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Short Communication

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Cavity Tightness Preferences of Overwintering *Halyomorpha halys* (Hemiptera: Pentatomidae)

Benjamin D. Chambers,[1,4] Tracy C. Leskey,[2] John P. Cullum,[1] Annie R. Pearce,[3] and Thomas P. Kuhar[1]

[1]Department of Entomology, Virginia Polytechnic Institute and State University, Price Hall, Room 216A, Virginia Tech, 170 Drillfield Drive, Blacksburg, VA 24061, [2]Appalachian Fruit Research Station, US Department of Agriculture, 2217 Wiltshire Road, Kearneysville, WV 25430, [3]Department of Building Construction, Virginia Tech, Bishop Favrao Hall, Room 330A, 1345 Perry Street, Blacksburg, VA 24061, and [4]Corresponding author, e-mail: bdc0112@vt.edu

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Abstract

Brown marmorated stink bug (*Halyomorpha halys*) (Stål) is a household nuisance pest that seeks shelter in buildings during the winter months. It has been found in a variety of cavities and spaces between building elements, as well as in the objects stored within buildings. This experiment examined the cavity tightness preferences for these insects as they settled in winter refugia. Adult overwintering *H. halys* were placed in two types of simulated refugia made from rigid material. Each type had a cavity of constant width, while one had a flat lid and constant tightness, and the other had a sloped lid that became tighter as insects moved inside. Adults were allowed to enter and settle, then their locations were recorded. In sloped lid cavities, *H. halys* tended to settle where the cavity tightness was between 4.5 and 5.5 mm. In the flat lid cavity boxes, *H. halys* tended to move all the way back. In both configurations, *H. halys* had a significant tendency to orient their heads towards the cavity entrance. A field comparison of cavity tightness in refugia with less rigid cardboard substrates was also performed, with spacers consisting of one or two layers of 3-mm cardboard. This comparison found differences in cavity selection by sex, with males more likely to pick single-spaced layers, and females more likely to select double-spaced layers. Understanding these preferences could be useful for collection, pest management, trap design, and study of impacts on structures.

Key words: diapause, home invasion, brown marmorated stink bug, urban pest, building envelope

Brown marmorated stink bug, *Halyomorpha halys* (Stål), is a major agricultural pest (Leskey and Nielsen 2018). It is also a major nuisance pest in homes due to its tendency to aggregate on exteriors each fall before entering in search of winter shelter, in numbers as high as tens of thousands (Inkley 2012). Observations of overwintering aggregations suggest that *H. halys* prefer tight spaces (Lee et al. 2014). However, rigid openings exclude most females at 4 mm and most males at 3 mm (Chambers et al. 2019a). The cloth, tarps, and wood piles from which they can be collected have varying and/or flexible opening and cavity sizes. The cavities in old buildings in which they shelter may also vary as buildings settle and warp with moisture and age.

Several kinds of overwintering shelters have been deployed to capture or store *H. halys* for research and management. Taylor et al. (2017) built shelters with a substrate of plastic foundation sheets spaced 1 cm apart, and shelters with substrates of paper towels and corn–starch based packing peanuts. Cira et al. (2016) used 18.9-liter buckets filled with 12.7-mm thick foam pipe insulation for insect storage. A slit trap shelter, used by Watanabe et al. (1994a, b), was a 90- × 180-cm rectangular panel leaning at a 30° angle, with 10-mm thick veneer panels, and three layers of 3-mm slits, though some details are unclear in the available translation. Bergh et al. (2017) described two overwintering shelters for *H. halys*. In one, opaque plastic containers contained concentric tubes of rolled 4.8-mm thick cardboard sheets, with a 5-mm space between layers, and extra space between tubes and outer walls. Insects were placed directly inside. The second design was used for fall collection. It consisted of a plywood box similar to a birdhouse, containing a stack of 3-mm thick cardboard sheets, spaced with strips of 3-mm cardboard, creating 3-mm high cavities, though those varied due to deformations in the cardboard. Spaces between sheets and the outer walls were 13 or 25 mm. This inexpensive and easily built wooden box design has been used by researchers in the mid-Atlantic United States for several years, including the relatively flexible cardboard substrate. However, if this 3-mm spacing were rigid, it would exclude most males and all females (Chambers et al. 2019a), which suggests questions about the tightness preferences of these insects, and optimal substrate spacing.

Understanding tightness preferences could be useful in trap design for collection or pest management, and also to study the impacts of *H. halys* aggregations on structures. The work presented here examines cavity tightness preferences of *H. halys* using rigid materials, and also examines the influence of cardboard spacing in shelters used for field collection.

Methods

Rigid Cavity Laboratory Test Subjects

All insects used in rigid cavity experiments were overwintering *H. halys* adults collected in southwest Virginia in late 2016 from the sides of houses during fall and from overwintering clusters in human structures during winter. Insects were kept in containers full of paper and foam pipe insulation in darkness at 10°C until the time of the experiment. No insects were reused.

Rigid Cavity Laboratory Apparatus

Rigid cavity boxes (Fig. 1) were laser cut from 3.2-mm hardboard and 1.6-mm acrylic. Acrylic windows were created to simulate a cavity entrance while preventing insect escape. All pieces were designed to interlock, with smooth sides facing inward. Boxes were 300 mm wide × 600 mm long, and floor-to-lid spacing at the windows was 9.5 mm. Sloped lid boxes grew tighter as insects went further inside, as floor-to-lid spacing ran from 9.5 to 0 mm, with a slope of 0.0192, over a 495-mm length. Flat lid box cavities had 9.5-mm floor-to-lid spacing over a 495-mm length. Each box was lined with plain white paper. Ceramic tiles were placed atop boxes to prevent warping. Boxes were laid flat on a table, so insect movement was horizontal.

Rigid Cavity Laboratory Procedure

Rigid cavity experiments were conducted with 16 replications for each cavity type, with 10 adult *H. halys* per replicate of mixed sex, for an expected n = 160 bugs for each cavity type. Ten bugs were used in a sample to best simulate natural conditions in which bugs aggregate in their overwintering shelter sites (Toyama et al. 2006, Inkley 2012, Chambers et al. 2019b). Four boxes of each type at a time were arranged on a table in a room kept at 15°C and 65% relative humidity, with a single overhead light, holding a compact fluorescent bulb (CF13EL/MICRO/865/BL2 6500K, OSRAM Sylvania, Wilmington, MA). Boxes were wiped with a damp cloth and given at least 24 h to air out between trials, after which fresh paper was inserted.

For each flat or sloped lid box, 10 adult *H. halys* were removed from an overwintering shelter and chilled in a freezer at −15°C for 4 min to facilitate placement. The insects were then placed right-side-up in the window section, and acrylic windows were replaced. After 24 h, lids were carefully lifted, and photographs were taken of box floors and the undersides of lids. Ruling lines were drawn on the images in intervals of 12.7 mm using the regularly spaced interlocking tabs as a guide.

For each individual in sloped lid cavities, the furthest ruling line from the release point where at least 50% of the pronotum was past was recorded. The location (lid or floor) and direction the insect was facing were also recorded. Cavity tightness at pronotum location was determined from the ruling lines, and analyzed. For flat lid cavities, ruling lines were also 12.7 mm apart. Bug locations were noted, including distance into the refuge, and location (lid or floor).

Fig. 1. *Halyomorpha halys* adults in the experimental (left) sloped lid box from above and from the side, and (right) flat lid box from above and from the side.

Cardboard Spacer Field Experiment

To examine the impact of cavity tightness on settling behavior of *H. halys* in the field with less rigid cardboard substrates, shelters with different substrate spacing were deployed. Experimental overwintering shelters, built to specifications described by Bergh et al. (2017), consisted of a 19 × 22 × 24 cm box constructed from 0.6-cm-thick plywood, with access to *H. halys* available through both an open bottom and a 1-cm opening in the top (Fig. 2). Each shelter contained cardboard inserts (0.3 × 17.8 × 21.6 cm). Inserts were oriented vertically and held in place by lips on the interior of the shelter. Standard cavity spacing shelters contained 16 cardboard sheets with spacers glued to each insert to create a 3-mm cavity between each insert. The double-spaced shelters contained eight cardboard inserts, each with two of these spacers stacked, resulting in a 6-mm cavity between each insert.

The deployment sites were Price Farms in Keedysville, MD (N39°030′17.13″ W77044′35.6″), a private residence in Sharpsburg, MD (N39°029′8.4″ W77066′1.9) and a private residence in Arden, WV (N39°024′50.1″ W7801′44.3″). All sites utilized lumber storage sheds with an open side and a history of *H. halys* pressure. Each replicate included two shelters screwed to an interior wall at 2-m height, about 1 cm apart. One had cavities created by single cardboard strip spacers, and one had double strip spacers. Interior walls were selected based on previous observations of aggregations of dispersing *H. halys* adults. Shelters were deployed in early September 2017 before dispersal. All *H. halys* adults inside each shelter were removed and counted after dispersal in early November 2017.

Analysis

Results were analyzed using JMP Pro 14 statistical software (SAS Institute 2018). For each laboratory experiment, separate chi-square tests were performed with an expected value of 50% for orientation to the window or back of the box, and location on lid or floor. ANOVA tests were used to check for effects of trial and box. For field trials, likelihood ratio chi-square tests with a Poisson distribution were used to check for effect of sex, location, spacing, and sex crossed with spacing.

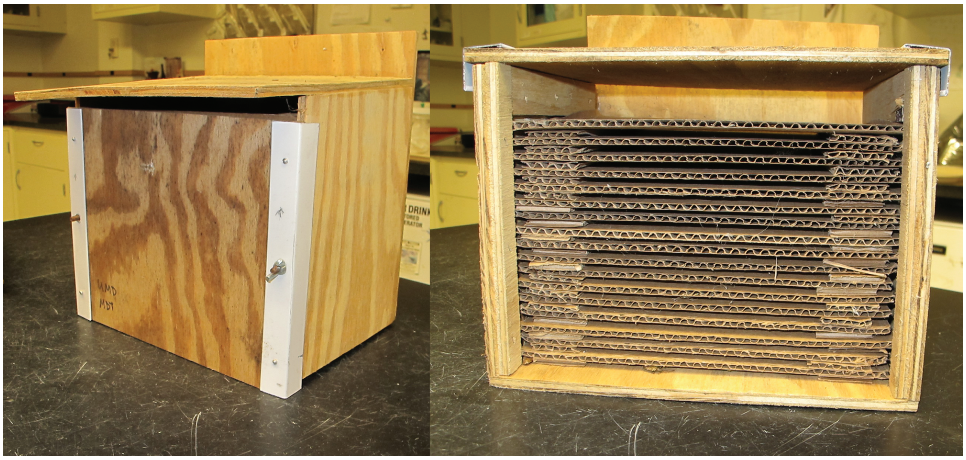

Fig. 2. Cardboard spacer field box (left) from the side, and (right) from below with single-spaced cardboard.

Table 1. Facing and locations for *H. halys* adults in rigid refuges by box type (df = 1 for all tests)

	Sloped lid	Flat lid
Facing entrance	101	92
Facing back	39	43
χ^2	28.4	18.2
P	<0.0001	<0.0001
On floor	129	42
On lid	11	93
χ^2	117.0	19.8
P	<0.0001	<0.0001
Total	140	135

Results

Rigid Cavity Laboratory Results

Although 160 *H. halys* adults were included in each rigid cavity configuration experiment, some died, remained in the window area, or escaped when the lid was removed. These individuals were excluded, leaving n = 134 insects in flat lid cavities and n = 140 insects in sloped lid cavities. In both cavity types, insects oriented their heads towards the entrance in proportions significantly higher than random orientation (Table 1). Insects were also much more likely to be on the floor than on the lid of the box, in proportions significantly higher than a random expected 50% in both cases.

In the sloped lid cavities, the majority of *H. halys* (about 68%) settled where the tightness of the cavity was between 4.5 and 5.5 mm at pronotum, with a mean ± SEM of 5.33 ± 0.0936 mm, and a tightness range of 9.50 to 3.66 mm (Fig. 3). In flat lid boxes, *H. halys* settled towards the back (338.7 ± 14.09 mm), with 37% settling in the last 12.7-mm zone (Fig. 4). There was no evidence of effect of trial with either lid type, nor of box with sloped lids. However, there was evidence of effect of box in the flat lid type (F = 7.17, df = 3, P = 0.0002), with the mean location of insects being closer to the deployment zone in one box.

Cardboard Spacer Field Results

In the field trials with cardboard substrates, *H. halys* were successfully captured (n = 2,426) (Table 2). There was a difference in sex of insect captured (χ^2 = 455.23, df = 1, P < 0.0001), with more than twice as many males captured as females. Results show an effect of spacing (χ^2 = 4.86, df = 1, P = 0.0275), with more insects selecting single-spaced shelters. This is likely related to the disproportion of sex, and indeed an interaction is shown between sex and spacing (χ^2 = 59.08, df = 1, P < 0.0001), with males preferring single-spaced substrate and females preferring double-spaced. A location effect is

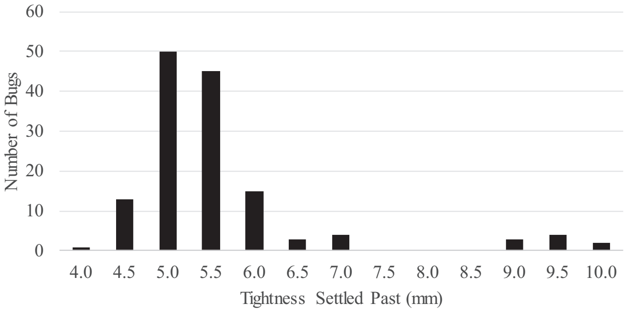

Fig. 3. Tightness below which *H. halys* settled in sloped lid boxes.

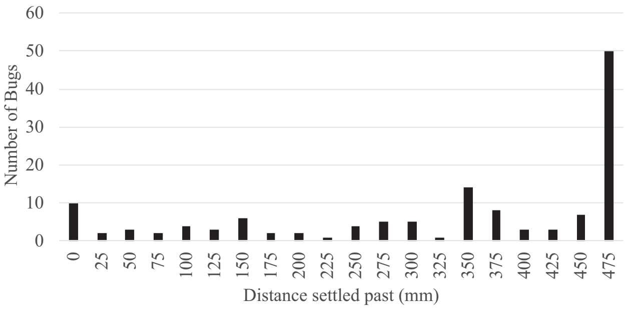

Fig. 4. Distance past which *H. halys* settled in flat lid boxes.

Table 2. Cardboard substrate spacing selections of *H. halys* adults settling in field tests

	Single-spaced	Double-spaced	Total
Male	1,068	680	1,748
Female	297	381	678
Combined	1,365	1,061	2,426

shown (χ^2 =16.62, df = 1, P = 0.0002), as one of the sites resulted in about 20% more captures than the others.

Discussion

In the sloped lid cavities, *H. halys* moved back into spaces not much larger than dorsoventrally limiting dimensions (Chambers et al. 2019a). Therefore, we conclude that *H. halys* exhibits dorso-ventral thigmotaxis during winter shelter seeking. We also conclude that *H. halys* tends to move as far as possible into a cavity of this size in the absence of dorso-ventral restriction. In the flat lid cavities, insects appeared to also aggregate along edges, even if they did not move all the way back. We also note an unintended result that the settled insects tended to be oriented towards the entrance, and suggest this as a topic for future research.

As previously noted, several experiments have already utilized overwintering boxes containing cardboard substrate, with cavities as tall as the cardboard is thick created by alternating sheets and strips. However, most commonly available kinds of corrugated cardboard are 4 mm thick or less (Twede et al. 2014), and the cardboard used in previous experiments was 3 mm thick (Bergh et al. 2017). This material is less rigid than the hardboard in our laboratory experiments and prone to warping with moisture, but the 3-mm spacing it creates is less than the preferred tightness found in the laboratory experiment, and less than the exclusionary rigid dimensions (Chambers et al. 2019a). We conclude that shelters with a single-spaced cardboard substrate of 3 mm are sufficient for capturing *H. halys*, and may be preferable to males. However, our results suggest that larger spacing is preferable to females, and that insert spacing could potentially affect the proportion of *H. halys* captured by sex. These results may, therefore, be of interest for future trap design for collection, pest management, and research purposes.

Acknowledgments

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