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Evaluating Chemical Cues Associated with *Halyomorpha halys* Toward Enhanced Sensitivity of Surveillance for *Trissolcus japonicus*

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Abstract

In Asia, *Trissolcus japonicus* (Ashmead) (Hymenoptera: Scelionidae) is the predominant egg parasitoid of the brown marmorated stink bug, *Halyomorpha halys* (Stål) (Hemiptera: Pentatomidae). Detections of adventive *T. japonicus* populations in North America since 2014, where invasive *H. halys* populations have impacted various specialty crops, spurred surveillance efforts to track *T. japonicus*, and yellow sticky cards (YSC) deployed in *H. halys* host trees have proven effective for this purpose. While *T. japonicus* exhibits positive behavioral responses to several olfactory stimuli associated with *H. halys* under laboratory conditions, these have not been evaluated for their potential utility to enhance surveillance of *T. japonicus* in the field. In northwestern Virginia, where both *H. halys* and *T. japonicus* are well-established, we examined the effect of baiting tree of heaven, *Ailanthus altissima* (Mill.) Swingle (Sapindales: Simaroubaceae), with lures containing the *H. halys* aggregation pheromone and pheromone synergist on the abundance of *H. halys* egg masses and captures of *T. japonicus* in YSC. We also assessed the effect of baiting YSC with newly-laid *H. halys* egg masses or *n*-tridecane, a component of *H. halys* tarsal prints, on *T. japonicus* captures. Destructive sampling of pheromone-baited and nonbaited trees revealed no significant differences in *H. halys* egg mass abundance on foliage. Similarly, YSC deployed in pheromone-baited and nonbaited trees showed no significant differences in *T. japonicus* captures. Moreover, YSC augmented with *H. halys* egg masses or *n*-tridecane showed no increase in *T. japonicus* captures compared with nonbaited controls. The implications for surveillance of adventive *T. japonicus* are discussed.

Key words: samurai wasp, brown marmorated stink bug, parasitoid ecology, biological control

In 2010, a widespread outbreak of the invasive brown marmorated stink bug, *Halyomorpha halys* (Stål) (Hemiptera: Pentatomidae), in the Mid-Atlantic region of the United States caused significant losses to tree fruits and other specialty crops (Leskey et al. 2012), and its impact on crop protection practices persists. Native to Eastern Asia, *H. halys* has established or been detected in 46 US states (stopbmsb.org 2022) and elsewhere in the Northern and Southern Hemispheres, where it is known to feed on more than 170 wild and cultivated host plants (Leskey and Nielsen 2018). In the United States, its management of affected crops has relied primarily on broad-spectrum insecticides, which in tree fruit systems have disrupted IPM practices and led to secondary pests outbreaks (Leskey et al. 2012). Thus, the natural enemies of *H. halys* and their effects against it have received considerable attention (Haye et al. 2015; Cornelius et al. 2016a,b; Morrison et al. 2016a; Ogburn et al. 2016; Abram et al. 2017; Dieckhoff et al. 2017).

In its native Asian range, several hymenopteran parasitoids attack *H. halys* eggs (Zhang et al. 2017), although their impact on *H. halys* populations has not been quantified empirically (Abram et al. 2020). In North America, a number of native predators and parasitoids attack various *H. halys* life stages (Cornelius et al. 2016a,b, Morrison et al. 2016a, Ogburn et al. 2016), but have not yet suppressed its populations adequately (Abram et al. 2017).

Trissolcus japonicus (Ashmead) (Hymenoptera: Scelionidae) is a solitary endoparasitoid of pentatomid eggs and the predominant parasitoid of *H. halys* in Asia (Zhang et al. 2017). In 2014, an adventive population of *T. japonicus* was discovered in Beltsville, MD (Talamas et al. 2015a, Herlihy et al. 2016), and subsequent detections have occurred elsewhere in the United States (stopbmsb.org 2022), Canada (Abram et al. 2019), Switzerland (Stahl et al. 2019), Italy (Peverieri et al. 2018), and Germany (Dieckhoff et al. 2021). In the United States, *T. japonicus* has been detected in urban and semiurban settings (Milnes et al. 2016, Hedstrom et al. 2017), at the edge of unmanaged woodlands (Quinn et al. 2021), and in agricultural systems (Kaser et al. 2019, Peterson et al. 2021). CLIMEX models predict the continued spread of *T. japonicus* to many regions where *H. halys* has or is likely to become established (Avila and Charles 2018). Thus, the development of sensitive and efficient sampling methods will be critical for tracking the distribution, spread, and growth of adventive *T. japonicus* populations, which have now been reported from 15 states in the United States (stopbmsb.org 2022). To date, new state records of adventive *T. japonicus* have been established via the use of naturally-laid or sentinel *H. halys* egg masses and yellow sticky cards (YSC). YSC are effective for surveillance of adventive *T. japonicus* populations (Quinn et al. 2019a, 2021), yielding more than 500 captures during a single season from recent work in Virginia (Dyer 2022), and are more efficient than sentinel eggs for *T. japonicus* surveillance across large geographic areas (Quinn et al. 2019a).

While the visual stimulus associated with YSC elicits attraction to these traps, integrating this stimulus with kairomonal stimuli from *H. halys* may further enhance their sensitivity and effectiveness for *T. japonicus* surveillance, particularly where adventive populations are in the early stages of development, and therefore rare. *Trissolcus japonicus* did not respond to *H. halys* egg masses in Y-tube olfactometer assays (Bertoldi et al. 2019), although other scelionid egg parasitoids have been shown to respond to kairomones from the chorion or egg adhesive of their pentatomid hosts (Bin et al. 1993, Borges et al. 1999, Conti and Colazza 2012). In laboratory bioassays, *T. japonicus* responded to tarsal 'footprints' left by adult *H. halys* (Boyle et al. 2020, Malek et al. 2021) that contain the defensive metathoracic secretion, *n*-tridecane, to which it also responds (Zhong et al. 2017, Malek et al. 2021). Here, we report studies conducted in northwestern Virginia, where *T. japonicus* is well-established (Quinn et al. 2021, Dyer 2022), that evaluated the effect of baiting trees with a commercial lure containing the *H. halys* aggregation pheromone and pheromone synergist on *H. halys* egg mass abundance and *T. japonicus* captures, and the effect of baiting YSC with fresh *H. halys* egg masses or synthetic *n*-tridecane on *T. japonicus* captures.

Methods

Sampling Trees

Across all studies, mature tree of heaven, *Ailanthus altissima* (Mill.) Swingle (Sapindales: Simaroubaceae), was the standard host used for sampling. Being among the most abundant and common wild host of *H. halys* in this region (Acebes-Doria et al. 2017), its fruiting structures support the development of *H. halys* nymphs (Acebes-Doria et al. 2016), and previous research in this area yielded *T. japonicus* detections from this species over several consecutive seasons (Quinn et al. 2019a,b, 2021). All trees selected for sampling had branching and foliage from the lower portion of the trunk to the top. *Ailanthus altissima* is a dioecious species; in 2019, trees of both sexes were used, while in 2020, only mature female trees bearing seed pods (i.e., samaras) were used.

H. halys Egg Mass Collections From Pheromone-Baited and Nonbaited Trees

To assess the effect of a commercial *H. halys* lure on the abundance of *H. halys* egg masses (the resource for *T. japonicus*) in the tree canopy, sampling was conducted in pairs of trees growing at the edge of contiguous forest or in tree lines, all of which adjoined tree fruit orchards or other agricultural land. In 2019, sampling sites were in Warren County, VA (10 sites) and in 2020, Frederick County, VA and Jefferson County, WV (5 sites). In 2019, sampling in June and August coincided with predicted periods of highest *H. halys* oviposition (Nielsen et al. 2016). In 2020, sampling was conducted only in June, due mainly to disruption from the COVID pandemic, and five tree pairs were used during all sampling intervals. The mean (±SD) distance between the trees in each pair was 95.09 ± 37.64 m in 2019 and 287.40 ± 225.35 m in 2020.

Three weeks before sampling egg masses, one tree per pair was baited with a BMSB Dual Lure (Trécé, Inc., Adair, OK) (hereafter referred to as pheromone lure) containing 5 mg of *H. halys* aggregation pheromone ((3*S*,6*S*,7*R*,10*S*)-10,11-epoxy-1-bis-abolen-3-ol and (3*R*,6*S*,7*R*,10*S*)-10,11-epoxy-1-bisabolen-3-ol) (Khrimian et al. 2014), and 50 mg of methyl decatrienoate (MDT), an *H. halys* pheromone synergist (Weber et al. 2014). Lures were attached at the top of a 4.6 m bamboo pole suspended from a mid-canopy branch via a wire hook near the top of the pole. Three weeks after lure deployment, baited and nonbaited trees were felled, and their height was recorded. In 2019 and 2020, respectively, the mean (±SD) height of the felled trees was 9.02 ± 2.04 m and 12.64 ± 1.92 m. All leaves from each tree were inspected for pentatomid egg masses, which were collected and recorded. Egg masses from *H. halys* were differentiated from those of other species via our experience with identifying its eggs and the field guide from Herbert et al. (2016), and were initially assessed for the number of eggs per mass and their status (hatched, unhatched, predated). Eggs classified as hatched were empty and showed either an egg burster, indicating prior emergence of *H. halys* nymphs, or ragged edges around the opening, indicative of adult parasitoid chewing during emergence. Unhatched egg masses were held in a controlled environment chamber at 25°C and a 14 hr photoperiod and monitored for up to five weeks to determine their ultimate fate (i.e., no emergence, emergence of stink bug nymphs, or parasitoid emergence). Adult wasps that emerged were counted and identified to species.

Sampling *T. japonicus* in Pheromone-Baited and Nonbaited Trees

To assess the effect of baiting trees with the pheromone lure on *T. japonicus* captures, two trees separated by at least 50 m were selected from tree lines adjacent to fruit orchards in Frederick County, VA in 2019 (*n* = 7 sites) and 2020 (*n* = 5 sites). One tree per pair was baited with a lure, as described above, and lures remained in the trees for 11 wk, spanning the duration of the study. Per manufacturer specifications, these lures remain attractive to *H. halys* for 12 wk in the field. Each year, sampling occurred for two, three-week intervals from late June to mid-July and from late July to mid-August, spanning the period of peak *T. japonicus* abundance in this area (Quinn et al. 2021, Dyer 2022). For the first interval, sampling began 3 wk after lure deployment. Following Quinn et al. (2021), one backfolding YSC (46 × 28 cm; AlphaScents, Inc., West Linn, OR) attached atop a bamboo pole was deployed in the mid-canopy of baited and nonbaited trees. YSC were collected and replaced weekly and inspected for *H. halys* parasitoids using a dissecting microscope. As described below, all parasitoid specimens of interest (i.e., potential *H. halys* parasitoids) were removed from the YSC and prepared for identification.

Augmenting YSC with *H. halys* Egg Masses

Adult *H. halys* were maintained in screened cages (12 × 12 × 12 in; BioQuip Products, Inc., Rancho Dominguez, CA) at Virginia Tech's Research and Extension Center, Winchester, VA, at 25°C, a 16 hr photoperiod, and 55% RH. Cages were provisioned with water, an assortment of seeds, nuts, fruits, and vegetables, and small, potted green bean plants as an oviposition substrate. Egg masses (≤24-hr-old) were collected daily from bean leaves, and leaf pieces with an intact egg mass consisting of at least 20 eggs were trimmed to a ca. 1.3 cm^2 square and used the same day.

For YSC baited with *H. halys* egg masses, two leaf pieces with a fresh egg mass were placed along the midline of the adhesive surface on both sides of the card (i.e., four egg masses per YSC) (Fig. 1A, upper image). Given that the tarsal footprints left by *H. halys* on plant surfaces influence *T. japonicus* host-searching behavior (Boyle et al. 2020) and that leaf pieces with its egg masses would presumably have been contaminated by its footprints, we attempted to control for the possible effect of these on *T. japonicus* captures. Bean leaves from plants held with the *H. halys* colony were cut into 1.3 cm^2 pieces and held in a Petri dish with three adult female *H. halys* for 30 min. Two of these leaf pieces were then affixed to each side of nonbaited YSC (Fig. 1A, lower image). When the YSC were folded in half and deployed in the field, each side of the card contained two leaf pieces with an egg mass (baited) or two leaf pieces without an egg mass (nonbaited).

Pairs of baited and nonbaited YSC were deployed periodically at sites in Frederick County, VA between mid-June and early August, 2019 and 2020. According to the availability of fresh egg masses, between one and five pairs of YSC were deployed on a given day, with 12 and 13 pairs used in 2019 and 2020, respectively. Using the bamboo poles described previously, one baited and one nonbaited YSC was placed in mid-canopy of the same tree, and separated by about 1.5 m. Because the acceptability of *H. halys* egg masses to *T. japonicus* declines after about three days (Qiu et al. 2007), deployment intervals were limited to 72 hr. The position of YSC treatments in each tree was randomly assigned initially and alternated at each redeployment. Parasitoid specimens of interest were removed and held for later identification. Egg masses on the baited YSC were removed upon retrieval and held at 25°C to monitor the emergence of *H. halys* nymphs, which confirmed their viability.

Augmenting YSC with *n*-Tridecane

Five pairs of trees at sites in Frederick County, VA were used for sampling *T. japonicus* in 2020, with trees in each pair separated by at least 50 m. YSC were baited with *n*-tridecane by dispensing 5 ml of the compound into 15 ml, UVC-resistant polyethylene vials (DWK Life Sciences, LLC, Millville, NJ) and attaching the vial to a 4.6 m bamboo pole at 3 cm below the base of a YSC attached at the top (Fig. 1B). Gravimetric analysis of the vials containing *n*-tridecane and deployed with the traps was conducted weekly during the study, yielding a mean (±SD) release rate of 0.236 ± 0.062 ml per vial per week. In each pair of trees, one baited and one nonbaited YSC was deployed in the mid-canopy for seven days over five weeks between July and mid-August. Baited and nonbaited YSC were randomly assigned to trees in each pair initially and alternated at each redeployment. The YSC were replaced weekly and inspected for *H. halys* parasitoids.

Scelionid parasitoids captured on a YSC or that emerged from an egg mass, respectively, were removed in situ on a small piece of the card or placed on a small piece of YSC. These were affixed to a Petri dish using double-sided sticky tape and identified following Talamas et al. (2015b). Sex determination of *T. japonicus* specimens was based on antennal morphology (Yang et al. 2016).

Statistical Analysis

Data analyses used SAS Studio 3.8 (SAS Institute Inc., 2018, Cary, NC) and all comparisons were considered significant at $\alpha = 0.05$. The distributions of *T. japonicus* captures and *H. halys* egg masses from pheromone-baited and nonbaited trees were assessed for normality using the Shapiro–Wilk

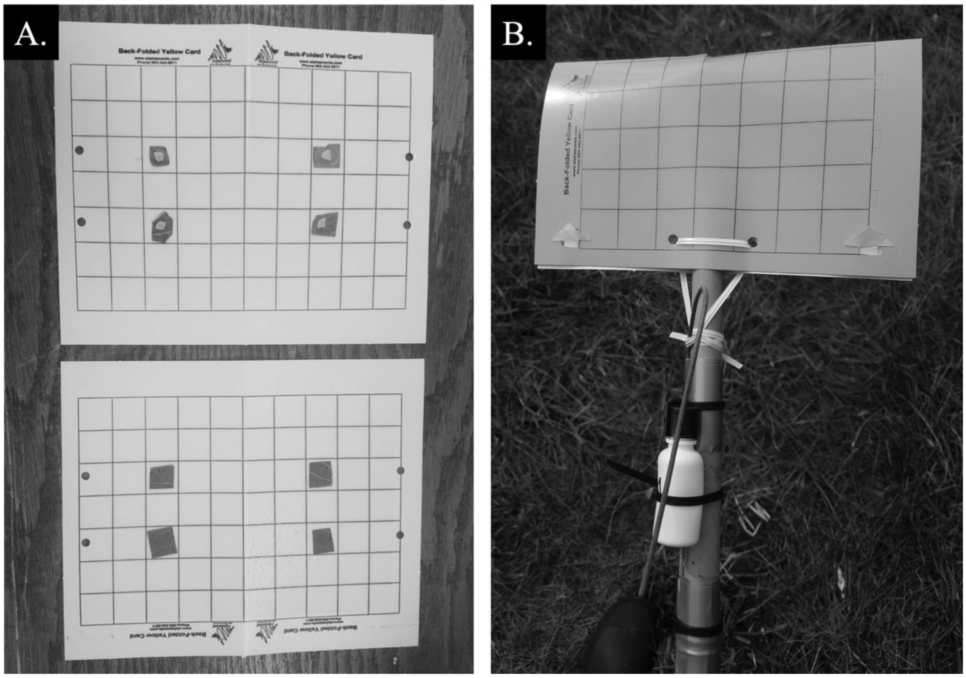

Fig. 1. (A) Backfolding yellow sticky card baited with *Halyomorpha halys* egg masses on bean leaf pieces (upper image) and nonbaited card with bean leaf pieces contaminated by *H. halys* tarsal footprints (lower image) (when cards were folded in half, each sticky surface contained two leaf pieces) and (B) synthetic *n*-tridecane in a polyethylene vial.

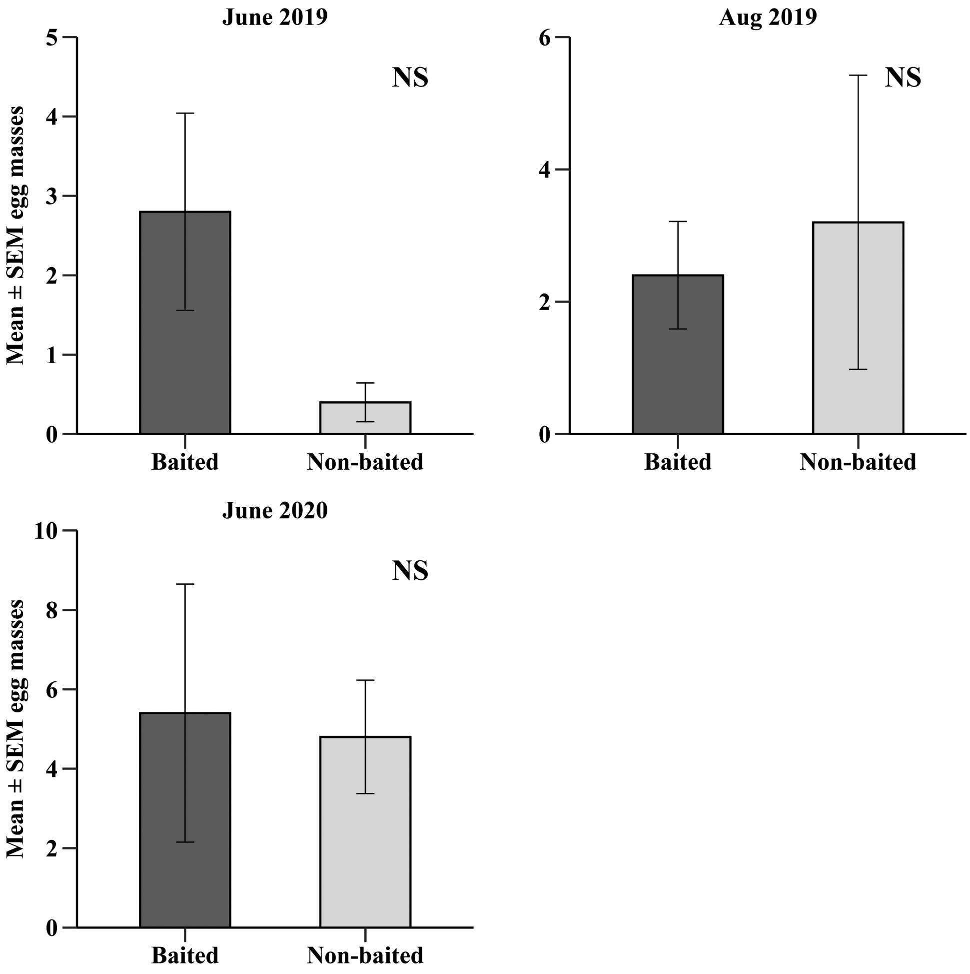

Fig. 2. *Halyomorpha halys* egg masses collected from pairs of felled pheromone-baited and nonbaited *Ailanthus altissima* in June and August, 2019 (*n* = 5 tree pairs per sampling period) and June, 2020 (*n* = 5 tree pairs) in Virginia.

test. Because counts of *H. halys* eggs masses from baited and nonbaited trees were not normally distributed, a nonparametric Wilcoxon signed-rank test was used. Data from early and late season sampling in 2019 were analyzed separately, based on the likelihood that hatched egg masses collected during the late season reflected cumulative oviposition since the onset of egg-laying in May or June. To compare *T. japonicus* detections between pheromone-baited and nonbaited trees, captures across each three-week sampling interval were pooled and analyzed using a paired t-test. At one site in 2019, no *T. japonicus* were captured in baited or nonbaited trees, and this site was excluded from analyses. Captures of *T. japonicus* were compared between egg-baited and nonbaited YSC using the Wilcoxon signed-rank test, while captures of *T. japonicus* in *n*-tridecane baited and nonbaited cards were pooled across sampling dates and compared using a paired t-test.

Results

H. halys Egg Mass Collections From Pheromone-Baited and Nonbaited Trees

In 2019, 25 and 28 pentatomid egg masses were collected from felled *Ailanthus* trees in June and August, respectively. In June 2020, 51 pentatomid egg masses were found. Of these, *H. halys* egg masses comprised 64% (*n* = 16) and 100% in June and August, 2019, respectively, and 100% in June, 2020. The mean (±SEM) number of *H. halys* eggs per mass was 24.59 ± 0.89 in 2019 and 26.86 ± 0.44 in 2020.

There was not a significant effect of baiting trees with an *H. halys* pheromone lure on the number of *H. halys* egg masses found in June ($P = 0.25, S = 4$) and August, 2019 ($P = 1.0, S = 0$) or in June, 2020 ($P = 0.63, S = -2.5$) (Fig. 2). None of the previously hatched *H. halys* egg masses found showed indications of prior parasitoid emergence, despite the subsequent emergence of parasitoids from unhatched eggs collected during each sample event (Table 1). Of the unhatched egg masses found, the percentage of those eventually yielding *H. halys* nymphs was highest in June 2019, and the percentage of masses producing adult parasitoids was very similar between the two samples in 2019, but increased substantially in June 2020 (Table 1). No *T. japonicus* emerged from eggs collected in June 2019, but the majority of parasitoids that emerged in August 2019 and June 2020 were *T. japonicus* (Table 2). Interestingly, despite the trees having been felled, in 2020 we also found three female wasps guarding egg masses, one of which was *Trissolcus brochymenae* (Ashmead) and the others *T. japonicus*.

T. japonicus Captures in Pheromone-Baited and Nonbaited Trees

In 2019 and 2020, respectively, 119 and 146 wasps from ≥6 scelionid species were captured, with *T. japonicus* predominating each year (Table 3) and showing a strong female bias in 2019 (93.7% female) and 2020 (92.9% female). However, there was not a significant effect of baiting trees with the *H. halys* aggregation pheromone on

Table 1. Status of *H. halys* egg masses collected from pairs of felled *Ailanthus altissima* in Frederick County, VA

Status	June 2019, n = 16 masses, %	Aug. 2019, n = 28 masses, %	June 2020, n = 51 masses, %
Previously hatched[a]	25.0	67.9	41.2
Predated	6.3	10.7	0
H. halys nymphs emerged[b]	56.2	3.6	33.3
Adult parasitoids emerged[c]	12.5	14.2	23.5
No emergence	0.0	3.6	2.0

One tree per pair was baited in mid-canopy with a commercial *H. halys* pheromone lure.

[a] All previously hatched egg masses had produced *H. halys* nymphs, indicated by the presence of egg bursters.

[b] Egg masses that were unhatched at collection and subsequently yielded *H. halys* nymphs.

[c] Egg masses that were unhatched at collection and subsequently yielded adult parasitoids.

Table 2. Number of parasitoid adults that emerged from *H. halys* egg masses collected from the foliage of pairs of felled *Ailanthus altissima* in Frederick County, VA

		Trissolcus (Scelionidae)			*Anastatus* (Eupelmidae)
Date	**Treatment**	*japonicus*	*thyantae*	*euschisti*	**spp.**
June 2019	Baited	0	13 (1)	0	2 (1)
	Nonbaited	0	0	0	0
	% of total	0.0	86.7	0.0	13.3 (1)
Aug 2019	Baited	49 (2)[a]	0	0	2
	Nonbaited	13 (1)	0	0	0
	% of total	96.9	0.0	0.0	3.1
June 2020	Baited	50 (2)	0	0	0
	Nonbaited	52 (2)	0	18 (1)	61 (7)
	% of total	56.4	0.0	9.9	33.7

One tree per pair was baited in mid-canopy with a commercial *H. halys* pheromone lure.

[a] Number of *H. halys* egg masses yielding specified parasitoid species shown in brackets.

Table 3. Number of Scelionidae parasitoids captured in yellow sticky cards deployed in pairs of *H. halys* pheromone-baited and nonbaited *Ailanthus altissima* in Frederick County, VA[a]

	Trissolcus				Telenomus	
Year and treatment	*japonicus*	*euschisti*	*thyantae*	*brochymenae*	*podisi*	spp.
2019						
Baited	39	9	3	4	8	2
Nonbaited	24	10	6	3	9	2
% of total[a]	52.9	16.0	7.6	5.9	14.3	3.3
2020						
Baited	21	24	1	6	30	3
Nonbaited	35	4	0	2	19	1
% of total	38.3	19.2	0.7	5.5	33.6	2.7

Yellow sticky cards deployed for two, 3-wk intervals in Aug., 2019 and June 2020.

[a] Percentages based on species for which >1 specimen was captured across both years.

T. japonicus captures in 2019 (t = 0.91, df = 5, P = 0.41) or 2020 (t = –1.69, df = 4, P = 0.17) (Fig. 3).

Augmenting YSC with *H. halys* Egg Masses and *n*-Tridecane

In 2019 and 2020, respectively, 91.7% and 100% of the fresh egg masses deployed on YSC yielded *H. halys* nymphs, confirming their viability. In 2019, one egg mass yielded *T. japonicus* adults. Total Scelionidae captures (≥6 species) were 46 and 99 in 2019 and 2020, respectively. Captures of *T. japonicus* predominated in 2019 but not 2020 (Table 4), and were female biased each year (2019: 77.8% female; 2020: 86.4% female). There was not a significant effect of baiting YSC with freshly-laid *H. halys* egg masses on *T. japonicus* captures in 2019 (P = 0.68, S = 6) or 2020 (P = 0.68, S = –5.5) (Fig. 4).

In 2020, a total of 66 *T. japonicus* (95.5% female) were captured in YSC baited with *n*-tridecane and nonbaited cards across five weeks of sampling, and there was not a significant difference (t = 0.19, df = 4, P = 0.86) in mean weekly captures between baited (6. 8 ± 3.40 SE) and nonbaited (6. 4 ± 1.44 SE) YSC. Of the 196

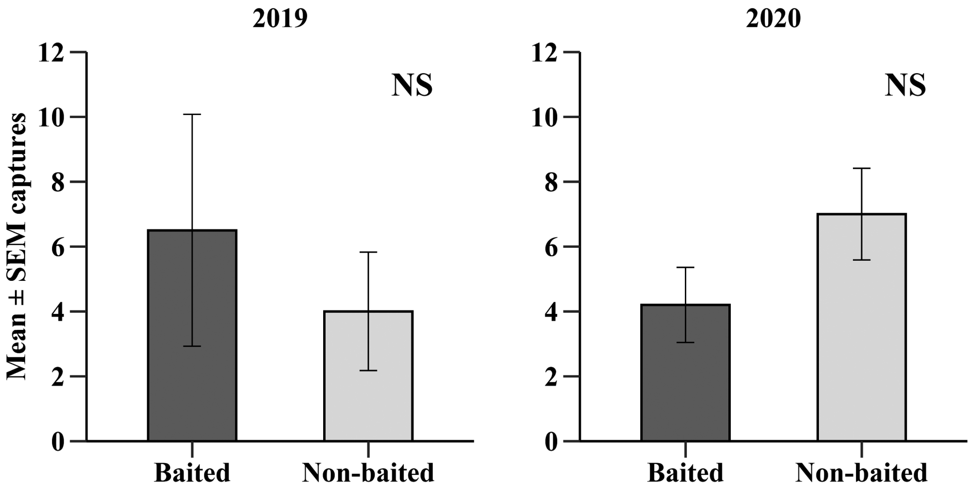

Fig. 3. *Trissolcus japonicus* captures in yellow sticky cards deployed in the mid-canopy of pairs of pheromone-baited and nonbaited *Ailanthus altissima* from 21 June to 12 July and 24 July to 14 August 2019 (*n* = 6 tree pairs) and from 1 July to 22 July and 29 July to 19 August 2020 (*n* = 5 tree pairs).

Table 4. Number of Scelionidae parasitoids captured in pairs of *H. halys* egg-baited and nonbaited yellow sticky cards deployed in the mid-canopy of individual *Ailanthus altissima* in Frederick County, VA

	Trissolcus				Telenomus	
Year and treatment	*japonicus*	*euschisti*	*thyantae*	*brochymenae*	*podisi*	spp.
2019						
Eggs	23	2	3	1	1	0
No. Eggs	13	2	0	0	1	0
% of total	78.3	8.7	6.5	2.2	4.3	0.0
2020						
Eggs	8	0	0	1	33	5
No. of eggs	14	2	1	0	32	3
% of total	22.2	2.0	1.0	1.0	65.7	8.1

Yellow sticky cards deployed for 72-hr intervals from 9 July to 14 Aug. 2019 (12 trap pairs) and from 12 June to 21 Aug. 2020 (13 trap pairs).

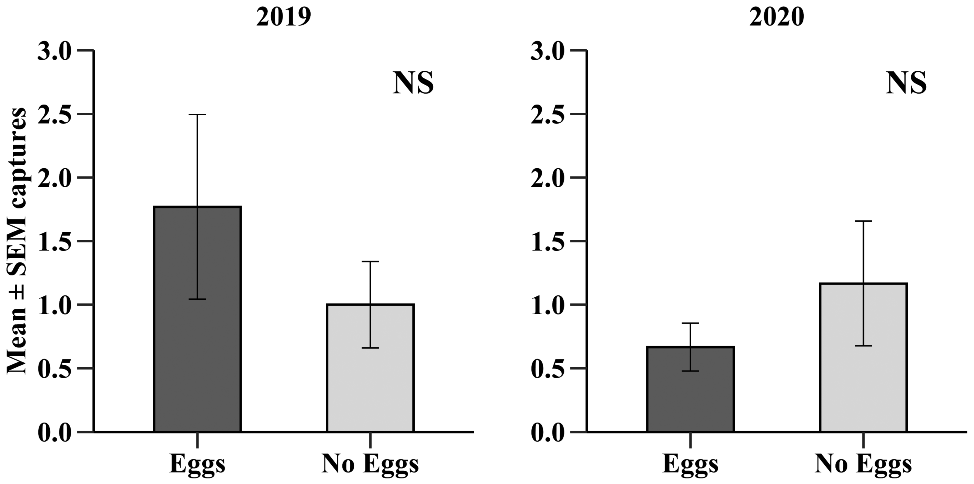

Fig. 4. *Trissolcus japonicus* captures in pairs of yellow sticky cards baited with fresh H. halys egg masses and nonbaited yellow sticky cards deployed for 72-hr in the mid-canopy of individual *Ailanthus altissima* from 9 July to 14 August 2019 (*n* = 12 pairs) and from 12 June to 21 August 2020 (*n* = 13 pairs).

scelionid specimens captured, 33.7% were *T. japonicus*, with the remainder being *Telenomus podisi* Ashmead (43.9%), *Trissolcus euschisti* (Ashmead) (15.8%), *Trissolcus brochymenae* (2.6%), *Trissolcus thyantae* Ashmead (2.6%), 2 other *Telenomus* spp. (1%), and *Hadronotus pennsylvanicus* (Ashmead) (0.5%).

Discussion

Lures containing the *H. halys* aggregation pheromone and its synergist, MDT, consistently produce increased densities of *H. halys* in their vicinity (Weber et al. 2014, Leskey et al. 2015, Morrison et al.

2016b, Formella et al. 2020), increased captures in traps (Acebes-Doria et al. 2018, 2019; Leskey et al. 2015, 2021), and increased retention duration of responding individuals on a baited plant (Morrison et al. 2016b). Our reasonable assumption of increased *H. halys* densities in trees baited with its pheromone lure, relative to a nonbaited tree, led to two hypothetical, nonexclusive outcomes, 1) increased egg mass abundance on leaves, and 2) increased concentration of tarsal footprints on plant surfaces.

Based on the following findings, a corollary to our hypotheses is the potential for increased abundance or retention of *T. japonicus* in baited trees with higher *H. halys* densities, thereby potentially enhancing the likelihood of its detection via surveillance methods. Some scelionid parasitoids are known to respond to kairomones associated with the eggs of their pentatomid hosts (Bin et al. 1993, Borges et al. 1999, Conti and Colazza 2012), although this was not shown for *T. japonicus* in olfactometer studies. In laboratory assays, female *T. japonicus* respond to tarsal prints from *H. halys* (Boyle et al. 2020, Malek et al. 2021), as do other Scelionidae with pentatomid hosts (Conti et al. 2004, Salerno et al. 2006, Colazza et al. 2007, Peri et al. 2016). Moreover, Arif et al. (2021) reported that *T. japonicus* discriminated between the sexes of *H. halys* based on tarsal residues, residing longer on patches visited by females. The response of female *T. japonicus* to *H. halys* footprints may be related to the presence of *n*-tridecane, a main component of its tarsal residues (Malek et al. 2021). This compound is a defensive metathoracic secretion and elicits electrophysiological and behavioral responses in female *T. japonicus* (Zhong et al. 2017, Malek et al. 2021), as well as behavioral responses in other pentatomid natural enemies, such as the minute pirate bug, *Orius insidiosus* (Say) (Fraga et al. 2017) and *Telonomus podisi* (Laumann et al. 2009). In laboratory olfactometers, *T. japonicus* also was attracted to volatiles from tomato plants on which *H. halys* had fed and oviposited (Bertoldi et al. 2019), suggesting that herbivore-induced plant volatiles (HIPVs) may facilitate host location by foraging female *T. japonicus*. However, despite the obvious presence and wide distribution of *T. japonicus* among our study sites, our experiments intended to manipulate/enhance or directly assess stimuli associated with *H. halys* did not result in higher detections or captures of *T. japonicus*, for which we offer the following possible explanations.

In semiurban settings in Blacksburg, VA, where *T. japonicus* has not yet been detected, Formella et al. (2020) examined the response of *H. halys* to an aggregation pheromone lure deployed in the lower canopy of ornamental trees using ground-based visual surveys, revealing significantly greater *H. halys* counts in baited versus nonbaited trees, but no effect on the number of egg masses observed. We conducted modified a version of the Formella et al. (2020) study, using a wild tree host in a natural setting and exhaustive inspection of leaves on felled trees. Because Quinn et al. (2019b) showed consistently and significantly higher captures of *H. halys* in the mid- and upper-canopy of *A. altissima,* compared with the lower canopy, and the highest numbers of *H. halys* egg masses in the upper portions of the canopy, our trees were baited with the lure in mid-canopy. However, results from pairs of pheromone-baited and nonbaited *A. altissima* yielded no significant differences in egg mass abundance, concurring with the results of Formella et al. (2020) and confirming that higher densities of *H. halys* did not result in higher numbers of its egg masses. The plume reach of *H. halys* pheromone lures and the area over which it attracts *H. halys* has been quantified (Kirkpatrick et al. 2019), but the aggregation response of this species is a proximate effect. Indeed, the influence of its aggregation behavior on mating and reproduction and the ultimate ecological role of its pheromone is poorly understood compared with other taxa such as Lepidoptera. Our results and those of Formella et al. (2020) highlight this knowledge gap.

The presumed increased concentration of *H. halys* tarsal prints on the surfaces of pheromone-baited trees did not translate to increased detections of *T. japonicus,* compared with nonbaited trees. While tarsal prints may be perceived by *T. japonicus* over short distances, gravimetric analyses of vials containing a main component of tarsal residues, *n*-tridecane, showed that *n*-tridecane volatilized to some extent. Yet in a separate study that directly assessed its effect, YSC baited with *n*-tridecane showed no effect on *T. japonicus* captures.

Kairomones on the egg chorion are typically also short-range or contact stimuli that may facilitate host recognition and acceptance in pentatomid parasitoids (Conti and Colazza 2012). Furthermore, adhesive secretions produced by ovipositing female stink bugs also can serve as contact kairomones for host recognition (Bin et al. 1993, Borges et al. 1999). Chemical extracts of the egg adhesive produced by *Euschistus heros* (F.) applied to glass beads elicited host searching and acceptance behaviors by *Tel. podisi* (Borges et al. 1999), and Bin et al. (1993) showed a similar response to the adhesive material from *Nezara viridula* (L.) eggs by its parasitoid, *Trissolcus basalis* (Wollaston). While olfactometer assays using egg volatiles from *Murgantia histrionica* (Hahn) suggested that *T. brochymenae* responded to them over some distance (Conti et al. 2003), Borges et al. (1999) noted that the volatility of egg adhesive kairomones is relatively low. We found no significant difference in *T. japonicus* captures between YSC with and without fresh *H. halys* egg masses, conforming with the results from laboratory assays by Bertoldi et al. (2019), and suggesting either no attraction to egg masses or a very localized, close-range attraction to egg kairomones that may have been present.

In combination, our results indicated that baiting host plants with the *H. halys* pheromone lure or augmenting YSCs with its egg masses or *n*-tridecane did not improve the effectiveness or sensitivity of YSCs for *T. japonicus* surveillance. In concurrence, Morrison et al. (2018) showed that neither predation nor parasitism of fresh *H. halys* egg masses deployed on the underside of tree leaves in the field were affected by the presence of a high-dose *H. halys* pheromone lure placed <1 m away on the same branch. Moreover, Morrison et al. (2018) also found that other semiochemicals, including the pheromone of two potential egg predators, *Podisus maculiventris* (Say) and *Coccinella septempunctata* (L.), and the plant volatile, methyl salicylate, used in combination with the *H. halys* pheromone in 1 m^2 field plots did not enhance predation or parasitism of *H. halys* egg masses. However, despite our negative results, these studies can inform future efforts to enhance the performance of *T. japonicus* surveillance tools, if only by demonstrating that the stimuli and deployment and sampling protocols used did not enhance *T. japonicus* detections in an area where its adventive populations are comparatively robust and widely distributed. Certainly, there remains an incomplete understanding of the foraging behavior and ecology of *T. japonicus* in the field, particularly given the relative scarcity of *H. halys* egg masses, the distribution of these eggs among many potential host tree species, and the amount of foliar surface area on which they are deposited. The integration of host-associated stimuli with the visual stimulus from YSC may yet have the potential to enhance the sensitivity of current sampling tools, which would be especially useful for surveillance where adventive *T. japonicus* populations are in early stages of establishment. For example, the role of HIPVs associated with *H. halys* feeding and oviposition on the foraging behavior female *T. japonicus* in the field remains to be explored, particularly in view of its response to them in a laboratory olfactometer (Bertoldi et al. 2019). Identification of the HIPVs to which *T. japonicus* responds and their use with traps is a potential avenue for exploration, as is evaluating its response to potential *H. halys* egg kairomones and, given a positive response, their identity, volatility, and concentration effects.

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