



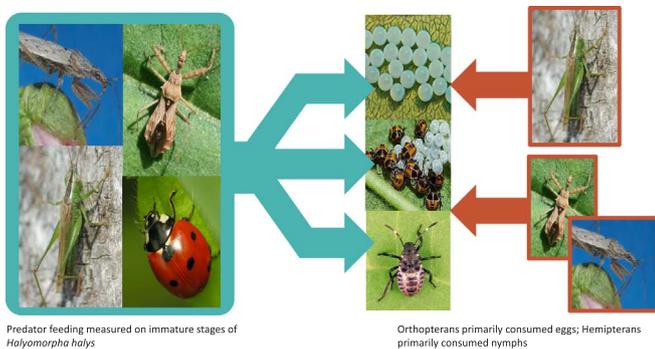
## Life stage specific predation of *Halyomorpha halys* (Stål) by generalist predators



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### GRAPHICAL ABSTRACT



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### ABSTRACT

*Halyomorpha halys* (Stål) is an invasive pest of agriculture in the USA. Feeding damage from *H. halys* affects dozens of crops yet little is known about the community of predators which prey on *H. halys* in its invaded range. Ten taxa of predatory or omnivorous insects were evaluated for their capacity to consume eggs and nymphs of *Halyomorpha halys* in laboratory mesocosm experiments. Predators were collected from agricultural ecosystems in New Jersey, starved for 24–48 h, and then exposed to *H. halys* eggs, first instar, or second instar nymphs. Survivorship of control prey in predator-excluding containers within the arenas was compared to that of predator treatment groups to determine the effect of predator presence. Stage-specific differences in *H. halys* survivorship among life stages were observed for several predator taxa indicating stage-specific predation. Acrididae, *Coccinella septempunctata* (L.), *Podisus maculiventris* (Say) (nymphs and adults), and Tettigoniidae reduced the hatch rate of *H. halys* eggs. Hemipteran predators, including *Nabis* spp. and Reduviidae, reduced the survivorship of first instar nymphs. Similarly, *Nabis* spp. and *P. maculiventris* nymphs reduced the survivorship of second instar nymphs. Acrididae, *Nabis* spp., *P. maculiventris* nymphs, Reduviidae, and Tettigoniidae showed stage-specific tendencies in their consumption of *H. halys*. Morphological similarities between the immature stages of *H. halys* may facilitate predator suppression of these mobile stages. These results indicate that predation estimates that rely solely on sentinel egg masses may underestimate the impact of generalist predators on other *H. halys* life stages.

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## 1. Introduction

Natural enemies play an important role in the regulation of insect pest populations (Hassell and May, 1986; Symondson et al., 2002). A meta-analysis of biological control literature over the last 10 years showed that, overall, biological control agents significantly reduce pest abundance compared to predator-protected control groups (Stiling and Cornelissen, 2005). Although predators may generally suppress insect herbivore populations, the effect of individual predator taxa on prey abundance can vary widely among prey life stages. Lycosid spiders can cause 91% mortality of small, third instar grassland acridids, but only 63.5% and 30.4% mortality of larger fifth instars and adults, respectively (Oedekoven and Joern, 1998). Predation on *Frankliniella occidentalis* (Pergande) flower thrips by *Orius insidiosus* (Say) varies among prey life stages and is mediated by shifts in prey behavior between nymph and adult stages (Baez et al., 2004). Predatory mites that consume eggs and larvae of *Tetranychus urticae* (Koch), also show life-stage preferences that vary with their diet breadth; oligophagous species prefer eggs, whereas generalist species show no preference for prey life stage (Blackwood et al., 2001).

Variation in behavior and other predatory cues among prey life stages may drive the demographic prey preferences of generalist predators. Although olfactory kairomones emitted by prey are often used by specialists for prey location, many generalists rely heavily on visual cues (Vet and Dicke, 1992). To reduce the risk of predation, arthropods often limit their visual detectability by reducing movement and foraging behaviors in the presence of predators (Lind and Cresswell, 2005; Nelson, 2007; Persons et al., 2001; Sih, 1986). Sessile prey such as eggs or female scale insects may provide weaker visual cues than mobile life stages, which may affect rates of predator attack. Alternatively, mobile prey may be more susceptible to attack by more visually-oriented predation strategies such as 'sit-and-wait' predation (Nelson, 2007; Olberg et al., 2000).

Differential impacts of predation on various pest life stages can have important implications for monitoring pest abundance, especially when the severity of pest damage varies among life stages. *Lygus hesperus* (Knight) is a pest of cotton at all life stages, but feeding by late instar nymphs and adults causes the majority of economic damage (Zink and Rosenheim, 2005). The primary predators of *L. hesperus* in cotton are *Geocoris* spp. which feed preferentially on the early, less damaging, nymphal instars. However, nymphs are more easily sampled than the winged adult *L. hesperus* and as a result, high nymphal predation complicates attempts to monitor the economically damaging adult populations. The disconnect between predator preferences and developmental changes in prey damage has important consequences for patterns of pest damage and for efforts to monitor pest populations (Zink and Rosenheim, 2007).

Brown marmorated stink bug (BMSB), *Halyomorpha halys* (Stål), is an invasive pest of agriculture across the US and parts of Europe (Hoebeke and Carter, 2003; Leskey et al., 2012a; Maistrello et al., 2016; Nielsen and Hamilton, 2009b). As a recently invaded species, *H. halys* can be a useful model organism for studying the role of generalist natural enemies on suppressing invasive prey populations. *Halyomorpha halys* feeds on multiple agriculturally important crops including apple, peach, tomato, pepper, corn, and soybean, making it a landscape-level pest (Leskey et al., 2012b; Rice et al., 2016, 2014). Late instar *H. halys* nymphs cause significantly more injury to peaches and more discolored depressions in apples than do younger nymphs (Acebes-Doria et al., 2016). As a result, predator impacts on early instars may result in less damage reduction than similar predation rates on older instars. The high density of *H. halys* in many agricultural settings may accelerate predator adaptation due to the high frequency of predator-prey encounters (Basnet et al., 2014; Nielsen et al., 2011; Nielsen and Hamilton, 2009). Sentinel egg masses have been used across habitats in the invaded region to study the effect of natural enemies on *H. halys*. Sentinel *H. halys* egg masses placed within agricultural sites in seven

states across the lower Midwest and Mid-Atlantic regions during 2013–2014 were parasitized at very low rates (< 1% of eggs produced an adult parasitoid) (Ogburn et al., 2016). In Mid-Atlantic ornamental nurseries, parasitism of *H. halys* egg masses varies greatly, possibly due to differences in parasitoid community (Cornelius et al., 2016; Jones et al., 2014). Predation on these egg masses, at least in agricultural settings, was markedly higher (up to 20% in some crops), suggesting that predators may have a greater effect on *H. halys* populations than do native parasitoids (Ogburn et al., 2016).

Due to the high abundance and impact of *H. halys* within diverse agroecosystems, it is important to understand how the generalist predator community is responding to this novel resource. Our understanding of predator impacts on *H. halys* to date has focused on one sessile life stage (eggs) and thus does not present a complete picture of predation. Evidence from *Nezara viridula* (L.) suggests that the community of predators consuming eggs and nymphs may be largely non-overlapping (Ragsdale et al., 1981). Behavioral differences exist between sessile *H. halys* eggs, aggregated first instar nymphs, and highly mobile second instar nymphs (Nielsen and Hamilton, 2009a), which may affect predation of these stages. Thus, the objectives of this research were to 1) determine the community of generalist predators that attack *H. halys* eggs, first and second instar nymphs in semi-natural arenas and 2) determine if predators differentially attack the various early life stages of *H. halys*.

## 2. Materials and methods

### 2.1. Predators and prey

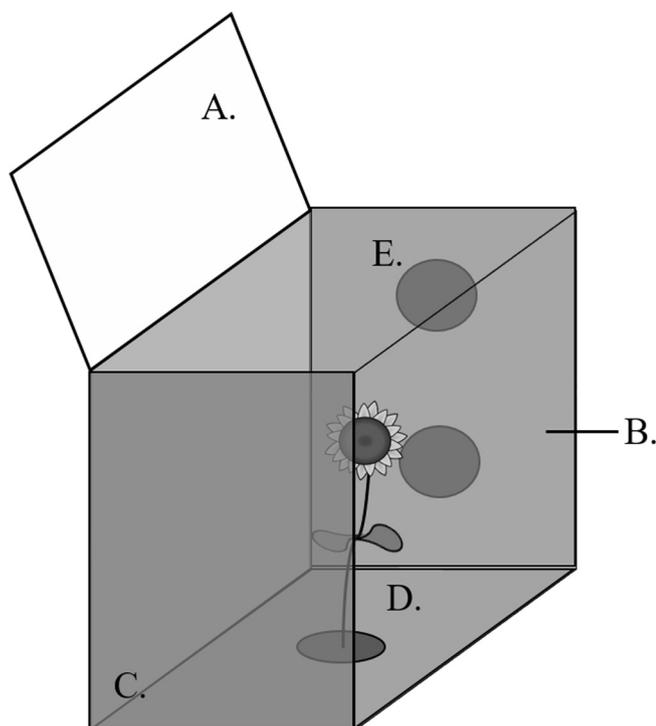
To test the acceptability of *H. halys* to natural enemies in laboratory, field collected predators were exposed to *H. halys* eggs, first instars, and second instar nymphs in no-choice predation assays.

Predators were collected at the Rutgers Agriculture Research and Extension Center (RAREC) in Bridgeton, NJ from organic soybean and rye by sweep-netting and beat sampling in conventionally managed peaches. From May through October of 2015–2016, preliminary sampling determined which taxa were sufficiently abundant to include in the study. Individuals of each abundant predator taxa were first tested in experimental arenas to determine if their behavior was significantly altered by this confinement and, as a result, ants and spiders were excluded from this study. Predator taxa tested in this experiment included Acrididae (represented by one unidentified morpho-species), *Coccinella septempunctata* (L.), *Coleomegilla maculata* (DeGeer), *Geocoris* spp., *Harmonia axyridis* (Pallas), *Hippodamia convergens* (Guérin-Méneville), *Nabis* spp., adults and 3rd-4th instar nymphs of *Podisus maculiventris* (Say), Reduviidae (consisting of *Arillus cristatus* (L.) and *Sinea spinipes* (Herrich-Schaeffer)), and *Conocephalus* spp. (see Table 1). *Halyomorpha halys* egg masses were acquired from a laboratory colony at the New Jersey Department of Agriculture Philip J. Alampi Beneficial Insect Laboratory in Trenton, NJ and nymphs were reared at RAREC using

**Table 1**  
Identity and life stages of taxa assessed for predation on *H. halys*.

Predator Taxa	Life Stage Tested
Acrididae	Adults
<i>Coccinella septempunctata</i>	Adults
<i>Coleomegilla maculata</i>	Adults
<i>Geocoris</i> spp.	Adults
<i>Harmonia axyridis</i>	Adults
<i>Hippodamia convergens</i>	Adults
<i>Nabis</i> spp.	Adults
<i>Podisus maculiventris</i>	Adults and Late Instar Nymphs
Reduviidae	Mixed <sup>1</sup>
Tettigoniidae	Adults

<sup>1</sup> Reduviidae consisted of a combination of *Sinea sinipes* adults and *Arillus cristatus* nymphs.



**Fig. 1.** Diagram of predation arenas. A. Hinged unpainted lid, shown in an open position, was closed and secured with masking tape during experiments, B. Vertical face, unpainted to allow observation of interactions inside the arena, C. exterior of all other vertical faces painted with opaque matte grey spray paint (illustrated as translucent for clarity), D. bottom surface with 8 cm opening for sunflower plants, grown from a small pot below each arena, E. two 3 cm ventilation holes covered with fine mesh. Note: although the sunflower is illustrated in full bloom, actual plants used in the present study were at the R2 stage of reproductive development.

organic carrot and green bean.

## 2.2. Predation arenas

Arenas used for predator assays were rectangular boxes constructed of 6 mm acrylic sheeting measuring 15 cm wide, 30 cm deep, and 30 cm tall (Fig. 1). A five cm hole was drilled into the bottom of each arena to allow for the insertion of a small sunflower plant. Sunflowers, *Helianthus annuus* L., are a known *H. halys* host plant and were selected because they grow easily in small pots (Soergel et al., 2015). Plants were grown from organic seed (var: grey striped sunflower, Johnny's Selected Seeds, Fairfield, ME) in individual 500 mL pots (seedling pads, grow light, room temperature and humidity) until the R1-R2 stage. Plants were used for multiple experiments, but were rinsed with water and searched for prey remains between runs. Sterile sand was spread across the bottom of each arena to simulate a natural substrate and to cover the soil. Two smaller 3 cm holes were drilled into one of the narrow vertical surfaces of the arenas and covered with window screening for ventilation. The top of each arena was covered by a hinged lid of acrylic sheeting sealed with masking tape to prevent insects from escaping. To deter insects from climbing the walls of the arenas, the base of each wall was painted with a five cm strip of fluon (Teflon PTFE 30, Dupont, Wilmington, DE). Matte grey spray paint was applied to three of the four walls to prevent activity outside the arenas from altering predator or prey behavior. The lid of each arena and one large vertical surface were left unpainted to allow light to enter the arenas (Fig. 1).

## 2.3. Assays

Each predator-life stage treatment was evaluated in two or three

temporal blocks each consisting of four replicates. Although a maximum of eight arena experiments could be conducted simultaneously, half-runs were occasionally conducted if predator species or prey stages were limiting. Predator treatments were selected each week based on availability and field abundance of predatory taxa in various crops at RAREC (listed above). After a predator treatment had been selected, ten individuals of this taxa were collected and starved for 48 h by providing only deionized water and a 10% sugar water solution. If fewer than eight individuals survived 48 h of starvation, this taxa was recollected and starved for only 24 h. Prey treatments consisted of 1) one egg mass consisting of ca. 28 *H. halys* eggs (< 48 h old), 2) approximately 28 *H. halys* first instar nymphs, or 3) five second instar nymphs. Egg masses and first instar nymphs aggregated on the hatched egg mass were affixed to the underside of a leaf of the sunflower plant with non-toxic craft glue (School Glue Gel, Elmer's, High Point, NC). Glue was applied only to the ovipositional substrate, not to eggs themselves. Second instar nymphs were transferred directly onto the leaves of the sunflower plant. In addition to the *H. halys* used as prey in the arenas, bugs of the same stage were stored in a small deli cup (30 mL, Solo Cup Company, Lake Forest, IL) within the arena containing moistened sand substrate and sealed with a paper lid to constitute negative controls. Prey were allowed to settle for 30 min prior to the introduction of a single predator in each arena. Throughout each trial, the sand substrate in the arenas and the cups containing the controls were misted with deionized water at least once a day and the sunflower plants were watered as needed. Aside from semi-daily misting, arenas were kept at ambient temperature, humidity and lighting (mixed fluorescent and natural light during daylight hours, half-intensity fluorescent at night). After 48 h, predators were removed and prey were assessed for mortality. Individual predators were used only once. Egg masses were assessed for signs of predation based on the symptoms identified by Morrison et al. (2016). Each predator-prey stage combination was repeated at least eight times, within two blocks of four simultaneously conducted assays. Replicates where the predator did not survive the 48 h assay were discarded (Tables 2–4).

## 2.4. Data analysis

A preliminary Welch two-sample *t*-test was performed to test for differences in prey survivorship between *A. cristatus* and *S. spinipes*, the two reduviid species in the experiment, to determine if these species could be considered and analyzed collectively as Reduviidae. To determine if predators caused a significant decrease in the survivorship of *H. halys* nymphs (or the hatch rate of *H. halys* eggs), Welch two-sample *t*-tests were performed. Arcsine-square root transformed proportion of survivorship (or proportion of hatch for egg masses) in treatment prey were compared to that of control prey for each predator and prey-stage combination. For each predator causing significantly lower survivorship than the control in at least one prey life stage, an analysis of variance (ANOVA) was used to test for significant differences in

**Table 2**

Mean ( $\pm$  SEM) effects of predators on *H. halys* egg hatch. *P* values were obtained from Welch two-sample *t*-tests; *P*-values in bold face were significant ( $P < 0.05$ ).

Predator ( <i>n</i> )	Treatment Hatch	Control Hatch	<i>P</i>
Acrididae (8)	0.549 ( $\pm$ 0.17)	0.957 ( $\pm$ 0.02)	<b>0.048</b>
<i>Coccinella septempunctata</i> (8)	0.886 ( $\pm$ 0.03)	0.978 ( $\pm$ 0.01)	<b>0.029</b>
<i>Coleomegilla maculata</i> (8)	0.867 ( $\pm$ 0.07)	0.772 ( $\pm$ 0.11)	0.480
<i>Geocoris</i> spp. (8)	0.572 ( $\pm$ 0.14)	0.472 ( $\pm$ 0.15)	0.633
<i>Harmonia axyridis</i> (8)	0.858 ( $\pm$ 0.12)	0.764 ( $\pm$ 0.10)	0.566
<i>Hippodamia convergens</i> (8)	0.655 ( $\pm$ 0.04)	0.797 ( $\pm$ 0.08)	0.152
<i>Nabis</i> spp. (8)	0.750 ( $\pm$ 0.10)	0.749 ( $\pm$ 0.09)	0.994
<i>Podisus maculiventris</i> adult (8)	0.581 ( $\pm$ 0.12)	0.879 ( $\pm$ 0.05)	<b>0.044</b>
<i>Podisus maculiventris</i> nymph (8)	0.699 ( $\pm$ 0.07)	0.917 ( $\pm$ 0.03)	<b>0.021</b>
Reduviidae (8)	0.901 ( $\pm$ 0.02)	0.875 ( $\pm$ 0.06)	0.677
Tettigoniidae (8)	0.241 ( $\pm$ 0.12)	0.709 ( $\pm$ 0.12)	<b>0.015</b>

**Table 3**

Mean ( $\pm$  SEM) survivorship of treatment and control *H. halys* first instar nymphs in various predator treatments. *P* values were obtained from Welch Two-Sample *t*-Tests; *P*-values in bold face were significant ( $P < 0.05$ ).

Predator (n)	Treatment Survivorship	Control Survivorship	<i>P</i>
Acrididae (8)	0.934 ( $\pm$ 0.02)	0.970 ( $\pm$ 0.02)	0.167
<i>Coccinella septempunctata</i> (8)	0.861 ( $\pm$ 0.07)	0.875 ( $\pm$ 0.02)	0.850
<i>Coleomegilla maculata</i> (8)	0.559 ( $\pm$ 0.14)	0.812 ( $\pm$ 0.05)	0.116
<i>Geocoris</i> spp (8)	0.894 ( $\pm$ 0.03)	0.956 ( $\pm$ 0.03)	0.138
<i>Harmonia axyridis</i> (8)	0.932 ( $\pm$ 0.03)	0.949 ( $\pm$ 0.03)	0.701
<i>Hippodamia convergens</i> (8)	0.823 ( $\pm$ 0.02)	0.855 ( $\pm$ 0.03)	0.423
<i>Nabis</i> spp (16)	0.869 ( $\pm$ 0.02)	0.967 ( $\pm$ 0.01)	<b>0.002</b>
<i>Podisus maculiventris</i> adult (8)	0.759 ( $\pm$ 0.10)	0.828 ( $\pm$ 0.07)	0.579
<i>Podisus maculiventris</i> nymph (8)	0.816 ( $\pm$ 0.09)	0.886 ( $\pm$ 0.03)	0.490
Reduviidae (8)	0.389 ( $\pm$ 0.13)	0.733 ( $\pm$ 0.04)	<b>0.031</b>
Tettigoniidae (7)	0.980 ( $\pm$ 0.01)	0.983 ( $\pm$ 0.01)	0.885

**Table 4**

Mean ( $\pm$  SEM) survivorship of treatment and control *H. halys* second instar nymphs in various predator treatments. *P* values were obtained from Welch two-sample *t*-tests; *P*-values in bold face were significant ( $P < 0.05$ ).

Predator	Treatment Survivorship	Control Survivorship	<i>P</i>
Acrididae (8)	0.819 ( $\pm$ 0.06)	0.825 ( $\pm$ 0.07)	0.947
<i>Coccinella septempunctata</i> (8)	0.725 ( $\pm$ 0.09)	0.850 ( $\pm$ 0.06)	0.283
<i>Coleomegilla maculata</i> (7)	0.800 ( $\pm$ 0.08)	0.914 ( $\pm$ 0.06)	0.259
<i>Geocoris</i> spp (8)	0.588 ( $\pm$ 0.14)	0.875 ( $\pm$ 0.05)	0.094
<i>Harmonia axyridis</i> (8)	0.875 ( $\pm$ 0.08)	0.825 ( $\pm$ 0.07)	0.634
<i>Hippodamia convergens</i> (8)	0.708 ( $\pm$ 0.06)	0.775 ( $\pm$ 0.08)	0.526
<i>Nabis</i> spp (8)	0.383 ( $\pm$ 0.09)	0.866 ( $\pm$ 0.05)	<b>&lt; 0.001</b>
<i>Podisus maculiventris</i> adult (8)	0.900 ( $\pm$ 0.04)	0.900 ( $\pm$ 0.04)	1
<i>Podisus maculiventris</i> nymph (8)	0.500 ( $\pm$ 0.08)	0.900 ( $\pm$ 0.04)	<b>0.001</b>
Reduviidae (8)	0.700 ( $\pm$ 0.13)	0.875 ( $\pm$ 0.05)	0.229
Tettigoniidae (8)	0.775 ( $\pm$ 0.05)	0.825 ( $\pm$ 0.06)	0.513

survivorship among prey life stages. First, prey survivorship in treatment groups was adjusted by the survivorship of the control group using Abbott’s Formula (Abbott, 1925). Next, a one-way ANOVA was performed for each predator to test for significant differences in corrected survivorship between prey life stages. Tukey’s HSD method was used for means separation in a post hoc analysis. Differences in survivorship means were considered significant at  $\alpha = 0.05$ . All data analyses were performed in R Studio v3.2.2 (“Fire Safety”).

### 3. Results

#### 3.1. Predation on eggs and nymphs

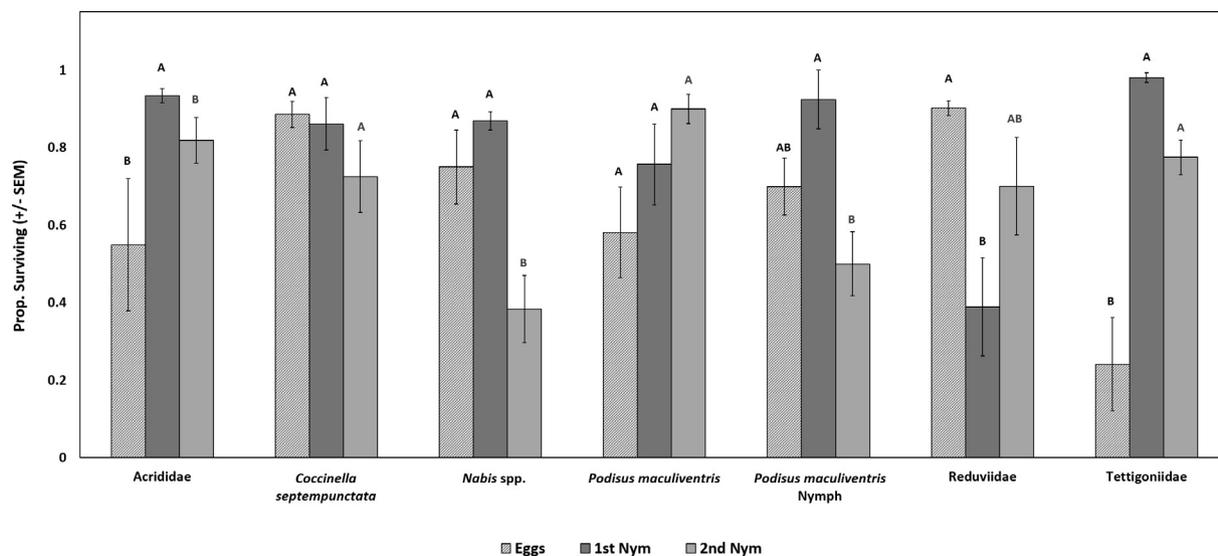
We detected no statistically significant difference in *H. halys* survivorship between *A. cristatus* and *S. spinipes* so these species were combined for subsequent analyses and are referred to simply as Reduviidae ( $t = 0.39$ ,  $P = 0.627$ ). Independent Welch *t*-tests revealed significantly lower hatch rate when egg masses were exposed to Acrididae ( $t = 2.379$ ,  $P = 0.0483$ ), *C. septempunctata* ( $t = 2.658$ ,  $P = 0.0287$ ), *P. maculiventris* adults ( $t = 2.313$ ,  $P = 0.0436$ ), *P. maculiventris* nymphs ( $t = 2.789$ ,  $P = 0.0215$ ), and Tettigoniidae ( $t = 2.788$ ,  $P = 0.0145$ ) (Table 2). When exposed to acridids and tettigoniids, 40.8% and 46.8% fewer *H. halys* eggs hatched, respectively, compared to unexposed controls (Table 2).

Survival of *H. halys* first instar nymphs was significantly lower than that of the controls when exposed to *Nabis* spp. ( $t = 3.609$ ,  $P = 0.0015$ ) and Reduviidae ( $t = 2.568$ ,  $P = 0.0311$ ) (Table 3). Reduviidae caused a 34.3% reduction in survivorship, whereas *Nabis* spp. reduced survivorship by 9.7%.

Only *Nabis* spp. ( $t = 4.94$ ,  $P = 0.0001$ ) and *P. maculiventris* nymphs ( $t = 4.413$ ,  $P = 0.0014$ ) reduced the survivorship of second instar nymphs compared to controls (Table 4), lowering it by 48.3% and 40%, respectively.

#### 3.2. Survivorship of prey life stages

Life stage-specific differences in *H. halys* survivorship were significant for *Nabis* spp. ( $F = 13.72$ ;  $df = 2,33$ ;  $P < 0.001$ ), Acrididae ( $F = 4.382$ ;  $df = 2,21$ ;  $P = 0.0261$ ), *P. maculiventris* nymphs ( $F = 3.967$ ,  $df = 2,21$ ;  $P = 0.0352$ ), Reduviidae ( $F = 3.698$ ;  $df = 2,21$ ;  $P = 0.0425$ ) and Tettigoniidae ( $F = 11.54$ ;  $df = 2, 20$ ;  $P < 0.001$ ). *Nabis* spp. and *P. maculiventris* nymphs significantly



**Fig. 2.** Mean ( $\pm$  SE) survivorship of *H. halys* eggs, first instar nymphs, and second instar nymphs following a 48 h exposure to a single predator of various taxa on a sunflower plant in a predation arena. Columns bearing different letters were significantly different within a taxon (ANOVA followed by Tukey’s HSD,  $P < 0.05$ ). Analysis was conducted on control-corrected survivorship, but uncorrected values are shown.

reduced survivorship of second instar nymphs more than that of first instar nymphs, whereas Acrididae and Tettigoniidae reduced egg hatch more than the survivorship of either first or second instar nymphs (Fig. 2). Reduviidae reduced first instar nymph survivorship more than the hatch rate of eggs, but preyed on first and second instar nymphs equally (Fig. 2).

#### 4. Discussion

The differences we observed among taxa in predation on various *H. halys* life stages is consistent with the findings of previous research on predators affecting other North American Pentatomidae. The predator guild consuming eggs of *N. viridula*, *Euschistus servus* (Say), *E. variolarius* (P. De B.), *Chinavia hilare* (Say), and *P. maculiventris* were primarily those with chewing mouthparts (Koppel et al., 2009; Ragsdale et al., 1981; Yeargan, 1979). In addition to chewing predators, *P. maculiventris* was identified as a predator of *N. viridula* eggs, consistent with the present study (Ragsdale et al., 1981). Similarly, *N. viridula* and *H. halys* nymphs were most commonly preyed upon by sucking predators, including Reduviidae and *P. maculiventris* and markedly different predator communities were shown to feed on the eggs and nymphs of *N. viridula* (Ragsdale et al., 1981).

We observed significant differences in survivorship between first and second instar *H. halys* nymphs exposed to *Nabis* spp. and *P. maculiventris* nymphs. Variation in predation between nymphal instars may be explained by stage-specific differences in defensive compounds. Pentatomidae are known for the odorous compounds they release when disturbed (Aldrich, 1988) and these compounds can vary with life stage (Borges and Aldrich, 1992). Members of the subfamily Pentatominae excrete (E)-4-oxo-2-decenal when agitated, but only during the first instar (Borges and Aldrich, 1992). In other species this compound is emitted by all nymphal instars but is notably absent in the secretions of adults (Pareja et al., 2007). The chemical composition of *H. halys* nymphal secretions has not been thoroughly studied, but variation in these compounds could lead to the observed variation in predation on different nymphal instars.

Our work is one of several recent studies attempting to identify the native natural enemies affecting *H. halys* eggs in agricultural settings. Visitors to *H. halys* sentinel egg masses in agricultural settings in Michigan and New Jersey were identified using closed circuit video cameras (Poley et al. unpublished) and included members of Acrididae, Anthracoridae, Araneae, Coccinellidae, Forficulidae, Gryllidae, Miridae, Parasitica, Pentatomidae, and Tettigoniidae. Many of these also fed on *H. halys* eggs in the present study. Our findings confirm the findings of earlier Petri dish assays in which Tettigoniidae were among the predator taxa feeding most frequently on *H. halys* eggs (Morrison et al., 2016). Abram et al. (2014) also tested the acceptance of *H. halys* eggs by several endemic generalist predators and, as in the current study, *C. maculata* proved to be a poor predator of *H. halys* eggs, with adults accepting them in less than seven percent of trials and consuming few of them (< 0.1 egg per predator in 24 h).

Many studies attempting to identify or quantify predation on phytophagous Pentatomidae have focused on the egg stage (e.g. Ogburn et al., 2016; Yeargan, 1979) but the present results broaden our understanding of *H. halys* natural enemies by identifying predation on nymphal stages. In the present study, three taxa were confirmed as predators of nymphs (*P. maculiventris*, *Nabis* spp., and Reduviidae). Due to the observed preference of hemipteran predators for the nymph stage of *H. halys*, it is possible these 'sucking' predators may attack the nymphs of other members of Pentatomidae. Our findings suggest that sentinel egg mass studies alone underestimate the role of predation in the population dynamics of *H. halys*, as well as other pentatomid pests.

Orthopteran omnivores readily attacked the eggs of *H. halys* but, unlike other chewing predators, consumed the entire egg mass leaving no trace of their activity (Morrison et al., 2016). Studies using sentinel egg masses to quantify predation may underestimate predation by

tallying eggs consumed by orthopterans as lost or missing. Missing egg masses accounted for 9.7–12.8% of sentinel eggs in the survey by Ogburn et al. (2016) and 37% of egg predation observed in peach orchards was consistent with feeding by Orthoptera (Morrison et al., 2016). In Southeast Asia, Orthoptera regularly consume the eggs of many rice pests and are considered effective natural enemies in these cropping systems (Chitra et al., 2002). Orthopteran omnivores are abundant in many cropping systems affected by *H. halys*, but it is unclear whether the consumption of *H. halys* eggs is supplemental, coincidental or actually preferred over plant matter.

Variation between the predator complex attacking *H. halys* and other native or less recently invasive Pentatomidae may be partially explained by the 'enemy release hypothesis' (ERH). According to the predictions of the ERH, *H. halys* arrived in the US without its coevolved natural enemies and is outcompeting native species as a result. Although the mechanism for the invasive success of *H. halys* is unclear, native predators may be maladapted to specific behavioral or chemical defenses of *H. halys*, allowing it to thrive in its invaded range (Keane and Crawley, 2002). Although Pentatomidae employ defensive secretions to avoid predation, the chemical composition of these may vary widely among local and invasive species. Invasive plant species often produce novel defensive compounds not found in native species and, as a result, experience less herbivory than non-invasive species (Cappuccino and Arnason, 2006; Cappuccino and Carpenter, 2005). Similarly, invasive *Harmonia axyridis* eggs are chemically defended from predation while eggs of native species of coccinellids are more prone to predation and cannibalism (Cottrell, 2004). However, the primary defensive compound emitted by *H. halys*, (E)-2-decenal, is a component of the secretions of at least five species of New World Pentatomidae including *N. viridula* (Borges and Aldrich, 1992; Harris et al., 2015). Although this compound is common throughout Pentatomidae, further study might determine if the secretions of *H. halys* and indigenous pentatomids vary in minor components. Direct comparisons of defensive secretions and other anti-predator behaviors between exotic and native Pentatomidae are needed to test the predictions of the ERH and to clarify the role of predators in the success of *H. halys* in its invaded range.

Although the ecological root of this phenomenon remains unclear, *H. halys* is outcompeting and displacing native pentatomids in agroecosystems across the Eastern US. Evidence in ornamental crops and soybean suggests that *H. halys* has become the most abundant pentatomid species in these systems, comprising over 50% of all Pentatomidae in many locations (Bakken et al., 2015; Nielsen and Hamilton, 2009a). Although the current utilization of *H. halys* by natural enemies is relatively low, the high relative abundance of this pest may change local predator preferences and adaptations over time (Carlsson et al., 2009; Carlsson and Strayer, 2009). High *H. halys* abundance in the absence of competitors could create strong selection pressure for predators able to consume this abundant, but underutilized, food resource. The enhanced fitness of individuals that successfully utilize *H. halys* as prey may drive dietary shifts toward more *H. halys* consumption (Carlsson et al., 2009; Elliott, 2004; Jaworski et al., 2013). Continuing surveys of natural enemy impacts on *H. halys* could reveal dietary shifts and how these affect overall pest mortality and abundance.

The absence of a single predator capable of competently attacking all life stages of *H. halys* implies that biological control of this pest will only be provided by a community of predators, not by an individual species. However, the lack of any highly effective single natural enemy will affect the potential roles of biological control in pest management. For example, augmentative releases or chemical attractants for a single predator species are unlikely to aid in the suppression *H. halys* populations in agricultural settings. However, the predator community may be enhanced through cultural tactics such as conservation biological control, use of trap crops, or intercropping with predator-attracting crops (Blaauw and Isaacs, 2012; Nielsen et al., 2016; Soergel et al.,

2015). Additionally, the responsible usage of chemical control methods, such as border-only spray programs, may further protect natural enemies in the crop interior while simultaneously suppressing *H. halys* at the crop edge where they are most abundant (Blaauw et al., 2015).

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