goal was not to be comprehensive. Rather, we aimed to conduct a preliminary assessment of whether there could be inter-line variation in the parameters we measured. Single females from each collection were used to establish an iso-female line~~s~~ for each strain, which were reared in climate-controlled rooms for at least 5 generations of sib-mating before testing. Using this iso-female line approach, any phenotypic differences among lines should be due mostly to genetic differences (Beukeboom and Zwaan, 2007), although we cannot exclude the possibility that other differences such as microbiomes (e.g. facultative or obligate microbial endosymbionts), long-term rearing effects, or maternal effects could also be a factor, especially because detailed genetic analyses of these lines has not been performed. Table 1 shows the parasitoid lines tested and their origins. Fresh (<24h since laying) *P. maculiventris* eggs were offered to the parasitoids on a regular basis for colony maintenance. To collect parasitoid females for use in experiments, parasitized egg masses were kept in 1.5 mL plastic microtubes and observed daily for newly emerged female parasitoids. Females were always paired with males from the same strain.

Halyomorpha halys individuals were originally collected from two locations in Chilliwack, British Columbia, in 2016 and were reared thereafter at the Agassiz Research and Development Center (BC). Stink bugs were maintained in mesh cages provided with potted pepper (*Capsicum annum* L.) plants and store-bought corn and carrots as food sources. Eggs laid by females on Reemay fabric, plant leaves, paper towels, and the sides of the cages were collected daily for laboratory experiments.

All insect colonies were maintained at 23 ± 2°C, 40-60% relative humidity, and a photoperiod of 16L:8D.

1.2.2 Intraspecific variation in acceptance and development

The goal of the first experiment was to compare the level, latency, and rate of behavioural acceptance of *H. halys* eggs among strains of *T. euschisti* and *T. podisi*. Prior to use in experiments, newly emerged parasitoids from each species and strain were isolated daily in 1.2 mL Eppendorf tubes with a drop of honey water (70% honey and 30% water) for food, kept with a male for 24 hours to allow mating, and then kept alone for one week to allow time for ovariole maturation. For each trial, individual female parasitoids were transferred to small petri dishes (diameter: 5.0 cm, height: 1.0 cm) where a fresh *H. halys* egg mass containing 28 eggs was secured to the bottom of the dish with a small amount of white, non-toxic glue. The trials were recorded for 5 h using a mini-button CCD camera connected to a digital microscope (Dino-Lite, London, Ontario, Canada). Each treatment conducted in a climatic chamber (25°C and 16L:8D) was replicated between 17 and 22 times for each strain of each parasitoid species. The experiment was performed in 42 testing days over a total period of 129 days. For each set of egg masses exposed to parasitoids (i.e. on each day of the experiment), unexposed egg masses (controls) were subjected to the same conditions as the treatment egg masses except that they were not exposed to a parasitoid. These unexposed egg masses (n=85) allowed the measurement of baseline levels of *H. halys* egg abortion that was not due to parasitoids (see Abram et al., 2016).

After the recording period, parasitoids were removed, and both exposed and unexposed egg masses were maintained under standard rearing conditions for at least 30 days, observing them daily to record the number of host nymphs and parasitoids emerging. After 30 days, any unhatched eggs were dissected and classified as: (i) emerged or fully developed but unemerged *H. halys* nymphs or (ii) aborted, containing only undifferentiated liquid contents. The dissections yielded no fully developed parasitoid adults that failed to emerge.

When analyzing each video (n=198), a single observer recorded the occurrence and timing of ovipositions, indicated by the parasitoid inserting its ovipositor into a host egg followed by marking behaviour (see Abram et al., 2014). This allowed us to determine the level of acceptance of the host eggs (the total number of eggs accepted), the latency to the first successful oviposition, and the total time period over which parasitism took place (time between the first and last ovipositions). Self-superparasitism was relatively uncommon in our trials (5.2% of all ovipositions for *T. euschisti*; 6.8% of all ovipositions for *T. podisi*), and the total number of ovipositions was almost perfectly correlated with the number of eggs accepted for both *T. euschisti* (r = 0.99) and *T. podisi* (r = 0.99). Thus, superparasitism events were disregarded in the analysis, and the number of eggs in which at least one oviposition was observed was used as the metric of behavioural acceptance.

We expected the level of acceptance to indicate the readiness of native parasitoids to oviposit in *H. halys* eggs. We anticipated that the time until the first oviposition occurred could be a proxy for the time necessary for native parasitoids to recognise the eggs of the host and accept it as a resource. Finally, we expected acceptance rate, calculated as the total time to complete parasitism of the *H. halys* egg mass divided by the number of eggs attacked, to measure the efficiency with which the different strains of the two species of parasitoids exploited *H. halys* eggs. Moreover, the acceptance rate would be driven by other behaviours that decrease this efficiency, such as increased time required to drill into eggs, host rejections, superparasitism, and short patch leaving bouts (Field 1998).

Next, we determined the level of *H. halys* egg abortion associated with unsuccessful ovipositions by comparing the number of host eggs the parasitoids oviposited into and the developmental outcomes of eggs. The number of eggs aborted due to oviposition in each replicate was calculated with the following formula (see also Abram et al. 2016):

$$\text{eggs aborted due to oviposition} = AbE - \left(\frac{\text{AbC} * \text{TotE}}{\text{TotC}}\right) \qquad \text{(eqn. 1)}$$

Where AbE and AbC are, respectively, the number of aborted eggs observed in the exposed and control egg masses, and TotE and TotC are the total number of eggs in the exposed and control, respectively. Abortion levels in each exposed egg mass (AbE and AbC) were adjusted using pooled values of TotE and TotC from paired egg masses set up on the same date.

Two factors contribute to the numbers of aborted *H. halys* eggs among parasitoid strains. The first is variation in the number of eggs attacked; the second is variation in the per-attack probability of aborting host development. To investigate whether parasitoid strains differed with respect to the per-attack probability of inducing *H. halys* egg abortion, we applied the following formula to calculate abortion efficiency, calculating the number of eggs aborting due to oviposition (for eggs accepted by parasitoids only), and then dividing it by the total number of accepted eggs,:~~first calculated the corrected number of eggs aborted in each replicate (eqn. 1), considering only accepted eggs.~~

$$\text{Abortion efficiency} = \frac{AbE_A \cancel{AbE} - \left(\frac{\text{AbC} * \text{TotE}_A \cancel{\text{TotE}}}{\text{TotC}}\right)}{\text{TotE}_A \cancel{\text{TotA}}}$$

Where AbE_A and TotE_A are the number of aborted eggs accepted by parasitoids, and the total number of eggs accepted by parasitoids, respectively.

~~First, we calculated the corrected number of eggs aborted in each replicate (eqn. 1), considering only accepted eggs We then we divided this by the total number of eggs accepted by the parasitoid (TotA), giving the corrected number of aborted eggs per host accepted, which we term “abortion efficiency”~~..

1.2.3 Laboratory tests of field-collected parasitoids on H. halys eggs

To complement detailed laboratory estimates of parasitoid developmental success and abortion induction with narrow ~~inter-linesgeneti~~inter-line comparisons~~e samples~~ within~~of~~ two species, we investigated variation in these parameters with a larger number of parasitoid species. We assumed that these encompassed a broader genetic diversity, because individuals were collected from sites across a relatively wide geographic region, although we did not directly assess the true amount of genetic variation present with molecular tools. The goal of these experiments was to explore to what extent the evolutionary trap posed by *H. halys* for native parasitoids generally applies across species present in British Columbia, and to what degree egg abortion caused by native parasitoids may be contributing to biological control of *H. halys*.

We used sentinel egg mass surveys, deploying eggs of the native species *P. maculiventris*, to investigate the diversity of egg parasitoids of stink bugs present in British Columbia and obtain parasitoids for laboratory screening against *H. halys*. From the beginning of May to the end of August of 2017, 1,264 *P. maculiventris* egg masses (21,310 eggs) were set out and retrieved at 10 sites in coastal (Fraser Valley) and interior (Okanagan Valley) British Columbia (Table 2), on between 2 and 8 different dates per site through the sampling period, on an opportunistic basis. All sites were semi-urban, urban, or suburban areas with mixed woody and herbaceous vegetation where breeding populations of *H. halys* had previously been detected (Abram et al., 2017a~~8~~; Abram et al., 2019b). Fabric squares containing egg masses (see *Insect Colonies* above) were stapled to the undersides of leaves of a variety of woody trees (e.g., *Prunus* spp., *Davidia* spp., *Acer* spp., *Gleditsia* spp., *Ailanthus* spp., *Sorbus* spp.) and shrubs (e.g., *Rubus* sp., *Mahonia* spp., *Symphoricarpos* spp., *Rosa* spp.). The egg masses were then retrieved before nymphs emerged, 3–7 days after being set out. Because of the variable retrieval intervals, parasitism rates measured here are intended only to determine which parasitoid species were present and to serve as sources of parasitoids for laboratory tests.

Retrieved egg masses were placed in Petri dishes (50 mm diameter, 9 mm depth) and kept under ambient laboratory conditions until parasitoid emergence. Emerging parasitoids were given cotton wicks with 10% sucrose as a food source. Newly emerged parasitoids from the Okanagan Valley collections were shipped overnight in a cooler with an ice pack (temperature ~8–10°C) to the Agassiz Research and Development Centre (Agassiz, BC) where samples from the Fraser Valley were located, and where all subsequent tests took place in the laboratory. Emerging parasitoids from ~~a subset of 139~~ parasitized egg masses were separated into 1.5 ml plastic Eppendorf tubes at least one day before they were tested. A subset of females was tested (n = 139), with the subset determined by the availability of fresh *H. halys* eggs when parasitoids were of the correct age for testing (see Table 2 and Figure 4 for per-species sample sizes). All females used in tests emerged in the presence of males from the same egg mass and were thus assumed mated. In all cases, parasitoids were 7–14 days old when used in these tests, enough time for post-emergence ovariole maturation to take place but well before the end of their typical lifespan, which ~~is~~ under~~in~~ ~~the~~ laboratory conditions is ~~(~~greater than ~~>~~40 days~~)~~ (see Abram et al., 2016). ~~For each parasitoid species a~~At least 5 replicates were conducted for each parasitoid species. Individual females were transferred from plastic tubes to Petri dish arenas (50 mm diameter, 9 mm depth) containing an *H. halys* egg mass of between 6 and 32 eggs (mean ± SE: 20.6 ± 0.52, n=191). The size of the egg masses used depended on the availability of fresh *H. halys* eggs on each testing day; egg masses were divided into smaller patches when availability of host material was limited~~, depending on the relative availability of *H. halys* eggs and adequately aged parasitoids~~. However, we accounted for variation egg mass size by including it as a covariate in the statistical analysis (see below). In all replicates, parasitoids were removed from dishes after 24 h. Egg masses were then incubated under standard rearing conditions and monitored for the emergence of host nymphs and parasitoids. After being incubated for at least 1 month, they were dissected to record unemerged parasitoids and aborted eggs, using the same classification criteria as the first experiment (see above). Throughout the period of the tests, a set of similarly-sized egg masses (24.3 ± 1.0 eggs, n=29) were set up in Petri dish arenas but not exposed to parasitoids as controls, inclubated, and then dissected to record levels of nymph emergence and egg abortion. These trials were run concurrently with the iso-female line experiments described above, under the same abiotic conditions and using *H. halys* egg masses of the same age from the same laboratory colony.

All parasitoids emerging from sentinel egg masses, including those used in laboratory tests with *H. halys* eggs, were subsequently preserved in 95% EtOH, point-mounted, and identified based on morphological characters (Talamas et al., 2015). Vouchers are deposited in the Florida State Collection of Arthropods ~~and Nematodes~~ and the Canadian National Collection of Insects, Arachnids, and Nematodes.

1.2.4 Statistical analysis

For laboratory trials where *H. halys* eggs were exposed to different iso-female lines of *T. podisi* and *T. euschisti,* the number of eggs accepted, acceptance latency, acceptance rate, the corrected number of eggs aborted, and abortion efficiency were compared among iso-female lines with Kruskal-Wallis tests followed by Dunn's Kruskal-Wallis multiple comparisons with Benjamini-Hochberg adjustments. These non-parametric tests were selected after finding that the response variables did not meet the assumptions of generalized linear models (with either G~~g~~aussian or P~~p~~oisson error distributions) in most cases. To test whether exposure to parasitoids from each iso-female line of the two species was associated with higher-than-baseline *H. halys* egg mortality, the corrected number of eggs aborted when exposed to each iso-female line was compared to zero with a Wilcoxon test.

The statistical approach was modified for the experiment measuring levels of egg abortion in *H. halys* eggs exposed to parasitoids emerging from field-collected *P. maculiventris* egg masses. First, because egg mass size varied among replicates, the proportion (rather than number) of aborted eggs was used as the response variable, and egg mass size (number of eggs) was included as a covariate in the analysis. Second, because multiple parasitoids were sometimes tested from the same sites and natal egg mass (which could result in pseudoreplication if not accounted for), "natal egg mass" and "collection site" were included in the statistical model as random effects. Third, because paired unexposed control egg masses could not be run for ~~each~~ every exposed egg mass due to limited *H. halys* egg availability on some days, the global level of abortion in unexposed egg masses set up over the course of the experiment was compared to levels of abortion in egg masses exposed to each parasitoid species. Thus, the proportion of *H. halys* ~~egg~~ eggs that aborted when exposed to different species of parasitoids or were not exposed to parasitoids, was compared with a generalized linear mixed model (GLMM) with a binomial error distribution, with parasitoid species and egg mass size as fixed effects and natal egg mass as a random effect. Observation-level random effects (Harrison, 2014) were also included to account for overdispersion.

All statistical analyses were analyzed using R version 3.6.0 (R Development Core Team, 2019).

1.3 Results

1.3.1 Intraspecific variation in acceptance and development

All iso-female lines of both *T. euschisti* and *T. podisi* oviposited in *H. halys* eggs, but the number of *H. halys* eggs in which parasitoids oviposited varied significantly among lines for both *T. euschisti* ($\chi^2_4 = 26.19$, $p < 0.0001$) and *T. podisi* ($\chi^2_4 = 29.32$, $p < 0.0001$), with the median number of eggs accepted ranging from 15 to 27 for *T. euschisti* and 7 to 28 eggs for *T. podisi*, out of the 28 *H. halys* eggs offered (Figure 1). Median acceptance latency by *T. podisi* ranged between 0.34 and 1.68 hours but did not differ among lines (χ^2_4 = ~~26.19~~7.42, p = 0.11~~< 0.0001~~) (Figure 2). However, acceptance latency did vary significantly among lines for *T. euschisti* ($\chi^2_4 = 10.80$, $p = 0.029$), with median time until the first observed oviposition ranging from 0.09 to 0.67 hours (Figure 2). The oviposition rate did not clearly differ among the lines of *T. podisi* ($\chi^2_4 = 8.00$, $p = 0.092$; medians

7.8 to 12.0 ovipositions/hour) or *T. euschisti* ($\chi^2_4 = 8.81$, p = 0.066; medians 5.6 to 10.0 ovipositions/hour).

There was no evidence in variation in the developmental success of parasitoid offspring among iso-female lines of either parasitoid species. In fact, only a single parasitoid offspring, a *T. euschisti* (line Te_3) emerged from one of the parasitized *H. halys* egg masses, despite a total of 1,907 eggs being oviposited in by *T. euschisti* across all replicates. We never observed the emergence of *T. podisi* offspring from *H. halys* eggs, which were observed ovipositing in a total of 1,588 *H. halys* eggs across all replicates.

After being corrected for natural levels of *H. halys* egg mortality, the number of eggs aborted by *T. podisi* varied among iso-female lines ($\chi^2_4 = 12.22$, p = 0.016), with all lines causing at least some *H. halys* egg abortion; median corrected numbers of aborted eggs ranged from 0.5–4.0 (Figure 3). These differences in *H. halys* egg abortion induction among lines were maintained when accounting for the number of ovipositions; abortion efficiency also varied among lines of *T. podisi* ($\chi^2_4 = 18.11$, p = 0.0012) (Figure 3), with per-strain medians of between 1.7% and 40.6% of attacked eggs aborting. For *T. euschisti*, numbers of aborted eggs did not vary among lines ($\chi^2_4 = 2.67$, p = 0.61), and medians were generally lower (range: 0.5–1.0 eggs), with only three out of five lines causing abortion levels significantly above zero (Figure 3). Likewise, abortion efficiency did not vary significantly among the lines of *T. euschisti* ($\chi^2_4 = 3.43$, p = 0.49), with an intraspecific mean of 6.0% of attacked *H. halys* eggs aborting.

1.3.2 Laboratory tests of field-collected parasitoids on H. halys eggs

Out of the total of 1,264 *P. maculiventris* sentinel egg masses deployed in the field, 15.3% were parasitized by scelionid egg parasitoids (Table 2). The percentage of egg masses with parasitoid emergence varied among sites from 8.4–24.0%. Of the five scelionid species emerging from sentinel eggs, *T. euschisti* was present at the greatest number of sites (10/10 sites), followed by *Te. podisi* (9/10 sites), *Trissolcus~~.~~ cosmopeplae* (Gahan) (3/10 sites), *Tr.~~.~~ utahensis* (Ashmead) (2/10 sites), and *Tr. hullensis* Harrington (1/10 sites). The latter three *Trissolcus* spp. were only found at sites in the Okanagan Valley; *T. euschisti* and *T. podisi* were found in both the Fraser Valley and Okanagan Valley regions. The only non-scelionid parasitoid reared from *P. maculiventris* eggs was a single individual in the family Encyrtidae (Table 2).

A total of 191 parasitoids, representing the five scelionid species emerging from *P. maculiventris* eggs, were tested on *H. halys* eggs in the laboratory. Oviposition was commonly observed at the beginning of these trials by all species tested, and post-oviposition brood guarding was often observed afterwards (P. Abram, personal observations). These informal observations, together with the direct observations on two of these species (*T. podisi* and *T. euschisti*) in the iso-female lines experiment under similar conditions, suggest that host acceptance occurred in many of these trials. However, of the 3,935 eggs exposed to these parasitoids, only a single *T. euschisti* individual emerged from an *H. halys* egg.

Levels of *H. halys* egg abortion in these experiments were very low, often with a median of zero (Figure 4). The proportion of *H. halys* egg abortion was not significantly higher in eggs exposed to any of the parasitoid species than in unexposed egg masses and did not vary significantly among parasitoid species to which they were exposed (GLMM, $\chi^2_{4,81}$= 2.92, p= 0.57) (Figure 4). Levels of egg abortion in exposed egg masses were not associated with variation in *H. halys* egg mass size ($\chi^2_{1,81}$= 0.013, p= 0.91) or its interaction with exposure treatment ($\chi^2_{4,81}$ = 2.60, p= 0.63).

1.4 Discussion

Our study adds to a growing body of evidence from laboratory and field studies indicating that native scelionid parasitoids in North America readily oviposit in the eggs of *H. halys* but rarely develop successfully (Abram et al., 2014; Haye et al., 2015b; Ogburn et al., 2016; Cornelius et al., 2016a, b; Dieckhoff et al., 2017; Abram et al., 2017b; Gariepy et al., 2019; Konopka et al., 2018, 2019, 2020). In fact, under our standardized laboratory conditions, the developmental success of parasitoid offspring occurred only twice out of the thousands of *H. halys* eggs that we exposed to parasitoids. In addition, we provide the first evidence that, in at least two native parasitoid species, behavioural parameters related to acceptance of *H. halys* eggs can vary intraspecifically. Finally, our results suggest that the biological control effects of parasitoid-induced abortion of *H. halys* eggs, when considering a broader range of native parasitoid genotypes than tested in past studies, is minimal. These findings have implications for the short-term biological control impact of native parasitoids on *H. halys* as well as their longer-term potential to adapt to this invasive host.

Several studies have suggested that native North American parasitoids may eventually adapt to be able to develop in *H. halys* eggs (Abram et al., 2014; Konopka et al., 2018; Tognon et al., 2019), and there are other examples where native natural enemies have eventually adapted to successfully exploit invasive prey or hosts (Keeler and Chew, 2008; Berthon et al., 2015). However, in order for natural selection to promote this shift in physiological host range, there would need to be genetic variation present in populations of native parasitoids for traits that influence their ability to successfully complete development in *H. halys* eggs. The low levels of emergence of native parasitoids, including *T. podisi* and *T. euschisti,* observed in past field studies using sentinel *H. halys* egg masses (e.g. Dieckhoff et al., 2017; Ogburn et al., 2016; Herlihy et al., 2016; Cornelius et al., 2016a, b), including in the area where parasitoids were collected for our second experiment (Abram et al., 2019b), indicates that this variation may exist. However, it is also possible that other abiotic and biotic factors in the field (e.g., unfertilized eggs, extreme temperatures, desiccation) could kill *H. halys* eggs at variable levels, making them more suitable for parasitoid development (similar to freeze-killing; e.g. Haye et al., 2015b), which would overestimate the ability of native parasitoids to develop in viable *H. halys* eggs. In addition, what proportion of *H. halys* eggs are located and unsuccessfully parasitized by native parasitoids in the field is unknown unless molecular diagnostic tools are used (Gariepy et al., 2019; Stahl et al., 2019b). Thus, to properly assess intraspecific variation in developmental success, standardized laboratory studies that observe parasitoid behaviour~~, and using *H. halys* eggs with low levels of baseline mortality, ~~are needed in conjunction with *H. halys* eggs with low levels of baseline mortality. In the current study, we found no evidence to suggest that there is significant intraspecific variation in the ability of *T. podisi*, *T.*

euschisti, or any of the other scelionid species we tested, to complete development in viable *H. halys* eggs. Across both experiments, only two individuals of *T. euschisti* emerged. It is, of course, possible that the hypothesized variation in developmental success exists in some populations of North American scelionids; our study did not include any such strains. ~~E~~However, even if initially very rare, after several years of strong selection, parasitoid ~~these~~ variants capable of developing in *H. halys*~~s~~ eggs would be expected to spread rapidly in areas where *H. halys* is common. ~~But also, in terms of variation, it's possible that some traits of an appropriate strains may needs different years of strong selections before showing their adaptation or avoidance to the target host.~~ ~~Even if initially rare, these variants would be expected to spread rapidly in areas where *H. halys* is common.~~ ~~However,~~ D~~d~~uring the period of 2013–2016, when our parasitoid lines were established, there is no evidence to suggest that there is significant intraspecific phenotypic variation in the ability of native North American scelionid parasitoids to consistently develop in viable *H. halys* eggs. However, the data and methodology presented here provide a basis for assessing how this situation could change in the future, and when considering more comprehensive samples of parasitoid strains present in areas invaded by *H. halys*.

There may be a significant fitness penalty for native parasitoids when they expend energy, time, and eggs while unsuccessfully exploiting *H. halys* eggs (Abram et al., 2014). Thus, natural selection could act on variation in parasitoid behaviours that mediate acceptance of *H. halys* eggs, leading to avoidance of the evolutionary trap by native parasitoids. We did observe variation in acceptance levels, measured as the number of eggs parasitized in 5 h, of both *T. podisi* and *T. euschisti*. Likewise, we observed significant variation in the time it took different iso-female lines of *T~~r~~. euschisti*, but not *T. podisi*, to begin ovipositing in *H. halys* eggs. We did not observe any variation in the speed at which either parasitoid species oviposited in *H. halys* eggs once parasitism began. On the one hand, these results could provide preliminary evidence that intraspecific variation exists in behaviours or physiological variables that determine acceptance of *H. halys* eggs by native parasitoids. For example, greater latency to the first oviposition could indicate that native parasitoids are less able to locate *H. halys* egg masses at a short range or are more likely to delay oviposition once the egg mass is encountered. Likewise, fewer total ovipositions by females from some iso-female lines of the two species could indicate that some strains had a greater tendency to leave *H. halys* egg masses before fully exploiting them. On the other hand, the variation in acceptance and oviposition latency we observed could be underpinned by variation in other aspects of parasitoid behaviour or physiology that are unlikely to be acted upon by natural selection to reduce the level of exploitation of *H. halys* by native parasitoids. For example, some iso-female lines may simply have lower fecundity than others leading to fewer ovipositions before their egg load is exhausted; or lower locomotor activity leading to a delay in the amount of time required to find and begin ovipositing in *H. halys* eggs. We would not anticipate natural selection to act on these types of traits specifically to avoid the evolutionary trap posed by *H. halys*, as they would also affect their ability to exploit more suitable native host species. Any eventual behavioural avoidance or rejection of *H. halys* eggs by native parasitoids would need to be mediated by modified sensitivity to specific semiochemical, visual, or physical cues that are specific to *H. halys* eggs. Future studies that examine intraspecific variation in parasitoid behaviour throughout the process of host searching are needed to better understand the potential for the evolution of behavioural avoidance of *H. halys* eggs. In particular, experiments comparing the behaviour of different strains on a suitable, native host species in addition to *H. halys* (e.g., Abram et al. 2014) could help to determine whether behavioural variation of parasitoids in response to *H. halys* is due to specific parasitoid avoidance or increased acceptance of *H. halys* specifically, *versus* non-specific parasitoid life history variation.

Although almost none of the oviposition events by *T. podisi* and *T. euschisti* we observed in this study resulted in offspring production, some of them caused increases in the number of aborted *H. halys* eggs. In our first experiment, all five iso-female lines o~~i~~f *T. podisi* (1.7–~~-~~40.6% of attacked eggs aborted) and three out of five lines of *T. euschisti* (7.4 – 10.0% of attacked eggs aborted) caused a significant number of *H. halys* eggs to abort above natural levels (Figure 3). Thus, the unsuccessful attack of *H. halys* eggs by *T. podisi* and *T. euschisti* in some invaded areas may be ~~contributing to~~ increasing ~~its~~ egg mortality. Past studies using a single laboratory strain of *T. podisi* have recorded increased abortion levels of between approximately 24 and 30% (Abram et al., 2014; Abram et al., 2016) in parasitized eggs, while one other study on two native European *Trissolcus* spp. estimated abortion rates of about 10-15% on unsuccessfully attacked *H. halys* eggs (Haye et al., 2015b). Abortion levels observed in past studies are thus within the range measured in our first experiment. Additionally, our results demonstrate that abortion levels vary between species, and, in some cases, within species. The number of *H. halys* eggs aborted, as well as abortion efficiency, varied among our iso-female lines of *T. podisi*. In contrast, there was no clear intraspecific variation in *H. halys* egg abortion caused by *T. euschisti*, with only three out of five lines causing at least some *H. halys* eggs to abort and two lines causing no abortion whatsoever. In combination with past studies, the results of our first experiment imply that for *H. halys*, the biological control effect of egg abortion by native parasitoids is likely to vary widely in time and space depending not only on parasitoid species composition but also on the relative frequency of different intraspecific variants causing different levels of egg abortion.

Although some parasitoid lines we tested aborted a significant number of *H. halys* eggs, after taking into account variation in acceptance levels and intraspecific variation in abortion efficiency, the absolute numbers of eggs aborted were low overall. While the most efficient strain of *T. podisi* aborted, on average, about 40% of attacked *H. halys* eggs (= 11.3 eggs per completely exploited egg mass of 28 eggs) (Figure 3), many egg masses were not completely exploited (Figure 1), and it was more typical for *T. podisi* and *T. euschisti* to abort fewer than four eggs per exposed egg mass (Figure 3). Considering that less than 50% of *H. halys* egg masses are located and oviposited within by native parasitoids in the field (Gariepy et al., 2019; Konopka et al., 2018), our results indicate that the biological control effect of egg abortion induced by native parasitoids on *H. halys* in North America is probably limited or even negligible. In fact, in our second set of experiments testing the capacity of native parasitoids emerging from sentinel egg masses of *P. maculiventris* in British Columbia, we never observed increases in abortion levels of *H. halys* eggs above natural levels – even for the two species (*T. euschisti* and *T. podisi*) that were observed to oviposit in and abort *H. halys* eggs in the first laboratory experiment. This result could indicate that the populations we sampled in British Columbia are composed of strains of native parasitoids that are ineffective at aborting *H. halys* eggs. Because we did not conduct detailed, comprehensive behavioural observations in this experiment, however, we cannot rule out that acceptance levels of *H. halys* eggs may have been lower than in the first experiment, and that this may be responsible for the low abortion levels observed. This would be unexpected, however, given that (i) qualitative behavioural observations (oviposition, patch guarding) indicated frequent acceptance; and (ii) the exposure period in the second experiment (24h) was almost five times longer than the first experiment (5h), giving parasitoids much more time to exploit the available eggs under similar abiotic conditions. In any case, our results suggest that native scelionid parasitoids are probably not currently contributing significantly to biological control of *H. halys* in newly invaded areas of British Columbia, either through host egg abortion or offspring development.

The susceptibility of *H. halys* to parasitism and egg abortion would depend not only on intraspecific variation in the attacking parasitoid but also potentially on phenotypic variation in hosts that mediates their susceptibility ~~(~~(Kraaijeveld et al., 1998)~~Kraaijeveld and Godfray, 1999)~~. There is ~~some~~ evidence for genetic differences among geographic populations of *H. halys*, including between e~~E~~astern and w~~W~~estern North America (Gariepy et al., 2014; Valentin et al., 2017; Abram et al., 2017a~~8~~; Cesari et al., 2017), so it is possible that there are differences in the susceptibility of these different populations to parasitoid attack. Variation in host susceptibility is unlikely to be responsible for the significant differences in the traits we measured among lines of parasitoids, as host egg masses were randomly assigned among iso-female line treatments, and experiments were run ~~in parallel~~during the same time period. However, it is possible that some differences between our study (which used w~~W~~estern North American *H. halys*) and past studies (Abram et al. 2014, 2016, which used e~~E~~astern North American *H. halys*), could be due to the different population of hosts used. The fact that one of the *T. podisi* strains used in our study (Tp_1) is the same as those used in Abram et al. (2014, 2016) allows an informal comparison. In both studies, there was no successful -development of this strain in *H. halys* eggs. In the current study, this strain aborted an average of 12% of attacked eggs, which is considerably lower than the 24-30% estimates from past studies. As most aspects of the methodology of the two studies were similar, this comparison could indicate a difference in the susceptibility of w~~W~~estern North American *H. halys* to abortion by native parasitoids. However, as some parts of the methodology differed between the two studies – most importantly egg mass size – further research that includes direct comparisons is needed to investigate this matter.

Adaptation of native parasitoids to successfully recognize and successfully exploit invasive hosts is typically considered to occur on timescales of multiple decades (Cornell and Hawkins, 1993). Given our results as well as those of past studies (e.g., Dieckhoff et al., 2017), it appears that for native scelionid parasitoids exploiting *H. halys* in North America, the variation in traits required for this adaptation is not yet clearly present, as w~~W~~e found no evidence of variation in the developmental success of these ~~native~~ parasitoids.. Our results do provide some evidence to suggest that variation in behaviours mediating acceptance of *H. halys* may exist, although it is unclear whether avoidance of *H. halys* eggs by these parasitoids could feasibly be selected for given this variation. ~~We found no evidence of variation in the developmental success of the native parasitoids.~~ Determining whether these native parasitoids will adapt to the presence of *H. halys* by avoiding its eggs, or alternatively by gaining the capacity to exploit them successfully, ~~will require~~could include long-term studies in several areas invaded by *H. halys*, as well as studies of parasitoid behaviour and development across gradients of time since the invasive stink bug has established.~~.~~

Credit authorship contribution statement

Elena Costi: Validation, Conceptualization, Data curation, Formal analysis, Writing - Original Draft, Writing - Review & Editing

Warren Wong: Validation, Data curation, Writing - Review & Editing

Joan Cossentine: Review & Editing, Supervision, Resources, Funding acquisition

Susanna Acheampong: Review & Editing, Supervision, Resources, Funding acquisition

Tim Haye: Writing - Review & Editing, Supervision, Funding acquisition

Elijah J. Talamas: Writing - Review & Editing, Data curation, Funding acquisition

Lara Maistrello: Writing - Review & Editing, Supervision, Funding acquisition

Paul Abram: Conceptualization, Formal analysis, Resources, Writing - Original Draft, Writing - Review & Editing, Visualization, Project administration

Acknowledgements

We thank Peggy Clarke, Jason Thiessen, Jessica Leung, Nemo DeJong, Mairi Robertson, Jesse McDonald, David Nield, Laura Keery, Gioia de Iulis, Johanna Schwartz, Kathryn Ryan and Emma Gaudreault for technical assistance in the laboratory and field. We owe special thanks to Tara Gariepy, Emily Ogburn, Josée Doyon, Kim Hoelmer, Kathy Tatman, Mathilde Gaudreau, Jacques Brodeur, and Jana Lee for collection and shipment of parasitoids used to attempt to establish laboratory lines.

Funding

This research was funded by Agriculture and Agri-Food Canada through A-BASE #1526 and "new scientist" funding to Paul Abram. Funding to Susanna Acheampong was through the Governments of Canada and British Columbia through Growing Forward 2, a federal-provincial-territorial initiative. The work by CABI (Tim Haye) was conducted with core financial support from CABI's member countries (see http://www.cabi.org/about-cabi/who-we-work-with/key-donors/). Funding for Elena Costi and Lara Maistrello was provided by the Emilia Romagna region (Italy) within the Rural Development Plan 2014-2020 Op. 16.1.01 – GO PEI-Agri - FA 4B, Pr. "Halys", coordinated by CRPV. Elijah Talamas was supported in part by the Florida Department of Agriculture and Consumer Services- Division of Plant Industry and USDA Farm Bill: Identification, monitoring, and redistribution of *Trissolcus japonicus*– Biological Control of Brown Marmorated Stink Bug (BMSB).

Conflict of Interest

All authors declare that they have no conflict of interest.

Informed consent

Informed consent was obtained from all individual participants included in the study

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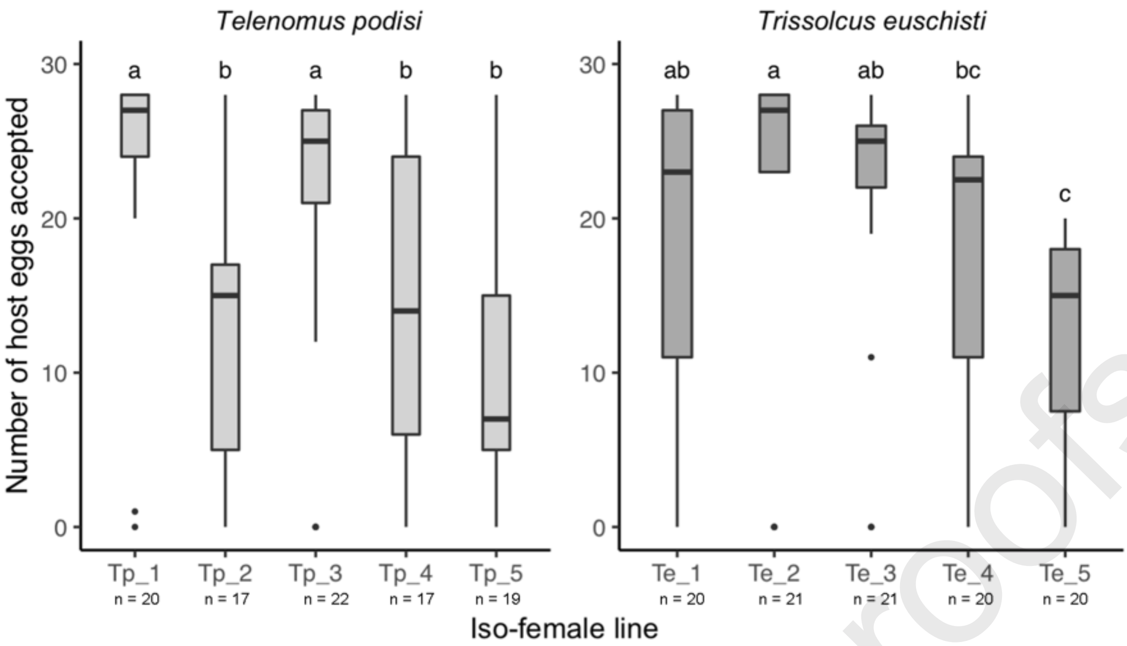


Figure 1. The number of *H. halys* eggs in which different iso-female lines of *Trissolcus euschisti* and *Telenomus podisi* oviposited when observed on *H. halys* egg masses of 28 eggs for 5 h. For lines within each parasitoid species, boxes not labeled with the same letter are significantly different ($p < 0.05$; Dunn's Kruskal-Wallis multiple comparison test). In boxplots, horizontal lines show medians, boxes contain the 25th-75th percentiles, whiskers show the upper and lower deciles, and points show outliers.

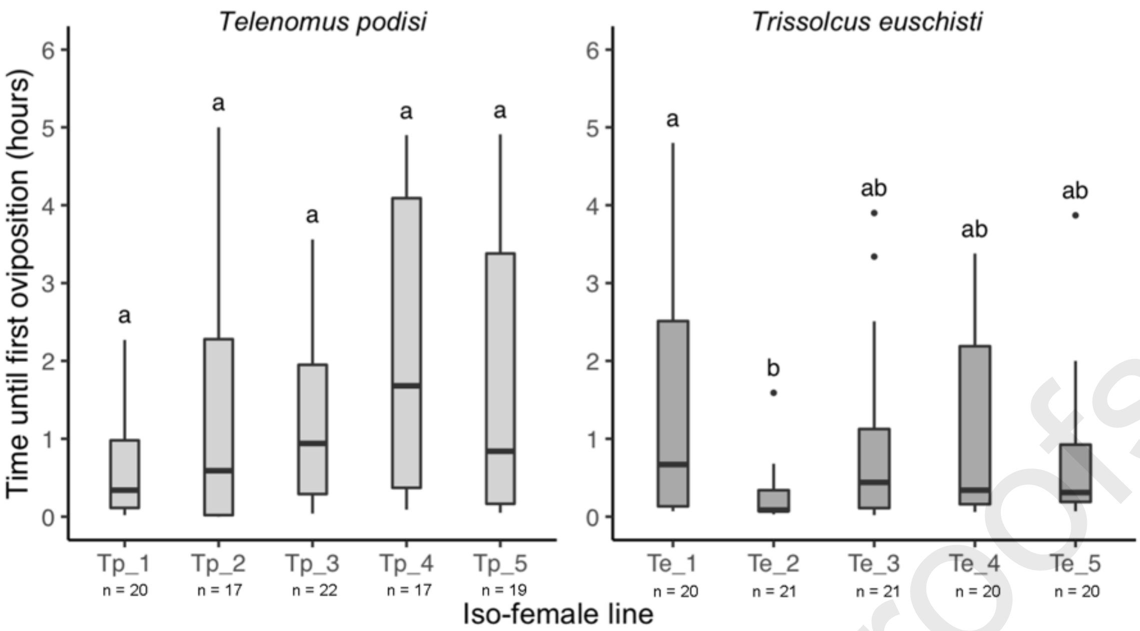


Figure 2. Acceptance latency (time until first oviposition) of different iso-female lines of *Trissolcus euschisti* and *Telenomus podisi* on *H. halys* egg masses. For parasitoid lines within each species, boxes not containing the same letter are significantly different ($p < 0.05$; Dunn's Kruskal-Wallis multiple comparison test). In boxplots, horizontal lines show medians, boxes contain the 25th-75th percentiles, whiskers show the upper and lower deciles, and points show outliers.

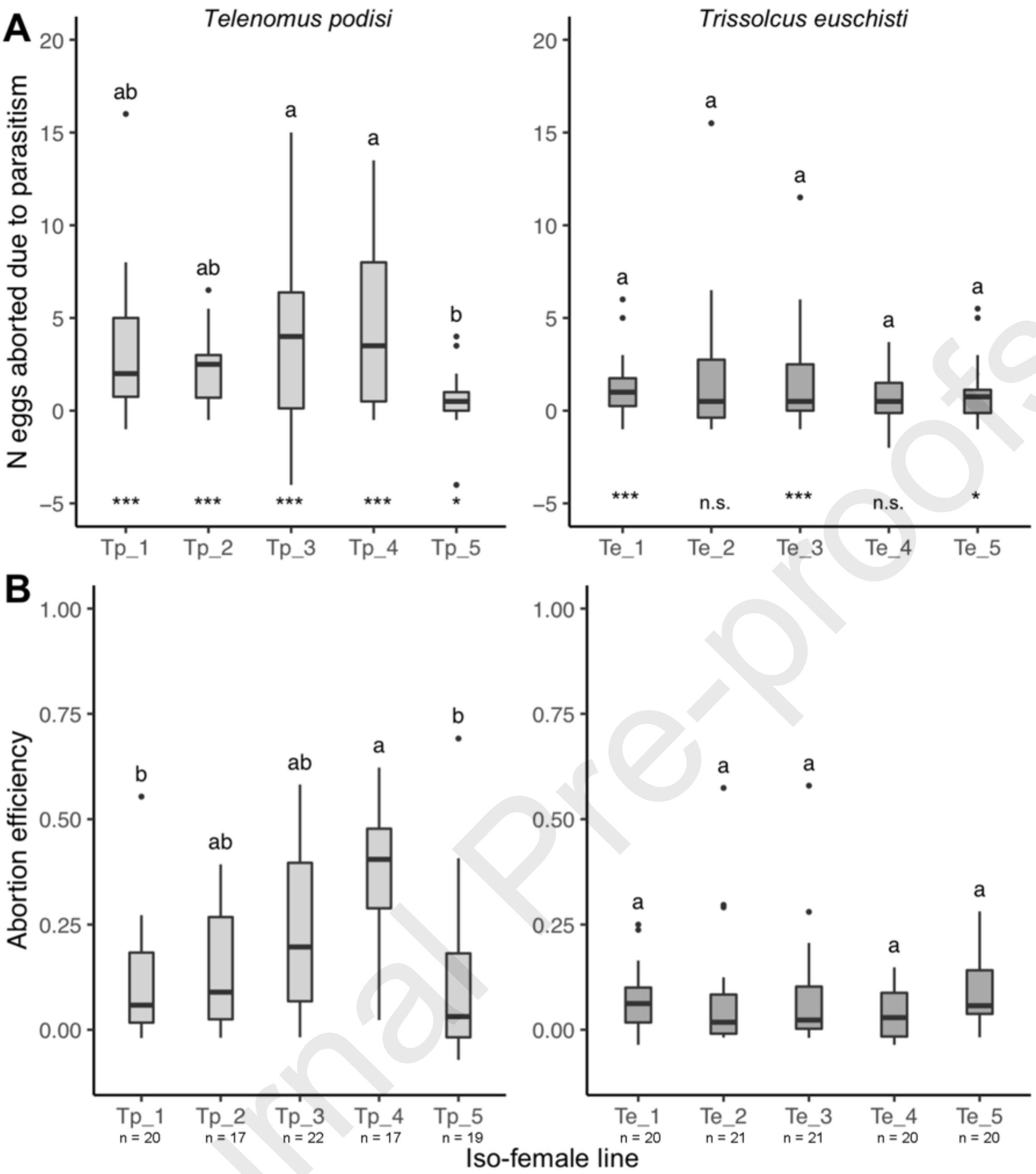


Figure 3. (A) ~~The corrected number of eggs aborting~~The number of eggs aborting due to parasitism, and (B~~) corrected~~ abortion efficiency (number of aborted eggs per parasitoid oviposition) when *H. halys* egg masses were exposed to different iso-female lines of *Trissolcus euschisti* and *Telenomus podisi*, after ~~correcting ~~adjusting for baseline levels of *H. halys* egg mortality. Negative values for some observations result from higher mortality in unexposed egg masses than exposed egg masses. In both panels, for lines within each parasitoid species, boxes not labeled with the same letter are significantly different ($p < 0.05$; Dunn's Kruskal-Wallis multiple comparison test). In (A), symbols underneath the boxes indicate whether the corrected abortion levels are significantly different from zero (n.s. – $p > 0.05$; *– $p < 0.05$; ***– $p < 0.001$; Wilcoxon

test). In boxplots, horizontal lines show medians, boxes contain the 25th-75th percentiles, whiskers show the upper and lower deciles, and points show outliers.

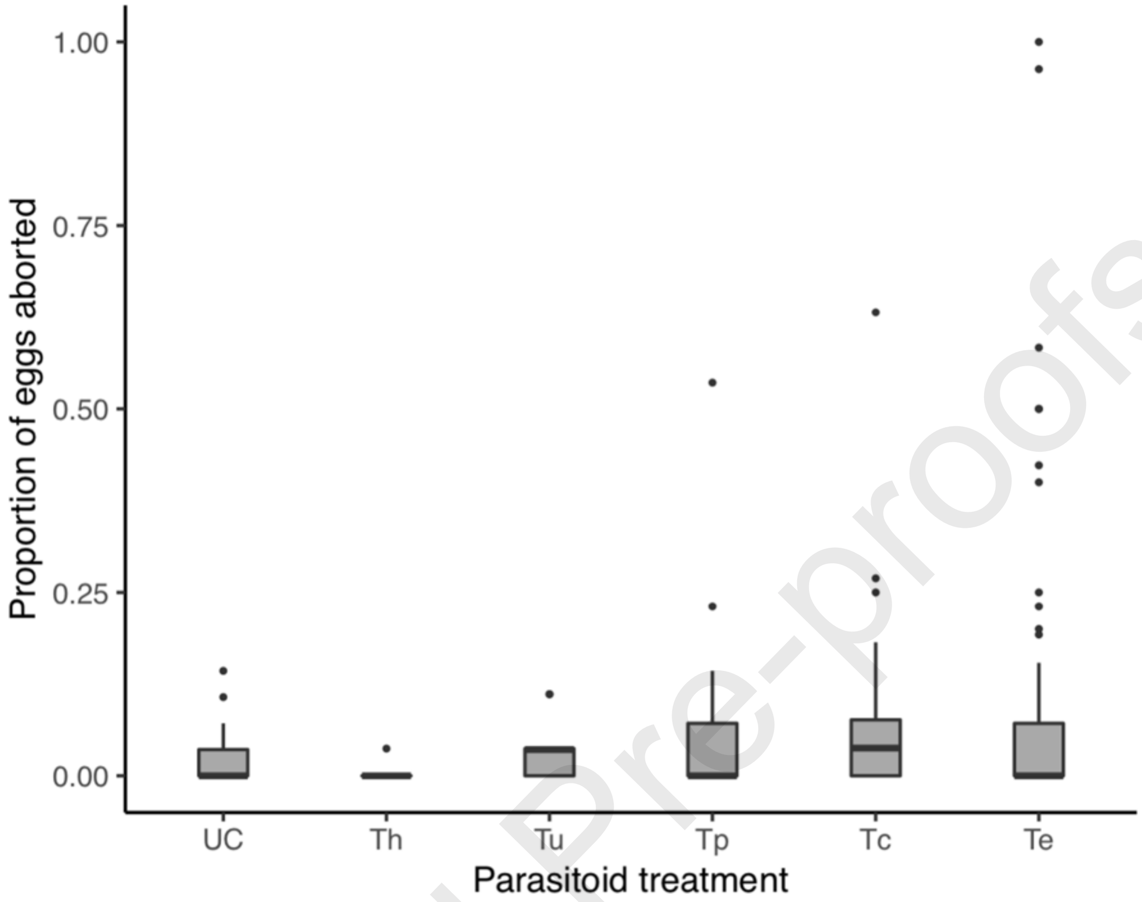


Figure 4. Percentage of *H. halys* eggs aborting in unexposed control egg masses (UC), compared to egg masses exposed to one of five parasitoid species in the laboratory for 24 h: Th – *Trissolcus hullensis* (n = 5); Tu – *Trissolcus utahensis* (n = 9); Tp – *Telenomus podisi* (n = 36); Tc – *Trissolcus cosmopeplae* (n = 46); Te – *Trissolcus euschisti* (n = 95). Horizontal lines show medians, boxes contain the 25th-75th percentiles, whiskers show the upper and lower deciles, and points show outliers. Abortion levels did not vary significantly among treatments (see the text of Results for statistical information).

Table 1. Sources of parasitoid iso-female lines used in experiments.

Species	Line name	State/Province, Country of Collection: GPS	Original emergence host	Year of collection
Telenomus podisi	Tp-1	Quebec, Canada: 45.556, -73.557	*P. maculiventris*	2013
	Tp-2	Ontario, Canada: 43.040, -81.219	*P. maculiventris*	2016
	Tp-3	Ontario, Canada: 43.028, -81.213	*P. maculiventris*	2016
	Tp-4	North Carolina, USA: 35.428, -82.564	*H. halys*	2016
	Tp-5	Ontario, Canada: 43.074, -81.337	*P. maculiventris*	2016
Trissolcus euschisti	Te-1	Ontario, Canada: 43.028, -81.213	*P. maculiventris*	2016
	Te-2	Ontario, Canada: 43.030, -81.209	*P. maculiventris*	2016
	Te-3	Ontario, Canada: 43.030, -81.209	*P. maculiventris*	2016
	Te-4	Ontario, Canada: 43.030, -81.209	*P. maculiventris*	2016
	Te-5	Quebec, Canada: 45.499, -73.346	*P. maculiventris*	2016

Table 2. Scelionid parasitoids emerging from *P. maculiventris* egg masses set out and retrieved from different sites in British Columbia, Canada, and the number of each species from each site that was subsequently offered *H. halys* eggs in the laboratory: Te – *Trissolcus euschisti*; Tp – *Telenomus podisi*; Tc – *Trissolcus cosmopeplae*; Tu – *Trissolcus utahensis*; Th – *Trissolcus hullensis*.

Region	Site Name (GPS)	# sentinel egg masses set out (total # eggs)	% egg masses parasitized per species[a] [# individuals lab-tested against *H. halys*]					
			Te	Tp	Tc	Tu	Th	Total[b]
Fraser Valley	Chilliwack #1 (49.158, -122.003)	107 (1,635)	4.67 [1]	3.74 [1]	0.00	0.00	0.00	8.41
	Chilliwack #2 (49.159, -121.997)	55 (807)	14.55 [5]	9.09 [2]	0.00	0.00	0.00	21.82
	Rosedale (49.184, -121.800)	53 (721)	3.77 [0]	7.55 [3]	0.00	0.00	0.00	11.32
	Abbotsford (49.003, -122.263)	25 (364)	20.00 [3]	4.00 [1]	0.00	0.00	0.00	24.00
	Langley (49.122, -122.657)	10 (138)	10.00 [0]	10.00 [0]	0.00	0.00	0.00	20.00
Okanagan Valley	Penticton[c] (49.483, -119.603)	579 (10,208)	4.49 [21]	1.04 [7]	8.29 [46]	2.76[9]	0.69 [5]	14.51
	Kelowna #1 (49.885, -119.484)	115 (1,958)	20.00 [31]	0.00	0.00	0.00	0.00	20.00
	Kelowna #2 (49.884, -119.456)	106 (1,821)	4.72 [5]	15.09 [15]	0.94[0]	0.00	0.00	18.87
	Kelowna #3 (49.880, -119.485)	104 (1,804)	16.35 [20]	2.88 [2]	0.96[0]	0.96[0]	0.00	20.19
	Kelowna #4 (49.871, -119.490)	110 (1,854)	6.36 [9]	2.73 [5]	0.00	0.00	0.00	9.09
	Total	1,264 (21,310)	7.75 [95]	3.32 [36]	3.96[46]	1.34[9]	0.32[5]	15.34

[a] Percentage of egg masses from which at least one individual of a given ~~each~~ species emerged

[b] Percentage of egg masses parasitized. This value is sometimes lower than row totals due to multiparasitism; i.e., multiple parasitoid species emerging from a single egg mass (this occurred in a total of 18 egg masses).

[c] One individual of an unidentified Encyrtidae emerged from a single egg mass at this site.

Credit authorship contribution statement

Elena Costi: Validation, Conceptualization, Data curation, Formal analysis, Writing - Original Draft, Writing - Review & Editing

Warren Wong: Validation, Data curation, Writing - Review & Editing

Joan Cossentine: Review & Editing, Supervision, Resources, Funding acquisition

Susanna Acheampong: Review & Editing, Supervision, Resources, Funding acquisition

Tim Haye: Writing - Review & Editing, Supervision, Funding acquisition

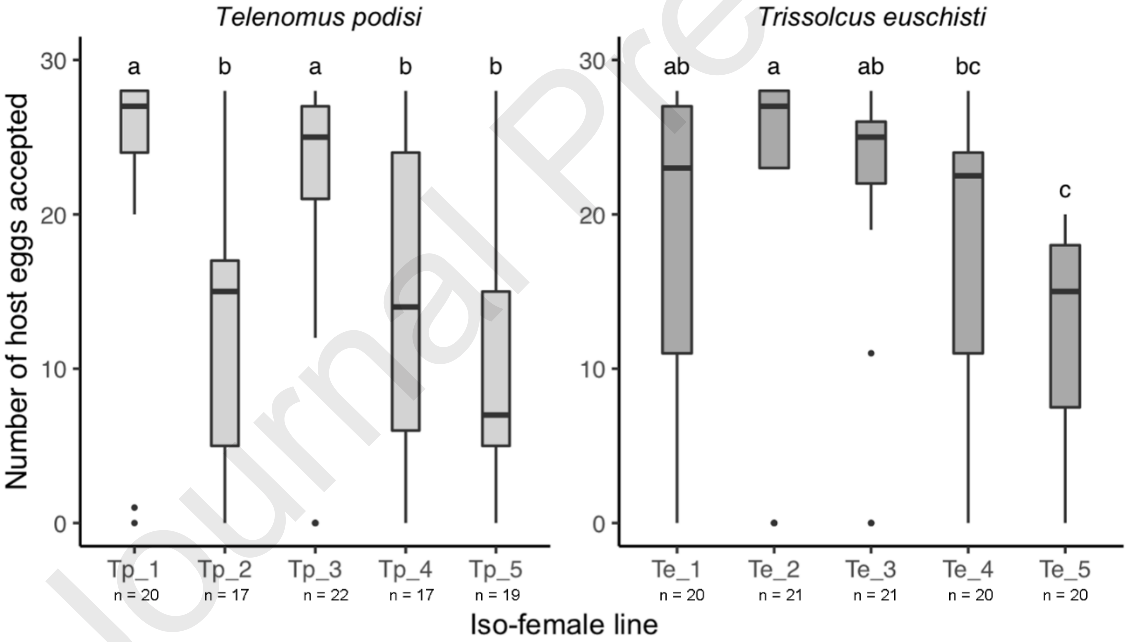
Elijah J. Talamas: Writing - Review & Editing, Data curation, Funding acquisition

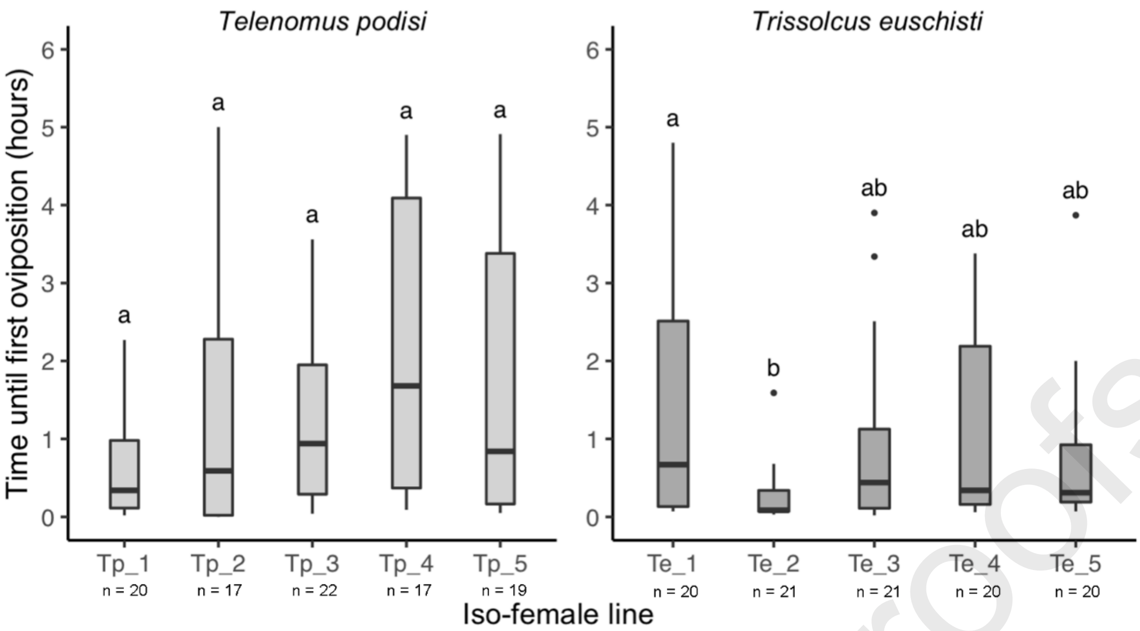
Lara Maistrello: Writing - Review & Editing, Supervision, Funding acquisition

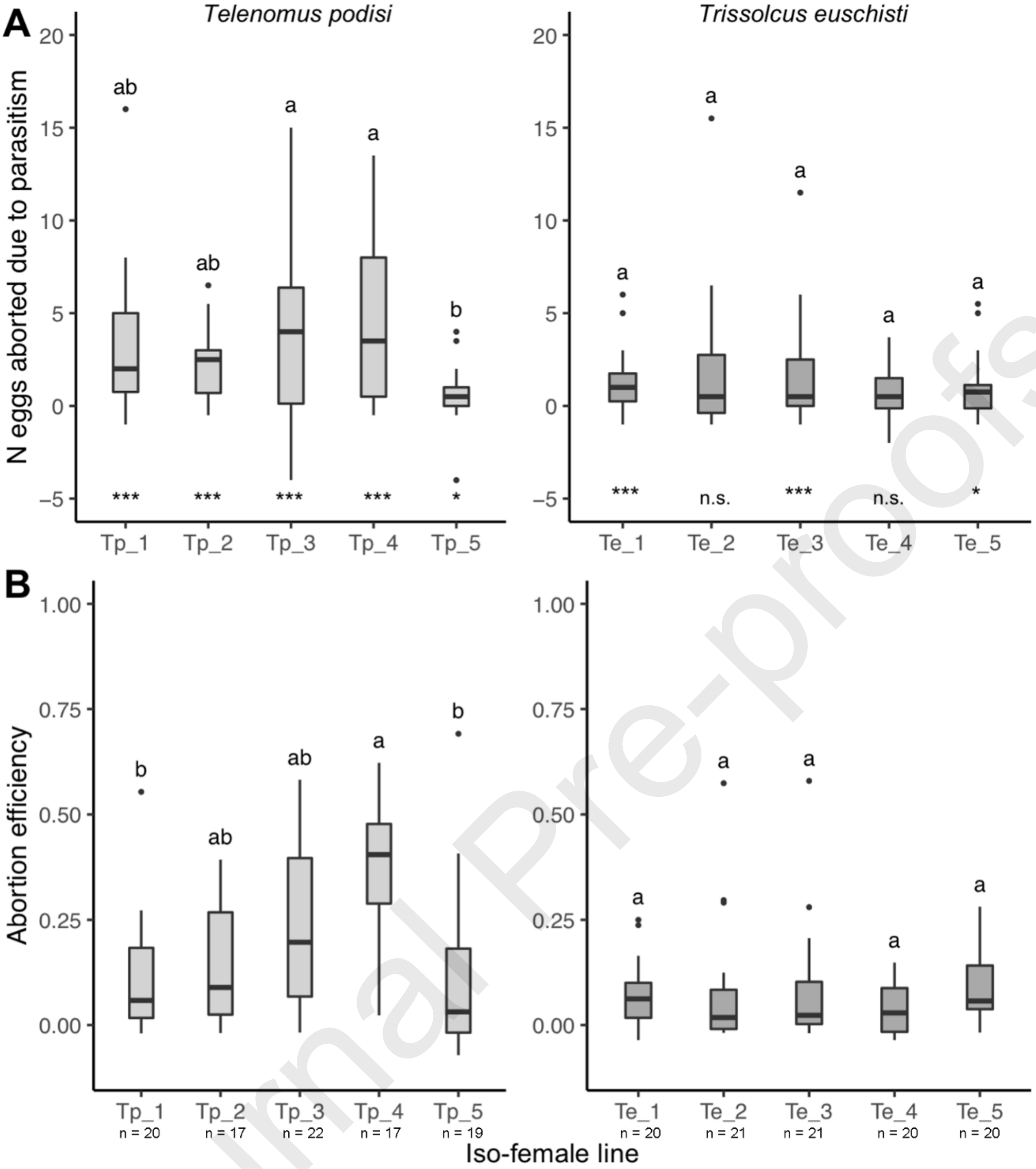
Paul Abram: Conceptualization, Formal analysis, Resources, Writing - Original Draft, Writing - Review & Editing, Visualization, Project administration

Highlights

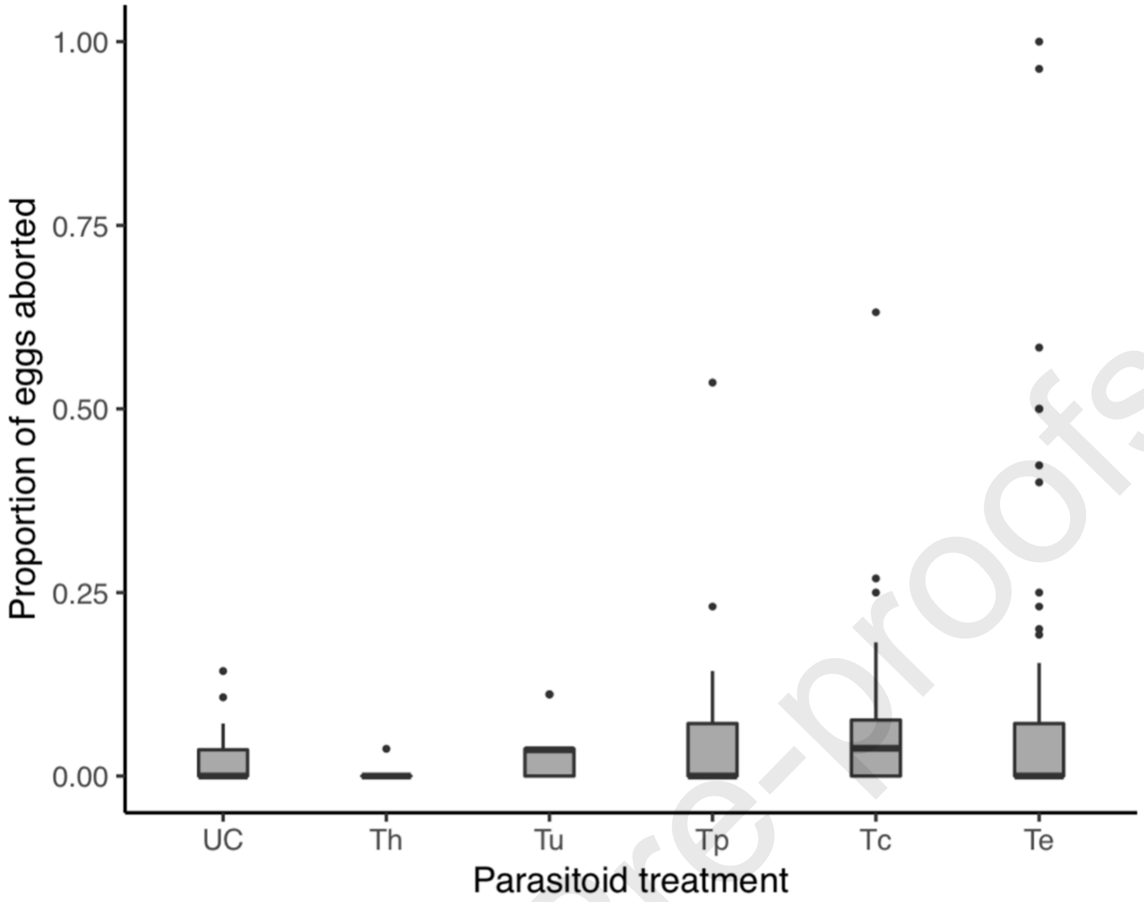
- Native North American scelionid parasitoids unsuccessfully attack eggs of *Halyomorpha halys*.
- Intra-specific variation in parasitoid acceptance behaviours was observed
- Developmental success of all species and intraspecific strains on *H. halys* eggs was almost zero.
- Variable, but low levels of ~~ariation in~~ parasitoid-induced host egg abortion were~~as~~ observed.~~ but levels were usually low.~~
-




Telenomus podisi
Trissolcus euschisti
Time until first oviposition (hours)
0
1
2
3
4
5
6
a
a
a
a
a
b
ab
ab
ab
Tp_1
Tp_2
Tp_3
Tp_4
Tp_5
Te_1
Te_2
Te_3
Te_4
Te_5
n = 20
n = 17
n = 22
n = 17
n = 19
n = 20
n = 21
n = 21
n = 20
n = 20
Iso-female line


A
Telenomus podisi
Trissolcus euschisti
N eggs aborted due to parasitism
20
15
10
5
0
−5
ab
ab
a
a
b
a
a
a
a
a
*** *** *** *** *
*** n.s. *** n.s. *
Tp_1 Tp_2 Tp_3 Tp_4 Tp_5
Te_1 Te_2 Te_3 Te_4 Te_5
B
Abortion efficiency
1.00
0.75
0.50
0.25
0.00
b
ab
ab
a
b
a
a
a
a
a
Tp_1 n = 20
Tp_2 n = 17
Tp_3 n = 22
Tp_4 n = 17
Tp_5 n = 19
Te_1 n = 20
Te_2 n = 21
Te_3 n = 21
Te_4 n = 20
Te_5 n = 20
Iso-female line

858


Proportion of eggs aborted
1.00
0.75
0.50
0.25
0.00
UC
Th
Tu
Tp
Tc
Te
Parasitoid treatment