Behavior of and Crop Injury Induced by Native and Exotic Stink Bugs in Mid-Atlantic Soybean

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ABSTRACT

Cage studies were performed to determine if the current thresholds for stink bugs in soybean (one bug per 0.3 row m) need to be adjusted based on current soybean production practices and species present. Several soybean development stages were infested by two native stink bug species for three weeks using small cages in a field of double crop soybean at sites in Virginia, Maryland, and Delaware during 2007-2009. Large field cages were infested by an introduced species for two weeks in 2010-2011. Results showed that *Euschistus servus* Say and *Acrosternum hilare* Say adults or nymphs did not cause different levels of injury to soybean seed quality or effects on yield. Both *A. hilare* and the introduced *Halyomorpha halys* Stål injured soybean seed in a similar fashion at threshold-level densities. Full flowering R2 stage soybean were least affected by stink bug feeding, and full pod and beginning seed R4-R5 stage soybean were slightly more sensitive to injury than R6 although not at the Maryland 2011 site. Several sites had increased seed injury and decreased yield at threshold density populations.

Finally, visual observations of stink bug vertical distribution inside soybean canopies were taken several times per day and compared with ambient and within-canopy temperature and relative humidity. Results indicated that these conditions did not influence the percentage of stink bugs below the top 38 cm sweep net intercept zone. In both years of observations, between 15 and 20% of stink bugs were observed below the 38 cm sweep net zone.

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Introduction

Stink bugs are one of the most important insect pests of soybean grown in the South and mid-Atlantic states. They can cause reductions in soybean seed quality and yield as well as delaying plant maturity. An economic threshold of one stink bug per 0.3 row m that was created in the late 1960's through the 1970's is still being used in many states. Thresholds are applied to the combined members of the phytophagous stink bug complex in soybean. However, both the soybean growing practices and stink bug species complex in soybean fields have changed. In the mid-Atlantic U.S., the invasive brown marmorated stink bug, *Halyomorpha halys* Stål, has become an important stink bug species in the complex.

A soybean field must be scouted in order to determine if insect pests are present at population levels that merit control. Sweeping soybean fields with a 38 cm diameter sweep net is the most common technique for scouting fields. The net is swung either across or between rows, passing through the upper 38- 42 cm. Little research has been done to determine whether or not stink bugs change their vertical distribution on soybean plants, and what environmental influences may affect this movement. If stink bugs move below this 38-42 cm upper canopy level, then a sweep net will not intercept them.

The goal of this research was to determine if current thresholds for stink bugs in soybean need to be adjusted, and if sampling techniques are adequate under fluctuating climatic conditions. There were three objectives:

1. Analyze data from previous field cage studies to evaluate the impact of native stink bug species' feeding on soybean seed quality and yield in mid-Atlantic soybean.
2. Examine seed and pod injury and damage potential of the exotic brown marmorated stink bug.

3. Examine the influence of temperature and relative humidity on the diurnal vertical distribution of stink bugs in the soybean canopy.

Chapter 1

Literature Review

Stink bugs (Hemiptera: Pentatomidae) have received increasing attention as major pests in field crop ecosystems in the United States. Sporadic stink bug injury to crops has been recorded since 1855 (Underhill 1934). Stink bug damage to crops as diverse as peaches and lima beans has been recorded since 1911 and 1889, respectively (Whitmarsh 1914, Underhill 1934). During previous decades, stink bug populations were controlled by frequent broad-spectrum insecticide applications that targeted other insect pests such as the corn earworm, *Helicoverpa zea* Boddie (Lepidoptera: Noctuidae), the tobacco budworm, *Heliothis virescens* Fabricius (Lepidoptera: Noctuidae), and the cotton boll weevil, *Anthonomus grandis* Boheman (Coleoptera: Curculionidae) (McPherson and McPherson 2000). During the late 1970's and 1980's, cotton was repeatedly sprayed with insecticides, mostly pyrethroids and organophosphates, in an attempt to eradicate the boll weevil. Due to the success of the boll weevil eradication program and the widespread adoption of Bt-transgenic crops, the number of broad-spectrum insecticide applications has decreased significantly (Lu et al. 2010). Furthermore, in other crops, there has been increased adoption of more narrow-spectrum (typically Lepidoptera-specific) insecticides that do not control stink bugs, further reducing the number of broad-spectrum applications that stink bugs are exposed to (McPherson and McPherson 2000). In this reduced broad-spectrum insecticide application environment in many field and vegetable crops, native stink bug populations and their damage has increased (McPherson and McPherson 2000).

Stink bugs in soybean

Soybean production has greatly increased in the southern U.S. over the last several decades, both in terms of yield and acreage. In Virginia, the highest acreage of field crops grown in the state is soybean, with over 231,000 hectares (USDA/NASS 2009), whereas in 1969, soybean acreage was estimated at 123,000 hectares (Plotkin et al. 1977). Most of the soybean acreage in the state occurs in the eastern region. Cash receipts for Virginia soybean in 2009 was estimated at 130 million dollars, and was the second most valuable crop, behind corn (USDA/NASS 2009).

Stink bugs, among other insects, have become important pests of soybean (Newsom et al. 1980). In the southern U.S., soybean is an attractive late season crop that allows for several stink bug species to produce additional generations and to increase food stores in preparation for overwintering (Jones and Sullivan 1982). These higher populations in the southern U.S. have increased their pest status, especially in soybean and cotton (Musser et al. 2010, Bacheler et al. 2010).

Stink bugs are attracted to soybean in high numbers when the plants begin producing pods (Thomas et al. 1974). The flowering stage is called R2, beginning pod is R3, full pod is R4, beginning seed is R5, full seed is R6, senescence is R7, and harvest maturity is R8 (Fehr et al. 1971). Stink bug pod feeding injury is independent of other types of soybean plant injury. Defoliation and stink bug feeding have an additive effect on soybean injury. Below threshold populations of defoliators and stink bugs may still need to be controlled. For instance, if defoliator populations reach half of their threshold, and stink bug populations reach half of their threshold, than the sum of the population proportions relative to their individual thresholds equal one, and thus warrant control (Simmons and Yeargan 1990).

Important members of the phytophagous stink bug complex that feed in soybean are the green stink bug, *Acrosternum hilare* Say (also *Chinavia hilaris*), southern green stink bug, *Nezara viridula* L., the brown stink bug, *Euschistus servus* Say (and other *Euschistus* spp.), the red banded stink bug, *Piezodorus guildinii* Westwood, and *Thyanta* spp. In several mid-Atlantic states, the recently introduced brown marmorated stink bug, *Halyomorpha halys* Stål, is becoming increasingly abundant (Nielsen and Hamilton 2009), whereas *N. viridula* and *P. guildinii* occur much farther south (Kamminga 2008). Stink bug feeding on soybean can cause reductions in seed quality and yield with densities of four bugs per row m (Daugherty et al. 1964) and maturity delays with variable densities (Daugherty et al. 1964, Boethel et al. 2000). This maturity delay is a result of the soybean plants' compensation for injury. Severely affected soybean plants redirect photosynthate from developing pods to increasing vegetative growth in an effort to increase flower and pod production until frost, long after the rest of the field has senesced (Daugherty et al. 1964, Russin et al. 1987, Boethel et al. 2000). Attempts to better understand this maturity delay caused by stink bugs indicated that season long average stink bug populations of 3.1 bugs per row m can result in maturity delays (Russin et al. 1987). Short two-week infestations of high densities (19.7 bugs per row m) of *N. viridula* consistently resulted in maturity delays, especially in the R3-R5 soybean development stages. Even densities of one stink bug per 0.3 row m feeding for a short period of time can cause some delay, although not consistently (Boethel et al. 2000). When faced with areas of the field that have not senesced normally, farmers will either postpone the harvest or forgo harvesting severely affected parts of the field (Daugherty et al. 1964).

Stink bugs insert their stylets into above-ground plant tissues, but preferentially feed on pods (McPherson and McPherson 2000). Feeding duration lasts between 70 and 130 minutes per

feeding session, depending on species (Depieri and Panizzi 2011). Cells are initially destroyed by the act of stylet insertion, and salivary enzymes spread several millimeters from the stylet. Enzymes disrupt cell walls and dissolve proteinaceous contents (Depieri and Panizzi 2011). These internally damaged areas have a white, chalky appearance, and are identifiable externally by a dark spot where the stylets were inserted (Miner 1966). Severely damaged seeds have lower oil content and higher protein content than less damaged seed, and this effect may be further exacerbated by delayed maturity (Miner 1961, Daugherty et al. 1964, Miner 1966). Seeds may be destroyed early in their development, resulting in un-filled pods (Corrêa-Ferreira and de Azevado 2002). Stink bug feeding on maturing seeds often results in shriveled and wrinkled seed coats that may be fused to the cotyledon (Miner 1966). Longer feeding times result in greater damaged to cells. Different stink bug species also produce different amounts of saliva and there may be chemical compositional differences in the saliva among species (Depieri and Panizzi 2011).

Stink bug injury to seed is one factor reducing the value of harvested soybean (Miner 1961), and much research has been done to determine what infestation levels are required to injure a significant portion of seed and at what soybean development stages. Seed injury is least tolerated by buyers in the seed and export markets (Miner 1961). Stink bug injury can also lower the price received for soybean grown for grain because the injury can alter the chemical composition of the seed. Discounts are assessed once stink bugs damage two percent of the soybean seed. Discounts increase with the severity of damage (Musser et al. 2011). Russin et al. (1987) found significant increases in damaged seed percentages and significant reductions of yield adjusted for stink bug feeding with season long infestation of sub-threshold populations. When stink bug populations were estimated using a sweep net, populations exceeding nine bugs

per 25 sweeps caused reduced seed quality (McPherson et al. 1993). Higher populations have been shown to cause significant seed injury after a short 14-day infestation period on R5-R6 stage soybean (Young et al. 2008). After R7, stink bugs cause less injury to soybean and need to be present at high densities (over 2.3 bugs per 0.3 row m) to cause economic damage, unless soybean quality has been previously decreased due to factors including other insect injury, disease, and drought (Musser et al. 2011).

Acrosternum hilare biology

Acrosternum hilare is an important pest of several crops in Virginia. It constitutes a major component of the native stink bug pest complex that attacks corn, cotton, soybean, fruit trees, tomato, and other vegetables in Virginia (McPherson and McPherson 2000). Little research has been done comparing native species feeding habits, salivary secretions, and resultant injury to soybean. Miner (1966) found *A. hilare* to be more damaging than *E. servus*, yet a different study found *E. servus* to be slightly more damaging than *A. hilare* (McPherson et al. 1979). *Nezara viridula* is the most damaging of the native stink bug species to soybean and most abundant in the southern U.S. (McPherson et al. 1979, McPherson et al. 1993), but it is not found in the mid-Atlantic region (Kamminga 2008).

Acrosternum hilare ranges throughout the United States and southern Canada (McPherson and McPherson 2000). It has one generation in the northern parts of its range, and two generations south of Virginia and Illinois (Kamminga 2008, McPherson and Tecic 1997). It overwinters in the adult stage in debris in the leaf litter on deciduous forest floors (Underhill 1934). Adults will break diapause when temperatures climb above 10° C (Underhill 1934). Winters that vary in temperature, with extremely cold temperatures quickly following warm temperatures above 20°C that enable the bugs to become very active, can kill large numbers of

overwintering adults (Whitmarsh 1917). In May and June, adults fly from their wooded overwintering sites onto woody hosts such as wild cherry, locust, mimosa, and elderberry to feed and reproduce. Females lay clusters of about 30 eggs every eight to 10 days, and will lay up to five clusters (Underhill 1934). The time period between eclosion and the adult stage ranges from 50-75 days (Whitmarsh 1914). It usually migrates to crops either when native hosts decline in quality or towards the end of the season. Soybean is an attractive late season crop that can support development of a second generation (Jones and Sullivan 1982).

Current soybean production practices

The potential to limit stink bug damage through cultural control practices has been examined by several researchers in the Southeast. Some researchers have suggested removing alternate woody hosts near soybean fields (McPherson and McPherson 2000). Small seeded soybean varieties may be more tolerant of stink bug injury than those that produce larger seeds (Wada et al. 2006).

Planting date has also been examined as a possible means of limiting stink bug infestions. In the South, a large percentage of soybean acreage is managed using the early season production system, where soybean are planted earlier (in April) than the conventional full season production system soybean are planted (mid-May). Varieties used by the early season production system also mature earlier than varieties used in the full season production system so that the soybean matures before late summer's typically hot, dry weather sets in (usually late August-mid September). If soybeans mature prior to this period, they often have lower stink bug populations and decreased stink bug damage (Gore et al. 2006). However, the effect of planting date and maturity group on limiting stink bug populations is lost the later soybeans are planted, regardless of maturity group. This may be due to a dependence on photoperiod for stink bug reproductive

development, not just crop phenology. Stink bug populations peak in September to October, near the autumnal equinox, and will be present in any soybean field producing pods at this time, regardless of planting date and maturity group (Herbert and Toews 2011). Stink bugs that infest the earlier soybean reproductive stages cause greater seed quality and yield loss (Daugherty et al. 1964). Conversely, stink bugs infesting soybean in later development stages cause less damage, as there is less time before harvest and later maturing development stages are less susceptible to injury (Daugherty et al. 1964).

Many of the maturity group IV and V varieties that are planted in double crop and early season production systems exhibit indeterminate growth, and there has been a shift towards increased use of indeterminate soybean varieties, not just in the United States (Musser et al. 2011, Soybean and Corn Advisor 2012). Indeterminate varieties may respond better than determinate varieties to adverse conditions during their reproductive stages. Indeterminate soybean varieties continue vegetative growth and continue to set new pods throughout reproductive development, while determinate varieties flower at one time and have very limited growth after the R2 stage (Pyle 1982). Although both types of soybean attempt to compensate for stink bug feeding injury (Russin et al. 1982), indeterminate soybean varieties may be affected differently (Simmons and Yeargan 1990).

The early season production system accounts for more than three quarters of soybean planted in the southeastern U.S. This system has the added benefit of allowing many soybean fields to escape severe stink bug infestations. Stink bug populations typically peak in early September; if early maturing soybean varieties are planted early, they can be harvested before stink bug populations peak (Gore et al. 2006). However, care needs to be taken to prevent loss in later planted soybean or later maturing varieties. Stink bugs migrate into the early planted

soybean earlier and begin to reproduce in them. When the early planted soybean fields senesce, stink bugs migrate to later planted soybean, which are beginning to produce pods. Thus, the early soybean system can concentrate higher stink bug populations in later planted soybean (McPherson and Bondari 1991, McPherson et al. 2001).

It is unclear if the early planted soybean production system would benefit Virginia. It has been noted that late maturing soybean offer stink bugs a vital source of late season nourishment and is present long enough to produce a second generation of stink bugs. Early-season wild hosts are no longer as suitable for feeding or attractive for stink bug reproduction by the time late planted soybean is maturing (Jones and Sullivan 1982). In Virginia, soybean is often double-cropped; that is, they are planted after small grains are harvested, usually not before early June. In 2010, the number of acres planted after small grain harvest was about 40% of Virginia's total soybean acreage (USDA/NASS 2011). This means that potentially 40% of Virginia's soybean crop is planted late and is therefore potentially more susceptible to experiencing stink bug reproduction and damage in September-October. Even if the full season soybean acreage in Virginia were planted early, more than half of the crop could still encounter severe stink bug infestations.

Stink bugs also appear to prefer soybean over cotton, another crop for which they cause economic damage. This preference appears to be strong enough to draw stink bugs away from cotton and into more attractive soybean that is planted nearby (Bundy and McPherson 2000). Thus, it would seem that not only can different soybean varieties be planted to avoid stinkbug injury, but they may serve as an unintentionally planned trap crop for other surrounding crops.

***Halyomorpha halys* biology**

Halyomorpha halys is a recently introduced invasive pest from Asia. It was first found in Allentown, Pennsylvania around 1996, but not correctly identified until 2001 (Hoebeke and Carter 2003). In about 15 generations it has spread from one county in Pennsylvania to several states, and is becoming the dominant stink bug species in the mid-Atlantic region (Nielsen and Hamilton 2009). It is now reported from over 30 states, including Oregon and California (Belisle 2011). It was first discovered in Virginia in 2004, and is considered to be distributed statewide (Day et al. 2011). *Halyomorpha halys* has also spread into Ontario, Canada and Switzerland (Wermelinger et al. 2008).

Halyomorpha halys was first noticed because of its overwintering behavior inside man-made structures, often houses, and was subsequently recognized only as an urban nuisance pest. In the fall, adults fly *en masse* to structures, often aggregating on the outside before moving inside to overwinter. This behavior is similar to the multicolored Asian lady beetle, *Harmonia axyridis* Pallas (Coleoptera: Coccinellidae) (Hoebeke and Carter 2003). *Halyomorpha halys'* potential for being a severe agricultural pest in the United States was not recognized until recently. This could be due in part because of its wide host range, which may have allowed it to disperse throughout the agricultural landscape in low populations that were not recognized as an economic problem, especially in regions where native stink bug species are less important. It was not until large populations of *H. halys* had built up that it was found to be economically damaging on various crops, including orchard fruit (apples, peaches, cherries, etc.) small fruit (grapes and berries), field crops (corn and soybean), vegetables, ornamentals, and many native shrubs and trees (Hamilton 2009).

Halyomorpha halys is a known vector of plant diseases. In its native range, it transmits a mycoplasma disease called *Paulownia* witches' broom, which weakens and shortens the life span

of *Paulownia tomentosa* Thunberg trees, an invasive tree species in the United States (Hoebeke and Carter 2003). Research needs to be conducted to determine if *H. halys* is a competent vector of other phytoplasma diseases that could potentially affect fruit trees (Bernon 2004). *Halyomorpha halys* can also transmit the yeast *Eremothecium coryli* to plant tissue and fruiting tissue of vegetables, soybean, and cotton in similar fashion to the native phytophagous stink bugs (Brust and Rane 2011).

Halyomorpha halys undergoes between four and six generations in the southern parts of its range in China (Hoffman 1931). In the mid-Atlantic U.S., it appears to be mostly univoltine, (Nielsen et al. 2008), but may be bivoltine. While it is still spreading, its final distribution in the U.S. is likely to be very similar to *A. hilare*. It is likely to exhibit similar generational biology to *A. hilare*, which has two generations in Virginia and further south (Nielsen et al. 2008, Holtz and Kamminga 2010).

Native stink bugs exhibit strongly aggregated spatial distributions. Although aggregates may be found near field edges, especially next to woods, these aggregations can encompass large portions of a field, and some aggregations may be found nearer to the field interior (Reay-Jones 2010, Reay-Jones et al. 2010). *Halyomorpha halys* has been observed to aggregate most heavily along field edges (within three meters), with very few being found in the field interiors (Dively and Patton 2011). Soybean near field edges often exhibit delayed maturity, poor yield and poor seed quality due to extremely high stink bug populations. Due to such extreme edge aggregation, the possibility for managing *H. halys* in soybean by spraying only the perimeter of the field seems promising, and research is ongoing (Dively and Patton 2011).

Although the exact aggregation pheromone of *H. halys* has not been identified, it is attracted to methyl 2, 4, 5-decatrienoate, a pheromone for the stink bug *Plautia stali* Scott in East

Asia (Sugie et al. 1996). When tested in baited traps in the United States, this compound was found to cross attract *Thyanta* spp. and *A. hilare*, as well as several Tachinid parasitoids (Aldrich et al. 2007). If *H. halys* produces this compound or a similar compound as an aggregation pheromone, then large perimeter aggregates of *H. halys* could draw in large numbers of native stink bugs. Cross-attracted native stink bugs may aggregate in larger areas throughout the field, not necessarily confined to a narrow perimeter.

Halyomorpha halys also exhibits a very strong startle response. Stink bugs quickly fly away from approaching objects in the field (Hoffman 1931) or enter a state of anabiosis by dropping off of plants in cooler temperatures (<23°C). This behavior can make scouting and insecticide application difficult (Li et al. 2007). *H. halys* also seek out shaded dark areas in the fall when they are seeking overwintering refuge sites. During the growing season, stink bugs are often found under leaves out of direct sunlight (Toyama et al. 2011).

Research justification

Relatively little information is available on the injury comparisons between native species in field crops, let alone introduced species. Differences in injury inflicted on cotton by different stink bug species have been detected, with *E. servus* causing more damage to cotton than *Euschistus quadrator* Rolston (Hopkins et al. 2009). Miner (1966) found that *E. servus* apparently did not injure soybean seeds as severely as *A. hilare* or *N. viridula.* The introduced red banded stink bug, *Piezodorus guildinii* Westwood (Hemiptera: Pentatomidae) is more damaging to soybeans than the native stink bugs and more difficult to control (Baldwin et al. 2010, Allen et al. 2011). Adult *P. guildinii* are more prone to flying and escaping insecticide application (LSU AgCenter 2011), which is a similar behavior to the introduced *H. halys*. The differences between *P. guildinii* and native stink bug species have led to much lower thresholds

for *P. guildinii* (24 bugs in 100 sweeps vs. 36 bugs in 100 sweeps for native stink bug species) (Baldwin et al. 2010). To date, little direct species comparison work has been done between *H. halys* and other native stink bugs. Such research could have implications for economic threshold development.

The current economic threshold for most states in the Southeast is one stink bug per 0.3 row m for soybean in the pod filling development stage (Table 1.1) (Allen et a. 2011, Baldwin et al. 2010, Herbert 2009, Linker et al. 1999, Lorenz et al. 2006, Thompson et al. 2006). These thresholds are similar to those suggested by Miner (1966), and correspond to between 3.6 bugs per 15 sweep sample (Herbert 2009) and 5.4 bugs per 15 sweep sample (Rudd and Jensen 1977). However, many researchers have expressed concern that these thresholds need to be reevaluated. Soybean production practices have changed substantially since these thresholds were developed in the 1960's. For instance, the average yield of soybean in Iowa during the 1960's was about 1,880 kg per hectare, while today it is about 2,690 kg per hectare (Whigham 2002). These production trends are similar for other soybean producing states. In addition, soybean is now commonly grown on narrower row spacings or drilled, many soybean fields are double-cropped after small grain harvest, and the varieties in production today are different from those in production when the thresholds were first developed.

Stink bug scouting

The stink bug density per row unit can be found using a standard 0.91 m beat sheet to sample (Turnipseed 1974, Rudd and Jensen 1977). However, researchers are at odds as to which method more closely estimates the true number of stink bugs; most often, the sweep net is used because it is the easiest and the most economic means of scouting soybean for pest insects (Rudd and Jensen 1977). Attempts to calibrate sweep net catches in wide-row, narrow-row, and drilled

beans have also been done. These attempts usually convert catches to insect density, and this can be compared with traditional thresholds of insects per row unit (Pitre et al. 1987). The sweep net is the preferred choice among scouts when scouting for insect pests, including stink bugs, in reproductive stage soybean.

One drawback to using the sweep net is that it samples the uppermost portion of the soybean canopy, potentially missing insects in the lower canopy. Environmental conditions, such as temperature, wind, and relative humidity can also affect an insect population's distribution in a canopy, and, therefore, sweep net accuracy (DeLong 1932). If insects are deeper in the canopy, they may not be sampled adequately. Few studies have evaluated the climate within a crop canopy and environmental effects on the behavior and distribution of Hemipterans. When comparing time of day to sweep net catches of rice stink bug, *Oebalus pugnax* F., researchers found that counts varied by time of day in more than half of the sampling dates (Espino et al. 2008). However, possible causes of this relationship were not examined. Romney (1945) found that population estimates of the beet leafhopper, *Eutettix tenellus* Baker (Hemiptera: Cicadellidae) done by sweep net varied tremendously with temperature, wind speed, and time of day. Additionally, *Orius insidiosus* Say (Hemiptera: Anthocoridae) population estimates were influenced by time of day, temperature, and cloud cover. Other predatory true bug counts varied with both soil temperature and cloud cover (Dumas et al. 1962; Dumas et al. 1964). Shepard et al. (1974) studied *Geocoris* spp. (Hemiptera: Lygaeidae) vertical distribution in cages with regards to temperature throughout the day. They found that not only do *Geocoris* move from the top of cages to lower positions throughout the day, but that the numbers of *Geocoris* caught in sweep nets differed between times of day, with the most caught during the morning hours. A study done in Australia on *N. viridula* found that basking behavior

was greatest during the morning hours between 7:00 and 9:00 a.m. After that, bugs either moved under leaves or moved lower in the canopy. However, ambient temperature, humidity, and time of day were not highly correlated with stink bug movement (Waite 1980).

In Virginia, stink bugs are scouted in late summer when temperatures can generally reach their highest annual levels. If stink bugs change position throughout the day within the canopy, then this could influence both the accuracy of sampling as well as insecticide treatment efficacy. For example, sampling when temperatures are high and bugs are deep in the canopy could result in a 'false low' scouting report underestimating the population and potentially delaying an insecticide application. If stink bug populations are deeper in the soybean canopy, it is also possible that insecticide applications could be less efficacious.

References Cited

Aldrich, J. R., A. Khrimian, and M. J. Camp. 2007. Methyl 2, 4, 6-decatrienoates attract stink bugs and Tachinid parasitoids. J. Chem. Ecol. 33: 801-815.

Allen, C., G. Andrews, B. Burdine, A. Catchot, D. Cook, D. Dodds, J. Gore, M. Howell, R. Jackson, E. Larson, B. Layron, F. Musser, L. Owen, G. Snodgrass, and S. Winters. 2011. 2011 Insect Control Guide for Agronomic Crops. A. Catchot. Extension Service of Mississippi State University, Publication 2471.

Bacheler, J. S., J. Van Duyn, D. Mott, A. Herbert, E. Blinka, J. Greene, P. Roberts, M. Toews, and R. Smith. 2010. Managing stink bugs in cotton: research in the Southeast Region. Pp. 991-993. *In* Proceedings, 2010 Beltwide Cotton Conferences. National Cotton Council, Memphis, TN.

Baldwin, J., L. Foil, M. Grodner, A. Hammond, G. Henderson, N. Hummel, S. Johnson, R. Leonard, A. Morgan, D. K. Pollet, J. Pyzner, T. E. Reagan, D. Reed, R. N. Story, and M. Stout. 2010. Louisiana Insect Pest Management Guide. LSU AgCenter. http://www.lsuagcenter.com/NR/rdonlyres/B733BE31-7DEA-4264-93A6-AF6FED7023D3/66760/pub1838InsectPestMgmtGuide2010completebook188pgs.pdf. Accessed 6 April 2011.

Belisle, R. F. 2011. Nearly 400 Jam into Auditorium to Learn About Stink Bugs. Herald-Mail. http://www.herald-mail.com/news/tristate/hm-nearly-400-jam-into-auditorium-to-learn-more-about-stink-bugs-20110209,0,5317231,full.story. Accessed 7 March 2011.

Bernon, G. 2004. Biology of *Halyomorpha halys*, the Brown Marmorated Stink Bug (BMSB) Final Report. USDA CPHST. T3P01.

Boethel, D. J., J. S. Russin, A. T. Wier, M. B. Layton, J. S. Mink, and M. L. Boyd. 2000. Delayed maturity associated with southern green stink bug (Heteroptera: Pentatomidae) injury at various soybean phonological stages. J. Econ. Entomol. 93: 707-712.

Brust, G. and K. Rane. 2011. Transmission of the yeast *Eremothecium coryli* to fruits and vegetables by the brown marmorated stink bug. http://www.agnr.umd.edu/Extension/agriculture/mdvegetables/files/yeast%20transmission%20by%20BMSB%20in%20vegetables.pdf.

Bundy, C. S. and R. M. McPherson. 2000. Dynamics and seasonal abundance of stink bugs (Heteroptera: Pentatomidae) in a cotton-soybean ecosystem. J. Econ. Entomol. 93: 697-706.

Corrêa-Ferreira, B. S. and J. de Azevedo. 2002. Soybean seed damage by different species of stink bugs. Agr. Forest Entomol. 4: 145-150.

Daugherty, D. M., M. H. Neustad, C. W. Gehrke, L. E. Cavanah, L. F. Williams, and D. E. Green. 1964. An evaluation of damage to soybeans by brown and green stink bugs. J. Econ. Entomol. 57: 719-722.

Day, E. R., T. McCoy, D. Miller, T. P. Kuhar, and D. G. Pfeiffer. 2011. Brown marmorated stink bug. Virginia Cooperative Extension Publication Number 2902-1100.

DeLong, D. M. 1932. Some problems encountered in the estimation of insect populations by the sweeping method. Ann. Entomol. Soc. Am. 25: 13-17.

Depieri, F. A. and A. R. Panizzi. 2011. Duration of feeding and superficial and in-depth damage to soybean seed by selected species of stink bugs (Heteroptera: Pentatomidae). Neotrop. Entomol. 40: 197-203.

Dively, G. P. and T. Patton. 2011. Summary of 2010 studies of brown marmorated stink bug on soybean in Maryland. http://www.frederick.umd.edu/files/Stinkbug-Dively2010studiesofBMSBonSoybeaninMaryland.pdf. Accessed 5 February 2012.

Dumas, B. A., W. P. Boyer, and W. H. Whitcomb. 1962. Effect of time of day on surveys of predaceous insects in field crops. Fla. Entomol. 45: 121-128.

Dumas, B. A., W. P. Boyer, and W. H. Whitcomb. 1964. Effect of various factors on surveys of predaceous insects in soybeans. J. Kans. Entomol. Soc. 37: 192-201.

Espino, L., M. O. Way, and L. T. Wilson. 2008. Determination of *Oebalus pugnax* (Hemiptera: Pentatomidae) spatial pattern in rice and development of visual sampling methods and population sampling plans. J. Econ. Entomol. 101: 216-225.

Fehr, W. R., C. E. Caviness, D. T. Burmood, and J. S. Pennington. 1971. Stage of development descriptions for soybeans, *Glycine max* (L.) Merrill. Crop Sci. 11: 929-931.

Gore, J., C. A. Abel, J. J. Adamczyk, and G. Snodgrass. 2006. Influence of soybean planting date and maturity group on stink bug (Heteroptera: Pentatomidae) populations. Environ. Entomol. 35: 531-536.

Hamilton, G. C. 2009. Brown marmorated stink bug. Amer. Entomol. 55: 19-20.

Herbert, D. A. Jr. 2009. Insects: Soybeans. Pest Management Guide 2010 Field Crops. D. A. Herbert and S. E. Hagood, Virginia Cooperative Extension Publication Number 456-016-10: 4.61-64.76.

Herbert, J. J., and M. D. Toews. 2011. Seasonal abundance and population structure of brown stink bug (Hemiptera: Pentatomidae) in farmscapes containing corn, cotton, peanut, and soybean. Ann. Entomol. Soc. Am. 104: 909-918.

Hoebeke, E. R. and M. E. Carter. 2003. *Halyomorpha halys* (Stal) (Heteroptera: Pentatomidae): a polyphagous plant pest from Asia newly detected in North America. Proc. Entomol. Soc. Wash. 105: 225-237.

Hoffman, W. E. 1931. A Pentatomid pest of growing beans in South China. Peking Nat. Hist. Bull. 5: 25-26.

Holtz, T. and K. Kammimga. 2010. Qualitative analysis of the pest risk potential of the brown marmorated stink bug (BMSB), *Halyomorpha halys* (Stål), in the United States. USDA APHIS PPQ, pp 33.

Hopkins, W. B., A. E. Knutson, J. S. Bernal, M. F. Treacy, and C. W. Smith. 2009. Species composition, damage potential, and insecticide susceptibility of stink bugs in cotton in the lower Gulf Coast region of Texas. Southwest. Entomol. 35: 19-32.

Jones, W. A. Jr. and M. J. Sullivan. 1982. Role of host plants in population dynamics of stink bug pests of soybean in South Carolina. Environ. Entomol. 11: 867-875.

Kamminga, K. L. 2008. Species survey, monitoring, and management of economically important stink bug species in Eastern Virginia. Ph.D. dissertation, Virginia Tech, 2008.

Li, X., X-y. Yin, Y-h. Liu, J. Fu, and X-f. Liu. 2007. *Halyomorpha halys* behaviors and their application for control. Journal of Northwest A&F University (Nat. Sci. Ed.) 35: 139-145.

Linker, H. M., J. S. Bacheler, H. D. Coble, E. J. Dunphy, S. R. Koenning, and J. W. Van Duyn. 1999. Integrated Pest Management Soybean Scouting Manual. North Carolina Cooperative Extension Service pub. no. AG-385. http://ipm.ncsu.edu/soybeans/Scouting_Soybeans/soyfinal.pdf. Accessed 6 April 2011.

Lorenz, G., D. R. Johnson, G. Studebaker, C. Allen, and S. Young, III. 2006. Insect Pest Management in Soybean. University of Arkansas Division of Agriculture Cooperative Extension Service Publication Number MP197-10M-4-00RV.

LSU AgCenter. 2011. Overview of the red banded stink bug. http://text.lsuagcenter.com/en/crops_livestock/crops/soybeans/red_shouldered_stink_bug/overview/Overview+of+the+Redbanded+Stink+Bug.htm.

Lu, Y., K. Wu, Y. Jiang, B. Xia, P. Li, H. Feng, K. A. G. Wyckhuys, and Y. Guo. 2010. Mirid bug outbreaks in multiple crops correlated with wide-scale adoption of Bt cotton in China. Science 328: 1151-1154

McPherson, J. E. and D. L. Tecic. 1997. Notes on the life histories of *Acrosternum hilare* and *Cosmopepla bimaculata* (Heteroptera: Pentatomidae) in southern Illinois. Gt. Lakes Entomol. 30: 79-84.

McPherson, J. E. and R. M. McPherson. 2000. Stink Bugs of Economic Importance in America North of Mexico. Boca Raton, FL, CRC Press.

McPherson, R. M. and K. Bondari. 1991. Influence of planting date and row width on abundance of velvetbean caterpillars (Lepidoptera: Noctuidae) and southern green stink bugs (Heteroptera: Pentatomidae) in soybean. J. Econ. Entomol. 84: 311-316.

McPherson, R. M., G. K. Douce, and R. D. Hudson. 1993. Annual variation in stink bug (Heteroptera: Pentatomidae) seasonal abundance and species composition in Georgia soybean and its impact on yield and quality. J. Entomol. Sci. 28: 61-72.

McPherson, R. M., L. D. Newsom, and B. F. Farthing. 1979. Evaluation of four stink bug species from three genera affecting soybean yield and quality in Louisiana. J. Econ. Entomol. 72: 188-194.

McPherson, R. M., M. L. Wells, and C. S. Bundy. 2001. Impact of the early soybean production system on arthropod pest populations in Georgia. Environ. Entomol. 30: 76-81.

Miner, F. D. 1961. Stink bug damage to soybeans. Ark. Farm. Res. 10: 12.

Miner, F. D. 1966. Biology and control of stink bugs in soybeans. Ark. Agric. Exp. Stn. Bull. 708: 1-40.

Musser, F. R., G. M. Lorenz, S. D. Stewart, and A. L. Catchot, Jr. 2010. 2010 Soybean insect losses for Mississippi, Tennessee, and Arkansas. Midsouth Entomol. 4: 22-28.

Musser, F. R., A. L. Catchot, B. K. Gibson, and K. S. Knighten. 2011. Economic injury leves for southern green stink bugs (Hemiptera: Pentatomidae) in R7 growth stage soybeans. Crop Prot. 30: 63-69.

Newsom, L. D., M. Kogan, F. D. Miner, R. L. Rabb, S. G. Turnipseed, and W. H. Whitcomb. 1980. General accomplishments toward better pest control in soybean, pp. 51 - 98. *In* C. B. Huffaker (Ed.), New technology of pest control. Wiley-Interscience Publ, John Wiley & Sons, Inc., New York, NY. 500 pp.

Nielsen, A. L. and G. C. Hamilton. 2009. Life History of the Invasive Species Halyomorpha halys (Hemiptera: Pentatomidae) in Northeastern United States. Ann. Entomol. Soc. Am. 102: 608-616.

Nielsen, A. L., G. C. Hamilton, and D. Matadha. 2008. Developmental rate estimation and life table analysis for *Halyomorpha halys* (Hemiptera: Pentatomidae). Environ. Entomol. 37: 348-355.

Pitre, H. N., L. G. Thead, and J. L. Hamer. 1987. Prediction of field populations of soybean insects from sweep-net samples in narrow-row soybean plantings. J. Econ. Entomol. 80: 848-853.

Plotkin, M. D., R. L. Hagan, S. Kallek, and O. L. Wilhite. 1977. 1974 Census of Agriculture: Virginia State and County Data. U.S. Department of Commerce: Bureau of the Census 46.

Pyle, M. E. 1982. A comparison of determinate and indeterminate soybean lines for double cropping in Virginia. Ph.D. dissertation, Virginia Tech, 1982.

Reay-Jones, F. P. F. 2010. Spatial and temporal patterns of stink bugs (Hemiptera: Pentatomidae) in wheat. Environ. Entomol. 39: 944-955.

Reay-Jones, F. P. F., M. D. Toews, J. K. Greene, and R. B. Reeves. 2010. Spatial dynamics of stink bugs (Hemiptera: Pentatomidae) and associated boll injury in Southeastern cotton fields. Environ. Entomol. 39: 956-969.

Romney, V. E. 1945. The effect of physical factors upon catch of the beet leafhopper (*Eutettix tenellus* (Bak.)) by a cylinder and two sweep-net methods. Ecol. 26: 135-147.

Rudd, W.G. and R. L. Jensen. 1977. Sweep net and ground cloth sampling for insects in soybeans. J. Econ. Entomol. 70: 301-304.

Shepard, M., V. H. Waddill, and S. G. Turnipseed. 1974. Dispersal of *Geocoris* spp. in soybeans. J. Ga. Entomol. Soc. 9: 120-126.

Simmons, A. M. and K. V. Yeargan. 1990. Effect of combined injuries from defoliation and green stink bug (Hemiptera: Pentatomidae) and influence of field cages on soybean yield and seed quality. J. Econ. Entomol. 83: 599-609.

Sugie, H., M. Yoshida, K. Kawasaki, H. Noguchi, S. Moriya, K. Takagi, H. Fukuda, A. Fujiie, M. Yamanaka, Y. Ohira, T. Tsutsumi, K. Tsuda, K. Fukumoto, M. Yamashita, and H. Suzuki. 1996. Identification of the aggregation pheromone of the brown-winged green bug, *Plautia stali* Scott (Heteroptera: Pentatomidae). Appl. Entomol. Zool. 31: 427-431.

Soybean & Corn Advisor. 2012. Indeterminate soybeans replacing determinate varieties in Brazil. Soybean & Corn Advisor, Inc. http://cornandsoybean.com/news/Feb24_12-Inderterminate-Soybeans-Replacing-Determinate-Varieties-in-Brazil. Accessed 19 April 2012.

Thomas, G. D., C. M. Ignoffo, C. E. Morgan, and W. A. Dickerson. 1974. Southern green stink bug: influence on yield and quality of soybeans. J. Econ. Entomol. 67: 501-503.

Thompson, A., D. Hensley, M. Newman, S. Stewart, and R. Patrick. 2006. Crop Profile for Soybeans in Tennessee. NSF Center for IPM, USDA: NIFA. http://www.ipmcenters.org/cropprofiles/docs/tnsoybeans.pdf. Accessed 6 April 2011.

Toyama, M., F. Ihara, and K. Yaginuma. 2011. Photo-response of the brown marmorated stink bug, *Halyomorpha halys* (Stål) (Heteroptera: Pentatomidae), and its role in the hiding behavior. Appl. Entomol. Zool. 46: 37-40.

Turnipseed, S. G. 1974. Sampling soybean insects by various D-Vac, sweep and ground cloth methods. Fla. Entomol. 57: 217-223.

Underhill, G. W. 1934. The Green Stinkbug. Va. Agric. Exp. Stn. Bull. 294.

United States Department of Agriculture [USDA]: National Agricultural Statistics Service (NASS). 2009.

http://www.nass.usda.gov/Statistics_by_State/Virginia/Publications/Annual_Statistical_Bulletin/pg20-2109.pdf. Accessed 3 January 2011.

United States Department of Agriculture [USDA]: National Agricultural Statistics Service (NASS). 2011. http://www.nass.usda.gov/Statistics_by_State/Virginia/Publications/5YRCROP10122005.pdf. Accessed 18 April 2011.

Wada, T., N. Endo, and M. Takahashi. 2006. Reducing seed damage by soybean bugs by growing small-seeded soybeans and delaying sowing time. Crop Prot. 25: 726-731.

Waite, G. K. 1980. The basking behaviour of *Nezara viridula* (L.) (Pentatomidae: Hemiptera) on soybeans and its implication in control. J. Aust. Entomol. Soc. 19: 157-159.

Wermelinger, B., D. Wyniger, and B. Forster. 2008. First records of an invasive bug in Europe: *Halyomorpha halys* Stål (Heteroptera: Pentatomidae), a new pest on woody ornamentals and fruit trees? Mitt. Schweiz. Entomol. Ges. 81: 1-8.

Whigham, K. 2002. Iowa soybean production history. Iowa State University University Extnesion pub. no. IC-488. http://www.ipm.iastate.edu/ipm/icm/2002/1-21-2002/prodhistory.html. Accessed 13 April 2011.

Whitmarsh, R. D. 1914. The green soldier bug (*Nezara hilaris*). J. Econ. Entomol. 7: 336-339.

Whitmarsh, R. D. 1917. The green soldier bug. Ohio Agric. Exp. Stn. Bull. 310: 519-551.

Yeargan, K. V. 1977. Effects of green stink bug damage on yield and quality of soybeans. J. Econ. Entomol. 70: 619-622.

Young, S. Y., J. K. Greene, and G. M. Lorenz. 2008. Damage to soybean by *Acrosternum hilare* (Say) (Heteroptera: Pentatomidae). J. Entomol. Sci. 43: 257-267.

Table 1.1. Stink bug thresholds published in extension recommendations in mid-South, Eastern, and mid-Atlantic states for soybean

Beat sheet (number per row meter)	Sweep net (per 25 sweeps)	Soybean type or stage	State*
0.5	1.5-2.8	Seed	Louisiana, Georgia, North Carolina
1	3	Grain, bloom to mid-pod fill	Georgia, Mississippi, Tennessee
3	6-9	Grain, mid-pod fill to harvest	Georgia, Mississippi, Louisiana, Tennessee, Arkansas, North Carolina, Virginia

*data taken from each state's pest management guide.

Chapter 2

Effects of *Acrosternum hilare* and *Euschistus servus* feeding on soybean seed and pod injury in the mid-Atlantic U. S.

Abstract

Relatively few studies have assessed the effects of stink bug feeding injury to soybean in the mid-Atlantic states. Furthermore, soybean cropping systems have changed dramatically since thresholds were first developed in the southeastern U.S. Finally, several stink bug species comprise the stink bug complex that feeds on soybean, but little research has been done comparing injury to soybean caused by individual species. To address these research needs, yearly field cage studies were initiated at one site each in Maryland and Virginia, and two in Delaware from 2007-2009 using mesh cages enclosing 0.3 row m. Single *Acrosternum hilare* adults (or nymphs) and *Euschistus servus* adults were introduced into cages for three weeks at various soybean developmental stages in 2007 and 2008. In 2009, densities of zero, one, two, and four stink bugs per 0.3 row m were introduced into the cages for three weeks. After the three week infestation period, stink bugs were removed, and cages were resealed. At harvest, plants were hand shelled and assessed for seed quality, destroyed pods, yield, and cage seed number. Results showed no significant difference of injury to soybeans caused by either stink bug species or life stages. The full pod (R4) and beginning seed (R5) developmental stages were the most sensitive to stink bugs and to cage effects. Stink bug effects on soybean seed quality and yield were inconsistent over the course of the experiments.

Keywords: stink bugs, soybean, thresholds, cage studies

Stink bugs (Hemiptera: Pentatomidae) have received increasing attention as major pests in field crop ecosystems in the United States. Due to the success of the boll weevil eradication program and the widespread adoption genetically modified Bt crops, the number of insecticide applications has decreased significantly (Lu et al. 2010). Furthermore, the adoption of more Lepidoptera-specific insecticides that do not control stink bugs has reduced the number of broad-spectrum applications that control stink bugs. In this reduced broad-spectrum insecticide spray environment, average Hemipteran population densities, including stink bugs, have been increasing in many field crops and causing significant economic damage (Lu et al. 2010, McPherson and McPherson 2000). Landscape dynamics have also been changing with the expansion of soybean acreage in the southern U.S. in the last 40 years, a crop that is attractive in the late season to stink bugs for food and reproduction. This has allowed for additional stink bug generations and decreased overwintering mortality (Jones and Sullivan 1982). Interest in the economic aspects of soybean seed and plant injury resulting from stink bug feeding began in the early 1960's (Miner 1961). Feeding injury has been studied in terms of effects on chemical content, seed damage, germination, yield loss, and delayed soybean maturity resulting in harvest difficulty. Daugherty et al. (1964) found that season long stink bug densities of about four per 0.3 row meters were associated with an increase in undeveloped beans and crop maturity delays. If stink bugs infest soybean fields during the early reproductive stages, soybean plants will attempt to compensate for injury by increasing growth to set new pods and increasing the size of unaffected seeds (Daugherty et al. 1964, Boethel et al. 2000). Farmers will often delay their harvest until the entire field has dried to between 13-16% moisture (Daugherty et al. 1964), or they will not harvest affected portions of the field.

Little information is available on the injury differences among native stink bug species in field crops. Injury differences have been detected in cotton, with the brown stink bug, *Euschistus servus* Say, causing more damage than *Euschistus quadrator* Rolston (Hopkins et al. 2009). Miner (1966) found that *E. servus* did not injure soybean seeds as severely as the green stink bug, *Acrosternum hilare* Say (also *Chinavia hilaris*), or the Southern green stink bug, *Nezara viridula* L. if caged on a single raceme of a determinate variety for several days, but in other studies using field cages, *N. viridula* was found to be the most damaging species, *E. servus* was the second most, and *A. hilare* less damaging, depending on soybean variety (McPherson et al. 1979). Despite the differences observed among injury potential of the different species, current economic thresholds combine all three species into a stink bug complex.

From this early work, an action threshold of one stink bug per 0.3 row m was suggested (Miner 1966, Todd and Turnipseed 1974). This threshold corresponds to about nine bugs per 25 sweep sample using a 38 cm diameter sweep net (Rudd and Jensen 1977). Much of this early work also focused on *N. viridula* in more southern areas. In the mid-Atlantic region, *A. hilare* replaces *N. viridula* as the primary species found in soybean fields (Kamminga 2008), except in areas where the recently-introduced brown marmorated stink bug, *Halyomorpha halys* Stål has become well established (Nielsen et al. 2011). Other important members of this phytophagous stink bug complex include *E. servus* and *Thyanta* spp. Stink bugs are attracted to soybean fields just after flowering and seed production begins (Thomas et al. 1974) which usually occurs after the stink bugs' native hosts decline in quality. Soybean is an attractive late season crop that can support development of a second generation of *A. hilare* (Jones and Sullivan 1982).

Recently, researchers have expressed interest in re-examining the thresholds developed in the 1960's and 1970's to determine if they should be adjusted for current soybean production

practices in the mid-Atlantic U.S. Soybean production in the United States has increased dramatically over the last several decades, both in terms of yield per hectare and planted area. For instance, the average yield of soybean in Iowa during the 1960's was about 1,880 kg per ha while today it is about 2,690 kg per ha (Whigham 2002). These production trends are similar for other soybean producing states. In contrast to the 1960's, soybean fields are now commonly planted using narrower row spacings or are drilled, much of the acreage is double-cropped after small grain harvest, many soybean fields are planted using no-till cultivation practices, and the varieties in production today are different from those in production when the stink bug thresholds were first developed. Finally, the thresholds are static for all stink bug life stages, assuming that nymphs and adults feed equally. Field cage studies with the aim of refining current thresholds were conducted in Virginia, Maryland, and Delaware in 2007-2009 to examine the effects of a three week infestation of a current threshold density of stink bugs on soybean seed quality and yield.

Materials and Methods

2007. Small nylon hexagonal-mesh sleeve cages 69 × 105 cm enclosing 0.3 row m of soybean (between three and seven plants) were infested with a single *E. servus* adult, a single *A. hilare* adult or a single *A. hilare* nymph at the Virginia Tech Tidewater Agricultural Research and Extension Center at Suffolk, VA, the University of Maryland Lower Shore Research and Education Center at Salisbury, MD, and at two sites at the University of Delaware's research farm at Newark, DE, the Allen site and the Apiary site. Cages were supported by tying them to a fiberglass pole. Treatments were assigned according to Table 2.1. The indeterminate Southern States 'RT4451N' (maturity group (MG) 4.4) soybean variety was planted on 3 July at Suffolk, late June at Salisbury, and 22 June at both the Allen and Apiary sites in Delaware. Row spacing

was 0.38 m. Cages were arranged in a split plot randomized complete block design replicated three times with soybean development stage as the main plot factor and stink bug presence or absence as the sub plot factor. Cages were placed when soybean in the field had reached development stages R4 (full pod, 1.9 cm pod in top four nodes), R5 (beginning seed, 0.3 cm seed in top four nodes), R6 (full seed in the top four nodes), and R7 (beginning maturity) (only in Suffolk) (Fehr et al. 1971). Adult *A. hilare* were collected from local commercial soybean fields using sweep net, transported to the study sites, and released into cages within 24 hours of collection. Cages were periodically inspected, and missing or dead stink bugs were replaced for the first five to seven days after initial introduction. Stink bugs were removed from cages after three weeks and cages were resealed until harvest to prevent reinfestation by resident stink bugs from the field. Lepidopteran pests were controlled in Suffolk with two sprays of *Bacillus thuringiensis* (Thuricide®, Bonide™, Oriskany, New York) at a rate of 1.2 L product/ ha. At harvest, soybean plants were removed from the cages and taken into the laboratory to determine seed quality and yield data. Aborted pods were estimated by collecting pods from the ground underneath the individual cages, flat pods were categorized as such by counting pods that did not contain seed, and normal pods that had seed were tallied. Normal pods were hand shelled and seeds were visually examined for normal seeds, seeds with evidence of stink bug punctures (discolored spots with damage), diseases (molds and purple stain), and seed coat deformations (shriveled, crinkled, and immature seeds). Yields were determined from the adjusted dry seed weight and cage seed number. At the Salisbury site, only flat and normal pod data, diseased seed data, and yield data were taken, while at the Allen and Apiary sites, only flat and aborted pod data (numeric destroyed pods), diseased seed data, and yield data were taken.

2008. Cages from 2007 were used in 2008 to enclose 0.3 row m of soybean and were infested with a single *A. hilare* adult per cage at Suffolk, VA, University of Maryland's Central Maryland Research and Education Center at Beltsville, MD, and at University of Delaware's research farm at Newark, DE (Newark) and University of Delaware's Carvel Research and Education Center, Georgetown, DE (Georgetown). Southern States 'RT4440N' (indeterminate MG 4.4) was planted at the Suffolk site on 19 June with 0.91 m row spacing. Southern States 'RT 4451N' was planted in late June at the Beltsville, on 19 June at the Newark site, and on 20 June at the Georgetown site. Row spacing at both Delaware sites was 0.76 m. Cages were arranged in the field at the same soybean developmental stages and using the same experimental design as in 2007. Adult *A. hilare* were only introduced into the R5 and R6-initiated cages at the Beltsville site. Plant stands were thinned to three plants per cage at all sites. Stink bugs were removed from the cages after three weeks, and cages were resealed until harvest. Seed quality and yield data were collected as previously described. At the Beltsville site, only yield data were taken, while at both the Newark and Georgetown sites, only destroyed pod and flat pod data were taken and combined as numeric destroyed pods.

2009. In 2009, the cages enclosing 0.3 row m and soybean stands were thinned to three plants per cage were infested with field collected *A. hilare* adults in a similar manner as described for the 2008 cage study. Southern States 'RT4440N' was planted on 11 June at the Suffolk site and on 25 June at the Beltsville site, while Southern States 'RT4451N' soybean were planted on 25 June at both the Newark and Georgetown sites. Cages were infested with densities of zero, one, two, and four stink bugs per 0.3 row m at the soybean development stage R4 at the Suffolk and Beltsville sites, and at R6 at all the sites. Stink bugs were removed from cages after three weeks and cages were resealed until harvest. At harvest maturity, plants were removed

from the cages, taken into the laboratory and hand shelled to determine seed quality and yield data as previously described.

Data analysis of *A. hilare* adult infested cages. The percentages of diseased seed, punctured seed, shriveled seed, and wrinkled seed were combined into one category: damaged seed, with the exception of seed quality data from the 2007 Salisbury, Allen, and Apiary sites, where only diseased seed data were taken and analyzed. Similarly, flat pods and aborted pods, when measured, were combined into a destroyed pod category and calculated as a percentage of the total number of pods; an exception was at both Delaware sites in 2007 and 2008, where only numeric destroyed pod data were recorded and analyzed. Data from all *A. hilare* cage studies were analyzed using split plot analysis with PROC MIXED (Littell et al. 2006, SAS Institute Inc. 2008) in SAS, except for the data from the 2009 Newark and Georgetown sites, which were analyzed using a fixed model ANOVA in SAS JMP v 9.0, as only one soybean development stage was caged at these sites. In PROC MIXED, degrees of freedom were determined using the Kenwood Rogers method (Schaalje et al. 2001). Residuals were tested for heteroscedascity, and heterogeneous variables were repeated in the model with the "Group =" option. Each replicate and replicate × soybean development stage interaction was included as a random effect. Means comparisons were performed using Tukey's HSD (SAS Institute Inc. 2008).

Destroyed pod percentage data from the 2009 Newark and Georgetown sites were $\log_{10}$ transformed. Destroyed pod data from the 2007 Salisbury and the 2007 and 2009 Suffolk sites were square root transformed. Variables that did not exhibit a normal distribution after transformation were analyzed using PROC GLIMMIX (SAS Institute INC. 2008). Damaged seed percentage data from the 2008 Newark site were square root transformed and data from the 2009 Georgetown site were log transformed. Yield and cage seed number data from the 2007

Apiary site and the 2008 Suffolk site were $\log_{10}$ transformed. At the 2007 Allen site, only cage seed number data were log transformed. The numeric destroyed pod data from the 2007 Allen and Apiary sites were analyzed with PROC GLIMMIX using a Poisson distribution and an over dispersion parameter if needed as indicated by the conditional Pearson residuals (Littell et al. 2006). Diseased seed percentage data from the 2007 Allen and Apiary sites, and the damaged seed percentage data from the 2008 Suffolk site were analyzed using PROC GLIMMIX using a pseudo binomial distribution, because percentage data response variables were treated as if they had similar properties to a binomial variable (SAS Institute Inc. 2008). Yield and cage seed number data from the 2007 Suffolk site were analyzed using PROC GLIMMIX, rep × development stage interaction effect was included as a random variable.

Species comparison data analysis. Additional cages infested with *E. servus* and *A. hilare* nymphs were set up at several sites (Table 2.1). Data from the 2007 Allen and Apiary sites were analyzed using a fixed model ANOVA in SAS JMP v 9, as *E. servus* was only introduced onto caged soybean plants at the R4 development stage. The cage seed number data from the cages were $\log_{10}$ transformed from both sites. Yield and destroyed pod data at the 2007 Allen site were also $\log_{10}$ transformed. Diseased seed percentage data from both the 2007 Allen and Apiary sites, and destroyed pod data from the Allen site, were analyzed using the non-parametric Kruskal-Wallis test because data were not normally distributed, even after transformation (SAS Institute Inc. 2007).

At the 2007 Salisbury site, data from the *A. hilare* adult-infested cages and *A. hilare* nymph-infested cages were analyzed using ANOVA, as nymphs were only introduced onto caged soybean plants at the R4 development stage. Destroyed pod percentage and percentage of diseased seed data were square root transformed. At the 2007 Salisbury site, cages containing *E.*

servus and *A. hilare* were arranged in a split plot experimental design, and data were analyzed using PROC MIXED with soybean development stage as the whole plot and species as the subplot factors. Destroyed pod percentage data were square root transformed, and both yield and cage seed number data were log transformed.

Cages from the 2008 Beltsville site enclosed *A. hilare* nymphs, *A. hilare* adults, and *E. servus* adults at the R4 and R6 soybean development stages. Data were analyzed using PROC MIXED in SAS with soybean development stage as the whole plot and species as the subplot factor. Replicate, soybean development stage, and the replicate × soybean development stage interaction were included as random effects. Degrees of freedom were calculated using the Kenwood Rogers method (SAS Institute Inc. 2008). Destroyed pod percentage data from the 2008 Suffolk site were square root transformed. Damaged seed percentage data from the 2008 Suffolk site were analyzed using PROC GLIMMIX fitted with a pseudo binomial distribution (SAS Institute Inc. 2008). All means for the analyses were compared using Tukey's HSD (SAS Institute Inc. 2008). Data from cages enclosing *A. hilare* nymphs at the 2009 Suffolk site were analyzed using ANOVA since nymphs were only introduced on caged plants at the R5 development stage.

Results

Species comparison cages. *Euschistus servus* did not cause significantly differing levels of injury to soybean on any of the injury parameters analyzed in 2007 or 2008 compared with *A. hilare* (Tables 2.2, 2.3, 2.4). The 2007 Allen and Apiary sites had slightly increased soybean yield, and significantly increased cage seed number in cages containing stink bugs, irrespective of species (Tables 2.3, 2.5). The cage seed number from the R6-initiated cages at the 2007 Salisbury site was significantly greater than from the R4-initiated control cages (Tables 2.2, 2.6).

The damaged seed percentage in the cages was not significantly affected by either stink bug species or by soybean development stage for any site (Tables 2.2, 2.3, 2.4, 2.5). At the 2008 Suffolk site, *A. hilare* nymphs tended to increase pod destruction at the R4 soybean development stage, while *A. hilare* adults significantly increased pod destruction at the soybean development stage R5 (Table 2.6). Yield and cage seed number from cages at the 2008 Suffolk site were significantly lower in the R4 and R5-initiated cages, regardless of stink bug species (Tables 2.2, 2.4). Yield was significantly lower in the R4-initiated cages compared with the R6-initiated cages at the 2008 Beltsville site regardless of stink bug species (Tables 2.2, 2.4, 2.7).

At the 2009 Suffolk site, increasing density of *A. hilare* nymphs did not significantly affect soybean seed quality or yield (Table 2.5). Yield from the R5-initiated cages was higher than in either the R4 or R6-initiated cages containing *A. hilare* adults, regardless of stink bug density (Tables 2.8, 2.9).

2007-2008 *A. hilare* cages. Significant stink bug effects on soybean seed quality, pods, and yield were inconsistent between sites. *Acrosternum hilare* adults increased the percentage of damaged seed at the 2007 Suffolk site (Tables 2.10, 2.11), and the damaged seed percentages were greatest in R4 and R5-initiated cages (Tables 2.10, 2.11). However, at the 2008 Georgetown site, the damaged seed percentage was highest in the R6-initiated cages (Tables 2.10, 2.11). In 2008, stink bugs increased the damaged seed percentage at both the Newark and Georgetown sites, with increases of three and nine percent, respectively (Tables 2.10, 2.12). Diseased seed percentage data at the 2007 Allen site was greatest in R4 and R5-initiated cages, regardless of stink bug infestation (Tables 2.10, 2.11). None of the other sites in 2007 had significantly elevated levels of damaged seed (Table 2.10).

Stink bugs only caused an increase in the destroyed pod percentage at the 2007 Suffolk site (Tables 2.10, 2.11), and in 2008 in the R5-initiated cages (Tables 2.10, 2.13). Both the R4 and R5-initiated cages had a significantly greater destroyed pod percentage than those initiated at R7 (Tables 2.10, 2.11, 2.13). R4 and R5-initiated cages at the 2007 Suffolk site tended towards reduced yield (Table 2.11). At the 2007 Allen site, soybean plants from stink bug-infested cages actually had a slightly higher yield and greater cage seed number (Tables 2.10, 2.11). At the 2007 Apiary site, yield in the stink bug infested R5-initiated cages and the R5-initiated control cages were significantly greater than R4-initiated control cages, while all others were intermediate (Tables 2.10, 2.14). Plants from infested R6-initiated cages and R5-initiated control cages had significantly higher cage seed number than the R4-initiated control cages, with all others being intermediate (Tables 2.10, 2.14). The caged soybean yields from the 2008 Beltsville site and the caged soybean yield and cage seed number from the 2008 Suffolk site were significantly lower in the R4-initiated cages (Tables 2.10, 2.12). Stink bugs reduced yield only at the 2008 Newark site regardless of soybean development stage (Tables 2.10, 2.12). At the 2008 Georgetown site, cage seed number was significantly lower in both the stink bug-infested R4 and R6-initiated cages compared with the stink bug infested R5-initiated cages; control cages all had intermediate cage seed number (Tables 2.10, 2.13).

2009 *A. hilare* density cages. The high 4 stink bugs per 0.3 row m density resulted in significant seed damage at the 2009 Beltsville site, and a trend towards increased seed damage at the 2009 Suffolk site (Tables 2.9, 2.10). At the Beltsville site, R4-initiated cages exhibited higher damaged seed percentages than the R6-initiated cages (Tables 2.9, 2.10). The highest density stink bug infestation in the R6-initiated cages at the 2009 Georgetown site resulted in a significant 14% increase in damaged seed (Tables 2.5, 2.15). Stink bug infestation resulted in a

significantly higher percentage of destroyed pods at the Suffolk site (Tables 2.9, 2.10), but the only yield loss caused by stink bugs in 2009 was with the highest density at the Georgetown site's R6-initiated cages (Tables 2.5, 2.15). Stink bugs did not significantly affect the cage seed number in 2009 (Tables 2.5, 2.9, 2.10, 2.15).

Discussion

Stink bug injury to soybean seed has long been one factor leading to discounts from buyers (Miner 1961), and much research has been done examining what infestation levels are required to injure a significant portion of seed, and what soybean development stages are most susceptible. Seed injury is least tolerable for soybean grown for seed and for export consumption (Miner 1961). Even when grown for grain, stink bug injury can affect prices due to their effect on decreasing the oil content of seed (Daugherty et al. 1964, Miner 1961). Stink bug populations that exceed sweep net thresholds and sometimes populations that remain below thresholds per row m throughout the duration of the season result in significant seed quality reductions (Russin et al. 1987, McPherson et al. 1993). Threshold-level populations also corresponded to elevated seed damage in several of the locations in 2007 and 2008, but not in 2009. In 2009, infestation levels of 4 bugs per 0.3 row m caused significant seed injury, and stink bugs had a significant effect on seed quality when infesting R4 and R6 caged plants. Populations a little lower than this have been shown to cause significant seed injury after an infestation period of only 14 days on R5-R6 stage soybean (Young et al. 2008). After R7, stink bugs cause less injury to soybean and need to be at very high densities (over 2.3 bugs per 0.3 row m) to cause economic damage, unless soybean quality is already compromised by some other factors (Musser et al. 2011). This was especially evident at the 2009 Georgetown site, where stink bugs caused significant injury at R6, but yields were one third of those from the other sites

in 2009. Due to the significant increase in percentage of damaged seed at the threshold density in 2007 and 2008, the current thresholds should continue to be used for both R4 and R6 stage double-crop soybean grown for seed.

Different stink bug species and life stages were not associated with differences in seed number and did not cause more injury to soybean. These data support the all-encompassing threshold promoted for stink bugs in soybean. Miner (1966) suggested that *E. servus* might be less damaging than *A. hilare*. Conversely, McPherson et al. (1979) found that *A. hilare* was less damaging than *E. servus*, and both species were significantly less damaging than *N. viridula*. Both of these studies used higher infestation densities and longer infestation periods than were used in this study. It could be that density-dependent effects could produce differences among species injury by challenging the soybean plant under prolonged conditions with higher stink bug densities (Miner 1966) or for shorter time periods under extremely high densities (McPherson et al. 1979). There were no significant differences between nymph and adult-caused damage. This supports the conclusions of Todd and Turnipseed (1974). In contrast, McPherson et al. (1979) found that fifth instar stink bugs were more damaging than adults, regardless of species. Yeargan (1977) also found *A. hilare* nymphs to be more damaging, but he noted that adults are present in the field about five times longer and cause greater cumulative damage. Thresholds do not separate late instars from adults. Nymphs from the R5-initiated cages at the 2009 Suffolk site caused similar damage to adult-infested cages from the other two development stages, but yield was higher in the R5-initiated cages. Nymphs may have also molted into adults during the study which may have further masked any life stage difference in feeding injury. Based on these data stink bug thresholds in the mid-Atlantic do not need to be separated based on species or life stages for native stink bugs.

Maturity delays were observed in all cages, including control cages. This was likely due to a cage effect and compensation due to a long period of reduced photosynthesis resulting from shading in the cages (Yeargan 1977, Buntin 2001). Such compensation has been suspected in mitigating the effects of insect injury on plants that would have otherwise occurred in the field (Simmons and Yeargan 1990). Other researchers using caged populations of stink bugs did not see delayed maturity in non-infested control cages that were left in place until harvest, and attributed maturity delay to stink bugs destroying pods and developing seed (Daugherty et al. 1964). Boethel et al. (2000) did find that stink bug levels of one bug per 0.3 row m were capable of causing slight maturity delays when cages were infested at R4 for one week, and slight delays when infestations lasting two weeks were initiated at R5. Maturity delays from these studies were inconsistent, and were not much different from delays associated with even higher densities. Interpretation of cage studies on soybean injury needs to be conservative, as cages can have significant effects on soybean physiology (Simmons and Yeargan 1990). Cages may have also influenced the percentages of damaged and diseased seed by altering the microclimate to be more favorable to disease organisms, especially at the 2007 Allen site in Newark, DE. Furthermore, cages had a significant influence on flat and aborted pods when placed over plants at the R4 and R5 development stage in three of the four sites in 2007.

In five of the 10 sites where cages were initiated at multiple soybean development stages, soybean yield was lower in the R4-initiated control and infested cages. Yield was higher in the R5-initiated *A. hilare* nymph density cages from the 2009 Suffolk site. This probably is not due to a life stage difference so much as it further highlights the variable yield response from stink bug infestation. Cages stressed the soybean plants the most at the R4-R5 stage, which is also the period when they are more susceptible to stink bug injury. This is consistent with the findings of

other researchers demonstrating increased susceptibility to heavy damage in the earlier development stages (Todd and Turnipseed 1974, Boethel et al. 2000, Yeargan 1977).

Infested R5-intiated cages had a very high percentage of destroyed pods at the 2008 Suffolk site. However, in the 2007 cages, stink bugs did not have a significant effect on pod numbers. In the 2009 stink bug cage studies, densities of two bugs per 0.3 row m or greater significantly increased the percentage of destroyed pods when caged on plants at the R4 stage. Since this density is twice that of the earlier studies, it seems that populations need to be higher than threshold to cause significant pod destruction. Stink bug infestation in the 2009 cages did not result in significant pod destruction when caged on plants at the R6 stage, further highlighting the R4-R5 developmental stage sensitivity to stink bug feeding and to stress. Pod destruction was often correlated with reduced yield. Thomas et al. (1974) found similar results with reproducing populations of *N. viridula*. He attributed the lack of damage at the R6 stage to there not being enough time in the growing season before frost for nymphs to molt into the more damaging late instars and that pods were not as susceptible to injury. Russin et al. (1987) found increased numbers of pods that were missing seeds when exposed to stink bug densities ranging from 0.5 to 1.2 bugs per 0.3 row m from R4 to harvest maturity, but did not see any stink bug effect on soybean yield. This study used late planted MG V soybean, which have a longer period of development from the R4 to the R6 development stages.

There was a significant yield increase in the stink bug-infested cages at the 2007Allen site, and there was a similar trend at the 2007 Apiary site. It is possible that at low levels of feeding, soybean plants may be able to compensate for such injury, resulting in a slight increase in seed size, especially in small seeded varieties (Wada et al. 2006). Conversely, at the 2008 Newark site, yield was significantly reduced by the presence of stink bugs, and at the 2009

Georgetown site, high stink bug densities resulted in significantly lower yield in the R6-initiated cages. As noted previously, these soybean plants may have already been stressed, which indicates that stressed soybean plants that are more mature may still be susceptible to yield loss caused by stink bugs. The inconsistent yield response among the test sites may have also been influenced by the fact that plots were planted with an indeterminate double crop MG IV soybean. Daugherty et al. (1964) concluded that varieties and maturity groups that take longer to mature are more susceptible to stink bug injury than those from earlier maturity groups; however Young et al. (2008) did not see a significant stink bug injury difference between late planted MG IV and MG V soybean varieties. MG IV double crop soybean's R6-R8 (harvest maturity) period usually lasts about three weeks (Holshouser 2010). Once soybean plants reach the R7 stage, thresholds for yield production and additional quality reduction (assuming quality was not reduced earlier) have been suggested to be increased by at least double (Musser et al. 2011). It may be possible for early maturing varieties that require short time intervals between R6 and harvest maturity when planted late to escape significant stink bug injury to both seed quality and yield, but especially yield, if threshold level populations develop during this time frame. If threshold levels develop before R6, control is still warranted, especially since previous studies have shown that populations may rapidly increase in the early reproductive stages (Thomas et al. 1974, Newsom et al. 1980).

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References Cited

Boethel, D. J., J. S. Russin, A. T. Wier, M. B. Layton, J. S. Mink, and M. L. Boyd. 2000. Delayed maturity associated with southern green stink bug (Heteroptera: Pentatomidae) injury at various soybean phenological stages. J. Econ. Entomol. 93: 707-712.

Buntin, G. D. 2001. Techniques for evaluating yield loss from insects, pp. 23-49. *In* R. K. D. Peterson and L. G. Higley (Eds.). Biotic Stress and Yield Loss. CRC Press, Boca Raton, FL. 261 pp.

Daugherty, D. M., M. H. Neustad, C. W. Gehrke, L. E. Cavanah, L. F. Williams, and D. E. Green. 1964. An evaluation of damage to soybeans by brown and green stink bugs. J. Econ. Entomol. 57: 719-722.

Fehr, W. R., C. E. Caviness, D. T. Burmood, and J. S. Pennington. 1971. Stage of development descriptions for soybeans, *Glycine max* (L.) Merrill. Crop Sci. 11: 929-931.

Holshouser, D. L. 2010. Days to soybean physiological maturity. Virginia Cooperative Extension Pub. No. 3009-1459.

Hopkins, W. B., A. E. Knutson, J. S. Bernal, M. F. Treacy, and C. W. Smith. 2009. Species composition, damage potential, and insecticide susceptibility of stink bugs in cotton in the lower Gulf Coast region of Texas. Southwest. Entomol. 35: 19-32.

Jones, W. A., Jr. and M. J. Sullivan. 1982. Role of host plants in population dynamics of stink bug pests of soybean in South Carolina. Environ. Entomol. 11: 867-875.

Kamminga, K. L. 2008. Species survey, monitoring, and management of economically important stink bug species in Eastern Virginia. Ph.D. dissertation, Virginia Tech, 2008.

Littell, R. C., G. A. Milliken, W. W. Stroup, R. D. Wolfinger, and O. Schabenberger. 2006. SAS for mixed models, second edition. SAS Institute Inc., Cary, NC.

Lu, Y., K. Wu, Y. Jiang, B. Xia, P. Li, H. Feng, K. A. G. Wyckhuys, and Y. Guo. 2010. Mirid bug outbreaks in multiple crops correlated with wide-scale adoption of Bt cotton in China. Science 328: 1151-1154

McPherson, R. M., G. K. Douce, and R. D. Hudson. 1993. Annual variation in stink bug (Heteroptera: Pentatomidae) seasonal abundance and species composition in Georgia soybean and its impact on yield and quality. J. Entomol. Sci. 28: 61-72.

McPherson, R. M., L. D. Newsom, and B. F. Farthing. 1979. Evaluation of four stink bug species from three genera affecting soybean yield and quality in Louisiana. J. Econ. Entomol. 72: 188-194.

McPherson, J. E. and R. M. McPherson. 2000. Stink Bugs of Economic Importance in America North of Mexico. Boca Raton, FL, CRC Press.

Miner, F. D. 1961. Stink bug damage to soybeans. Ark. Farm. Res. 10: 12.

Miner, F. D. 1966. Biology and Control of Stink Bugs in Soybeans. Ark. Agric. Exp. Stn. Bull. 708: 1-40.

Musser, F. R., A. L. Catchot, B. K. Gibson, and K. S. Knighten. 2011. Economic injury leves for southern green stink bugs (Hemiptera: Pentatomidae) in R7 growth stage soybeans. Crop Prot. 30: 63-69.

Nielsen, A. L., G. C. Hamilton and P. W. Shearer. 2011. Seasonal phenology and monitoring of the non-native *Halyomorpha halys* (Hemiptera: Pentatomidae) in soybean. Environ. Entomol. 40: 231-238.

Newsom, L. D., M. Kogan, F. D. Miner, R. L. Rabb, S. G. Turnipseed, and W. H. Whitcomb. 1980. General accomplishments toward better pest control in soybean, pp. 51 - 98. *In* C. B. Huffaker (Ed.), New technology of pest control. Wiley-Interscience Publ, John Wiley & Sons, Inc., New York, NY. 500 pp.

Rudd, W.G. and R. L. Jensen. 1977. Sweep net and ground cloth sampling for insects in soybeans. J. Econ. Entomol. 70: 301-304.

Russin, J. S., M. B. Layton, D. B. Orr, and D. J. Boethel. 1987. Within-plant distribution of, and partial compensation for, stink bug (Heteroptera: Pentatomidae) damage to soybean seeds. J. Econ. Entomol. 80: 215-220.

SAS Institute Inc. 2007. JMP User's Guide. Cary, NC: SAS Institute Inc.

SAS Institute Inc. 2008. SAS/STAT® 9.2 User's Guide. Cary, NC: SAS Institute Inc.

Schaalje, G.B., J. B. McBride, and G. W. Fellingham. 2001. Approximations to distributions of test statistics in complex mixed linear models using SAS Proc MIXED. SUGI (Sas User's Group International) 26, 262. www2.sas.com/proceedings/sugi26/p262-26.pdf.

Simmons, A. M. and K. V. Yeargan. 1990. Effect of combined injuries from defoliation and green stink bug (Hemiptera: Pentatomidae) and influence of field cages on soybean yield and seed quality. J. Econ. Entomol. 83: 599-609.

Thomas, G. D., C. M. Ignoffo, C. E. Morgan, and W. A. Dickerson. 1974. Southern green stink bug: influence on yield and quality of soybeans. J. Econ. Entomol. 67: 501-503.

Todd, J. W. and S. G. Turnipseed. 1974. Effects of southren green stink bug damage on yield and quality of soybeans. J. Econ. Entomol. 67: 421-426.

Wada, T., N. Endo, and M. Takahashi. 2006. Reducing seed damage by soybean bugs by growing small-seeded soybeans and delaying sowing time. Crop Prot. 25: 726-731.

Whigham, K. 2002. Iowa soybean production history. Iowa State University University Extnesion pub. no. IC-488. http://www.ipm.iastate.edu/ipm/icm/2002/1-21-2002/prodhistory.html. Accessed 13 April 2011.

Yeargan, K. V. 1977. Effects of green stink bug damage on yield and quality of soybeans. J. Econ. Entomol. 70: 619-622.

Young, S. Y., J. K. Greene, and G. M. Lorenz. 2008. Damage to soybean by *Acrosternum hilare* (Say) (Heteroptera: Pentatomidae). J. Entomol. Sci. 43: 257-267.

Table 2.1. Stink bug species infesting field cages at sites in Maryland, Virginia, and Delaware from 2007-2009. Stink bugs were placed in cages enclosing 0.3 row m at several soybean development stages for three weeks. All stink bugs were adults unless specified

Location	Soybean Development Stage			
	R4	R5	R6	R7
Salisbury, MD 2007	*E. servus*, *A. hilare*, *A. hilare* nymphs	*E. servus*, *A. hilare*	*E. servus*, *A. hilare*	
Suffolk, VA 2007	*A. hilare*	*A. hilare*	*A. hilare*	*A. hilare*
Apiary site, Newark, DE 2007	*E. servus*, *A. hilare*	*A. hilare*	*A. hilare*	
Allen site, Newark, DE 2007	*E. servus*, *A. hilare*	*A. hilare*	*A. hilare*	
Beltsville, MD 2008	*E. servus*, *A. hilare*	*E. servus*	*E. servus*, *A. hilare*	
Suffolk, VA 2008	*A.* hilare, *A. hilare* nymph	*A. hilare*, *A. hilare* nymph	*A. hilare*, *A. hilare* nymph	*A. hilare*, *A. hilare* nymph
Newark, DE 2008	*A. hilare*	*A. hilare*	*A. hilare*	
Georgetown, DE 2008	*A. hilare*	*A. hilare*	*A. hilare*	
Beltsville, MD 2009	*A. hilare*		*A. hilare*	
Suffolk, VA 2009	*A. hilare*	*A. hilare* nymph	*A. hilare*	
Newark, DE 2009			*A. hilare*	
Georgetown, DE 2009			*A. hilare*	

Table 2.2. Effects of stink bug species and soybean development stage on soybean seed quality and yield

		Destroyed pod percentage			Damaged/diseased seed percentage			Dry soybean yield (g)			Seed number per cage		
Location	Source[a]	*F*	df	*P*	*F*	df	*P*	*F*	df	*P*	*F*	df	*P*
Beltsville, MD 2007	Sb species	0.17	2, 142	0.847	0.22	2, 140	0.799	0.08	2, 15.8	0.924	0.10	2, 140	0.901
	R stage	0.34	2, 143	0.711	0.40	2, 6.81	0.6872	2.01	2, 10.3	0.184	2.84	2, 12.9	0.095
	Sb species x R stage	0.47	4, 143	0.757	0.84	4, 139	0.5024	1.93	4, 14.3	0.161	2.66	4, 139	0.035*
Beltsville, MD 2008	*E. servus*	---	---	---	---	---	---	0.58	1, 110	0.447	0.41	1, 111	0.523
	R stage	---	---	---	---	---	---	4.62	2, 10.9	0.035*	4.20	2, 5.5	0.078
	E. servus x R stage	---	---	---	---	---	---	0.20	2, 110	0.817	0.69	2, 110	0.504
Beltsville, MD 2008	SB species	---	---	---	---	---	---	2.15	2, 88.9	0.123	2.57	2, 81.1	0.083
	R stage	---	---	---	---	---	---	9.95	1, 6.54	0.018*	6.23	1, 7.18	0.040*
	SB species x R stage	---	---	---	---	---	---	0.08	2, 88.9	0.926	0.43	2, 81.1	0.652
Suffolk, VA 2008	SB life stage	1.21	2, 58	0.304	0.59	2, 60	0.559	0.74	2, 38.2	0.486	0.40	2, 52	0.669
	R stage	23.57	3, 58	<0.001*	0.80	3, 60	0.498	48.93	3, 7.11	<0.001*	84.81	3, 6	<0.001*
	SB life stage x R stage	3.63	6, 58	0.004*	0.15	6, 60	0.988	1.31	6, 31.6	0.280	0.42	6, 52	0.864

*denotes significant treatment effect.

[a]R stage denotes soybean development stage whole plot factor

Table 2.3. Means (±SEM) of various soybean seed quality and yield factors after exposure to one *E. servus* adult, *A. hilare* adult, or *A. hilare* nymph per 0.3 row m. Soybean was infested using field cages at several reproductive development stages for three weeks in 2007

Apiary site, Newark, DE 2007 R4 cages				
Species[a]	Number destroyed pods	Diseased seed percentage *	Dry soybean yield (g)	Seed number per cage
E. servus	17.6 ± 3.8	3.6 ± 4.5	40.9 ± 1.5	245.7 ± 10.0 a
A. hilare	16.0 ± 3.0	2.4 ± 3.1	37.2 ± 2.9	239.9 ± 19.9 a
Control	11.2 ± 1.5	3.5 ± 3.9	32.2 ± 2.4	166.1 ± 20.1 b
Allen site, Newark, DE 2007 R4 cages				
Species	Number destroyed pods*	Diseased seed percentage*	Dry soybean yield (g)	Seed number per cage
E. servus	24.2 ± 3.1	1.2 ± 0.5	26.9 ± 2.9	207.6 ± 18.2
A. hilare	20.3 ± 3.7	0.9 ± 0.1	23.2 ± 1.4	193.9 ± 11.7
Control	23.1 ± 3.8	0.9 ± 0.9	19.1 ± 2.6	154.1 ± 19.6
Salisbury, MD 2007 R4 cages				
Species	Destroyed pod percentage	Diseased seed percentage	Dry soybean yield (g)	Seed number per cage
E. servus	5.7 ± 0.8	2.4 ± 0.4	37.8 ± 3.4	319.8 ± 26.9
A. hilare nymphs	4.0 ± 0.6	1.7 ± 0.5	37.1 ± 5.4	320.0 ± 57.6
A. hilare	5.0 ± 0.7	3.0 ± 0.3	32.9 ± 4.5	275.6 ± 35.3
Control	5.7 ± 0.7	3.1 ± 0.9	42.4 ± 2.0	329.8 ± 15.2
Salisbury, MD 2007 R4-R6 cages				
Species	Destroyed pod percentage	Diseased seed percentage	Dry soybean yield (g)	Seed number per cage
E. servus	4.6 ± 0.2	1.9 ± 0.1	43.7 ± 1.4	374.1 ± 11.8
A. hilare	4.5 ± 0.6	1.9 ± 0.2	44.4 ± 2.8	380.3 ± 25.6
Control	4.3 ± 0.4	1.7 ± 0.4	43.9 ± 2.6	380.0 ± 22.4
Development stage				
R4	4.7 ± 0.5	2.3 ± 0.4	38.7 ± 2.6	325.6 ± 20.7
R5	4.4 ± 0.3	1.6 ± 0.2	44.7 ± 1.8	379.5 ± 14.3
R6	4.3 ± 0.3	1.6 ± 0.2	48.7 ± 1.7	429.4 ± 14.5

*denotes factors tested using non-parametric Kruskal Wallis test. Values within the same column and treatment followed by a different letter are significantly different (Tukey-Kramer HSD, $P < 0.05$).

[a]All stink bugs in cages were adults, unless specified as being nymphs

Table 2.4. Means (±SEM) of various soybean seed quality and yield factors after exposure to one *E. servus* adult, *A. hilare* adult, or *A. hilare* nymph per 0.3 row m. Soybean was infested using field cages at several reproductive development stages for three weeks in 2008

Beltsville, MD 2008 R4, R6 cages				
Species[a]	Destroyed pod percentage	Damaged seed percentage	Dry soybean yield (g)	Seed number per cage
E. servus	---	---	66.4 ± 2.3	532.6 ± 18.1
A. hilare	---	---	60.3 ± 3.3	478.8 ± 25.3
Control	---	---	63.2 ± 2.8	507.7 ± 19.9
Development stage				
R4	---	---	54.6 ± 2.0 b	562.5 ± 17.2 a
R6	---	---	72.0 ± 1.9 a	450.3 ± 14.2 b
Suffolk, VA 2008 R4-R7 cages				
Species[a]	Destroyed pod percentage	Damaged seed percentage*	Dry soybean yield (g)	Seed number per cage
A. hilare nymph	11.2 ± 1.3	71.6 ± 3.5	98.5 ± 6.3	830.2 ± 69.1
A. hilare	11.3 ± 0.8	69.5 ± 3.9	99.1 ± 7.6	854.1 ± 73.4
Control	9.5 ± 1.1	65.8 ± 3.7	103.4 ± 6.2	867.1 ± 66.1
Development stage				
R4	11.8 ± 1.1 ab	68.2 ± 4.0	67.6 ± 3.1 c	488.5 ± 30.0 b
R5	14.9 ± 1.5 a	73.4 ± 4.0	83.1 ± 4.2 b	660.2 ± 41.1 b
R6	10.5 ± 0.7 b	70.4 ± 4.0	114.4 ± 4.3 a	1058.4 ± 45.0 a
R7	5.5 ± 0.4 c	63.9 ± 4.9	136.2 ± 5.7 a	1194.7 ± 48.0 a

*denotes factors tested using Proc GLIMMIX. Values within the same column and treatment followed by a different letter are significantly different (Tukey-Kramer HSD, $P < 0.05$).

[a]All stink bugs in cages were adults, unless specified as being nymphs

Table 2.5. ANOVA results of stink bug comparison cages and stink bug density cages. The 2007 Apiary, Newark, Delaware diseased seed data and destroyed pod and damaged seed percentage data from the 2007 Allen, Newark, Delaware sites were tested using non-parametric Kruskal Wallis test, reported are Chi Square values

		Destroyed pod percentage			Damaged/diseased seed percentage			Cage soybean yield (g)			Total number seed		
Location	Source	*F*	df	*P*	*F*	df	*P*	*F*	df	*P*	*F*	df	*P*
2007 Apiary, DE R4	Bug species	0.21	2, 42	0.814	0.13	2	0.938	8.81	2, 42	0.001*	5.46	2, 42	0.008*
2007 Allen, DE R4	Bug species	0.82	2	0.663	0.001	2	0.997	2.03	2, 41	0.145	2.55	2, 41	0.090
2007 Salisbury, MD R4	Bug species	0.28	3, 55	0.84	0.37	3, 55	0.77	1.85	3, 55	0.149	0.96	3, 55	0.420
2009 Suffolk, VA R5	*A. hilare* nymph density	2.88	3, 28	0.053	2.11	3, 28	0.122	0.75	3, 28	0.532	0.81	3, 28	0.498
2009 Newark, DE R6	*A. hilare* adult density	2.20	3, 28	0.111	1.83	3, 28	0.164	0.38	3, 28	0.766	0.44	3, 28	0.723
2009 Georgetown, DE R6	*A. hilare* adult density	1.53	3, 26	0.231	13.24	3, 28	<0.001*	3.68	3, 28	0.024*	1.58	3, 28	0.216

*denotes significant treatment effect.

Table 2.6. Means (±SEM) of the interaction between stink bug species and soybean development stage on caged soybean seed number and destroyed pods

Seed number per cage, Salisbury, MD 2007				
Species[a]	Development stage			
	R4	R5	R6	
E. servus	346.8 ± 26.8 ab	378.8 ± 17.8 ab	396.9 ± 18.3 a	
A. hilare	350.7 ± 57.6 ab	371.7 ± 73.1 ab	418.6 ± 29.7 a	
Control	279.2 ± 35.3 b	388.1 ± 20.1 ab	472.8 ± 33.5 a	
Destroyed pod percentage interaction, Suffolk, VA 2008				
Species	Development stage			
	R4	R5	R6	R7
A. hilare	9.7 ± 1.1 bcde	20.5 ± 1.6 a	10.4 ± 1.3 bcd	4.4 ± 0.6 e
A. hilare nymph	14.4 ± 0.3 ab	13.0 ± 0.3 abc	11.1 ± 1.1 bcd	6.4 ± 0.8 cde
Control	11.3 ± 1.3 bcd	11.2 ± 1.6 bcd	10.0 ± 1.3 bcd	5.7 ± 0.4 de

Values followed by a different letter are significantly different (Tukey-Kramer HSD, $p < 0.05$).
[a]All stink bugs in cages were adults, unless specified as being nymphs

Table 2.7. Means (±SEM) of various soybean seed quality and yield factors after exposure to one *E. servus* adult per 0.3 row m. Soybean was infested using field cages at several reproductive development stages for three weeks in 2008

Beltsville, MD 2008 *E. servus* cages		
Species	Dry soybean yield (g)	Seed number per cage
E. servus	67.3 ± 1.7	542.3 ± 13.5
Control	65.3 ± 2.3	528.4 ± 18.3
Development stage		
R4	56.4 ± 2. b	463.7 ± 16.6
R5	69.3 ± 2.1 ab	562.9 ± 17.1
R6	73.2 ± 2.3 a	579.4 ± 17.6

Values within the same column and treatment followed by a different letter are significantly different (Tukey-Kramer HSD, $P < 0.05$).

Table 2.8. Means (±SEM) of various soybean seed quality and yield factors after exposure to zero, one, two, and four *A. hilare* nymphs per 0.3 row m. Soybean was infested using field cages at the R5 development stage for three weeks in 2009

Suffolk, VA 2009 *A. hilare* nymph R5 cages				
Bug density per 0.3 row m	Destroyed pod percentage	Damaged seed percentage	Dry soybean yield (g)	Seed number per cage
0	5.0 ± 0.7	57.6 ± 3.2	101.8 ± 7.5	872.6 ± 59.7
1	6.3 ± 0.6	60.0 ± 1.8	88.5 ± 7.1	821.0 ± 43.7
2	7.2 ± 0.8	51.9 ± 5.4	97.3 ± 5.1	899.1 ± 27.3
4	7.8 ± 0.8	65.0 ± 3.6	91.5 ± 7.3	815.4 ± 43.6

Table 2.9. Means (±SEM) of various soybean seed quality and yield factors after exposure to zero, one, two, and four *A. hilare* adults per 0.3 row m. Soybean was infested using field cages at the R4 and R6 development stages for three weeks in 2009

Beltsville, MD 2009 *A. hilare* cages				
Bug density per 0.3 row m	Destroyed pod percentage	Damaged seed percentage	Dry soybean yield (g)	Seed number per cage
0	---	12.2 ± 0.9 b	67.7 ± 5.5	623.5 ± 48.7
1	---	13.2 ± 0.7 b	65.7 ± 5.4	607.3 ± 49.6
2	---	15.5 ± 1.4 ab	64.7 ± 2.7	622.8 ± 25.8
4	---	18.8 ± 1.4 a	62.7 ± 5.0	616.0 ± 51.6
Development stage				
R4	---	16.4 ± 0.6 a	61.2 ± 3.0	565.7 ± 27.2
R6	---	13.4 ± 0.9 b	69.2 ± 3.1	669.1 ± 28.1
Suffolk, VA 2009 *A. hilare* cages				
Bug density per 0.3 row m	Destroyed pod percentage	Damaged seed percentage	Dry soybean yield (g)	Seed number per cage
0	5.1 ± 0.4 c	50.5 ± 3.4	75.8 ± 3.3	731.7 ± 26.0
1	7.1 ± 0.6 bc	58.8 ± 4.2	68.0 ± 3.9	657.6 ± 31.5
2	8.0 ± 0.5 b	61.9 ± 4.1	70.8 ± 3.4	709.6 ± 33.1
4	11.2 ± 1.1 a	65.5 ± 4.8	70.0 ± 3.5	684.0 ± 30.0
Development stage				
R4	8.6 ± 0.8	58.0 ± 3.4	71.6 ± 2.2	686.8 ± 16.8
R6	7.1 ± 0.4	60.3 ± 2.6	70.7 ± 2.8	704.6 ± 25.4

Values within the same column and treatment followed by a different letter are significantly different (Tukey's-adjustment $P < 0.05$).

Table 2.10. Effects of *A. hilare* infestation and soybean development stage on soybean seed quality and yield during 2007-2009

		Destroyed pod percentage			Damaged/diseased seed percentage			Dry soybean yield (g)			Seed number per cage		
Location	Source[a]	*F*	df	*P*	*F*	df	*P*	*F*	df	*P*	*F*	df	*P*
Salisbury, MD 2007	SB presence	0.03	1, 57	0.862	0.32	1, 54.2	0.576	0.00	1, 55.4	0.983	0.00	1, 56	0.947
	R stage	0.08	2, 57	0.928	0.29	2, 7.48	0.753	2.00	0.180	0.180	3.35	2, 11.8	0.070
	SB presence x R stage	0.35	2, 57	0.706	0.47	2, 53.7	0.630	1.37	2, 54.8	0.263	1.22	2, 55.4	0.303
Suffolk, VA 2007	SB presence	4.40	1, 63	0.040*	4.46	1, 63	0.039*	1.02	1, 56.1	0.318	0.01	1, 57	0.941
	R stage	13.65	3, 63	<0.001*	13.71	3, 63	<0.001*	3.92	3, 8.07	0.054	15.16	3, 8	0.001*
	SB presence x R stage	2.34	3, 63	0.082	2.36	3, 63	0.079	1.17	3, 56.1	0.328	2.50	3, 57	0.069
Allen, Newark, DE 2007	SB presence	0.88	1, 111	0.351	0.00	1, 110	0.980	4.40	1, 19.7	0.049*	5.37	1, 29.6	0.028*
	R stage	4.52	2, 111	0.013*	3.70	2, 110	0.028*	0.97	2, 18.1	0.398	0.31	1, 29.5	0.737
	SB presence x R stage	1.67	2, 111	0.193	0.48	2, 110	0.619	0.12	2, 18.2	0.891	0.04	2, 29.6	0.958
Apiary, Newark, DE 2007	SB presence	0.00	1, 32.8	0.967	0.32	1, 111	0.574	3.08	1, 104	0.082	2.55	1, 111	0.113
	R stage	1.64	2, 23.1	0.215	1.63	1, 111	0.201	1.78	2, 6.25	0.245	2.07	2, 111	0.131
	SB presence x R stage	0.44	2, 23.2	0.652	1.71	2, 111	0.185	4.71	2, 104	0.011*	3.20	2, 111	0.045*
Beltsville, MD 2008	SB presence	---	---	----	---	---	---	0.71	1, 44.2	0.403	1.21	1, 43	0.278
	R stage	---	---	----	---	---	---	8.77	1, 5.68	0.027*	3.02	1, 6.19	0.132
	SB presence x R stage	---	----	---	---	---	---	0.17	1, 44.2	0.685	0.22	1, 43	0.638
Suffolk, VA 2008	SB presence	1.88	1, 38	0.179	2.20	1, 32	0.148	0.30	1, 32	0.588	0.00	1, 32	0.963
	R stage	27.32	3, 38	<0.001*	1.09	3, 6	0.423	35.92	3, 6	<0.001*	58.31	3, 6	<0.001*
	SB presence x R stage	7.27	3, 38	0.001*	0.44	3, 32	0.725	0.91	3, 32	0.449	0.35	3, 32	0.788
Newark, DE 2008	SB presence	---	---	---	12.31	1, 27	0.002*	7.44	1, 23.4	0.012*	3.01	1, 23.3	0.0961
	R stage	---	---	---	1.22	2, 27	0.312	0.92	2, 4.15	0.468	0.37	2, 4.12	0.714
	SB presence x R stage	---	---	---	1.54	2, 27	0.233	2.70	2, 23.4	0.088	3.19	2, 23.3	0.059
Georgetown, DE 2008	SB presence	---	---	---	14.84	1, 30	0.001*	0.35	1, 18.4	0.560	0.1	1, 23.6	0.739
	R stage	---	---	---	7.31	2, 30	0.003*	3.51	2, 12.4	0.062	2.57	2, 16.7	0.106
	SB presence x R stage	---	---	---	3.03	2, 30	0.063	3.72	2, 12.4	0.054	4.29	2, 16.7	0.031*
Beltsville, MD 2009	SB density	---	---	---	7.05	3, 18.5	0.002*	0.20	3, 29.6	0.899	0.03	3, 29.6	0.992
	R stage	---	---	---	7.59	1, 31.7	0.010*	1.96	1, 9.75	0.193	3.53	1, 9.73	0.090
	SB density x R stage	---	---	---	2.14	3, 18.5	0.130	0.22	3, 29.6	0.883	0.26	3, 29.6	0.855
Suffolk, VA 2009	SB density	17.87	3, 50	<0.001*	2.71	3, 50	0.055	0.94	3, 50	0.429	1.30	3, 46.1	0.287
	R stage	3.39	1, 3	0.163	0.13	1, 6	0.729	0.04	1, 3	0.853	0.38	1, 2.42	0.592
	SB density x R stage	1.43	3, 50	0.246	0.97	3, 50	0.412	0.85	3, 50	0.472	1.73	3, 46.1	0.173

*denotes significant treatment effect.

[a]R stage denotes soybean development stage whole plot factor

Table 2.11. Means (±SEM) of various soybean seed quality and yield factors after exposure to one *A. hilare* adults per 0.3 row m. Soybean was infested using field cages at several reproductive development stages for three weeks in 2007

Salisbury, MD 2007 *A. hilare* threshold cages				
Species	Destroyed pod percentage	Diseased seed percentage	Dry soybean yield (g)	Seed number per cage
A. hilare	4.6 ± 0.6	1.7 ± 0.2	44.5 ± 2.8	382.6 ± 25.6
Control	4.4 ± 0.4	1.9 ± 0.3	44.4 ± 2.4	380.3 ± 19.6
Development stage				
R4	4.5 ± 0.6	2.2 ± 0.7	37.5 ± 3.7	313.9 ± 30.2
R5	4.6 ± 0.5	1.4 ± 0.2	45.5 ± 2.6	385.6 ± 2.2
R6	4.3 ± 0.5	1.7 ± 0.2	50.4 ± 2.6	444.9 ± 22.6
Suffolk, VA 2007 *A. hilare* threshold cages				
Species	Destroyed pod percentage	Damaged seed percentage	Dry soybean yield* (g)	Seed number per cage*
A. hilare	31.5 ± 1.9 a	69.9 ± 1.3 a	33.3 ± 1.5	351.6 ± 17.1
Control	26.9 ± 1.6 b	65.5 ± 2.9 b	35.4 ± 2.5	353.5 ± 24.5
Development stage				
R4	35.1 ± 3.6 a	74.1 ± 2.0 a	28.4 ± 2.3	271.9 ± 21.7
R5	35.9 ± 2.2 a	74.8 ± 2.4 a	29.6 ± 3.1	313.3 ± 30.6
R6	24.9 ± 1.2 b	61.6 ± 2.4 b	39.1 ± 1.5	403.3 ± 19.3
R7	20.1 ± 1.4 b	60.5 ± 2.1 b	40.2 ± 1.7	421.8 ± 17.1
Apiary site, Newark, DE 2007 *A. hilare* threshold cages				
Species	Number of destroyed pods	Diseased seed percentage*	Dry soybean yield (g)	Seed number per cage
A. hilare	15.9 ± 0.9	4.8 ± 01.0	39.1 ± 1.1	232.1 ± 7.8
Control	17.1 ± 2.7	4.0 ± 1.2	35.3 ± 2.1	210.8 ± 17.0
Development stage				
R4	13.6 ± 2.0	5.2 ± 0.6	35.1 ± 2.3	203.0 ± 16.1
R5	17.8 ± 1.5	4.2 ± 0.4	40.1 ± 1.5	238.8 ± 10.7
R6	18.1 ± 1.5	3.7 ± 0.4	36.4 ± 1.5	222.5 ± 12.2
Allen site, Newark, DE 2007 *A. hilare* threshold cages				
Species	Number of destroyed pods*	Diseased seed percentage*	Cage soybean yield (g)	Seed number per cage
A. hilare	21.4 ± 1.1	1.6 ± 0.1	22.8 ± 0.8 a	191.9 ± 7.3 a
Control	23.1 ± 2.0	1.2 ± 0.3	19.2 ± 1.7 b	159.2 ± 13.2 b
Development stage				
R4	26.8 ± 2.7 a	2.0 ± 0.1 a	21.1 ± 1.3	174.0 ± 10.6
R5	19.0 ± 1.3 b	1.3 ± 0.2 ab	22.5 ± 1.4	180.4 ± 9.9
R6	21.0 ± 1.2 ab	0.9 ± 0.1 b	19.4 ± 0.9	172.4 ± 12.2

Values within the same column and treatment followed by a different letter are significantly different (Tukey-Kramer HSD, $P < 0.05$). *denotes variables analyzed using PROC Glimmix.

Table 2.12. Means (±SEM) of various soybean seed quality and yield factors after exposure to one *A. hilare* adult per 0.3 row m. Soybean was infested using field cages at several reproductive development stages for three weeks in 2008

Beltsville, MD 2008 *A. hilare* threshold cages				
Species	Destroyed pod percentage	Damaged seed percentage	Dry soybean yield (g)	Seed number per cage
A. hilare	---	---	60.3 ± 3.3	478.3 ± 25.3
Control	---	---	63.2 ± 2.8	508.9 ± 19.9
Development stage				
R4	---	---	53.1 ± 3.3 b	442.1 ± 28.7
R6	---	---	70.3 ± 2.3 a	545.1 ± 16.7
Suffolk, VA 2008 *A. hilare* threshold cages				
Species	Destroyed pod percentage	Damaged seed percentage	Dry soybean yield (g)	Seed number per cage
A. hilare	11.2 ± 1.3	69.1 ± 3.5	98.7 ± 6.3	867.0 ± 69.1
Control	9.5 ± 0.8	65.8 ± 3.9	103.4 ± 7.6	849.0 ± 73.4
Development stage				
R4	10.5 ± 0.9 b	68.7 ± 4.6	65.0 ± 3.4 d	475.6 ± 29.5 b
R5	15.8 ± 1.8 a	72.3 ± 5.0	85.1 ± 5.3 c	679.0 ± 53.4 b
R6	10.2 ± 0.9 b	67.6 ± 5.0	115.0 ± 4.8 b	1079.1 ± 53.0 a
R7	4.9 ± 0.4 c	61.4 ± 6.3	139.0 ± 7.2 a	1198.6 ± 64.3 a
Newark, DE 2008 *A. hilare* threshold cages				
Species	Destroyed pod percentage	Damaged seed percentage	Dry soybean yield (g)	Seed number per cage
A. hilare	---	5.5 ± 0.7 a	47.3 ± 1.4	357.9 ± 9.9
Control	---	2.9 ± .7 b	52.2 ± 1.4	336.6 ± 11.6
Development stage				
R4	---	5.0 ± 0.9	49.2 ± 2.2	338.6 ± 14.5
R5	---	4.2 ± 1.2	48.5 ± 1.8	347.8 ± 11.9
R6	---	3.4 ± 0.6	51.6 ± 1.3	355.4 ± 14.0
Georgetown, DE 2008 *A. hilare* threshold cages				
Species	Destroyed pod percentage	Damaged seed percentage	Dry soybean yield (g)	Seed number per cage
A. hilare	---	19.0 ± 2.4 a	18.5 ± 2.3	143.3 ± 10.3
Control	---	10.1 ± 1.0 b	19.4 ± 2.4	147.5 ± 9.4
Development stage				
R4	---	11.2 ± 1.4 b	18.0 ± 2.3	140.7 ± 9.6 ab
R5	---	13.1 ± 1.7 b	22.0 ± 2.5	164.9 ± 11.8 a
R6	---	20.6 ± 3.3 a	16.9 ± 2.6	130.7 ± 12.9 b

Values within the same column and treatment followed by a different letter are significantly different (Tukey-Kramer HSD, $P < 0.05$).

Table 2.13. Means (±SEM) of significant interactions between stink bug infestation and soybean development stages on destroyed pods and caged seed number from the 2008 *A. hilare* adult cages

Destroyed pod percentage interaction, Suffolk, VA 2008				
Species	Development stage			
	R4	R5	R6	R7
A. hilare	31.4 ± 1.1 bc	46.9 ± 1.6 a	32.6 ± 1.3 bc	20.2 ± 0.6 d
Control	34.0 ± 1.3 b	33.6 ± 1.6 b	32.0 ± 1.3 bc	23.9 ± 0.4 cd
Seed number per cage, Georgetown, DE 2008				
Species	Development stage			
	R4	R5	R6	
A. hilare	126.7 ± 11.9 b	187.7 ± 13.7 a	115.7 ± 12.5 b	
Control	154.7 ± 13.6 ab	142.2 ± 14.6 ab	145.7 ± 22.2 ab	

Values followed by a different letter are significantly different (Tukey-Kramer HSD, $p < 0.05$).

Table 2.14. Means (±SEM) of the interaction between stink bug infestation and soybean development stages on caged soybean yield and seed number in 2007

Dry soybean yield (g), Apiary site, Newark, DE 2007			
Species	Development stage		
	R4	R5	R6
A. hilare	41.3 ± 2.9	38.0 ± 1.6	38.0 ± 1.6
Control	28.9 ± 2.4	42.1 ± 3.5	34.8 ± 3.9
Seed number per cage, Apiary site, Newark, DE 2007			
Species	Development stage		
	R4	R5	R6
A. hilare	239.9 ± 18.7 ab	224.1 ± 13.0 ab	232.1 ± 12.9 a
Control	166.1 ± 24.4 b	253.4 ± 24.4 a	212.9 ± 25.7 ab

Values followed by a different letter are significantly different (Tukey-Kramer HSD, $p < 0.05$).

Table 2.15. Means (±SEM) of various soybean seed quality and yield factors after exposure to zero, one, two, and four *A. hilare* adults per 0.3 row m. Soybean was infested using field cages at the R6 development stage for three weeks at the 2009 Delaware sites

2009 Newark, DE *A. hilare* cages				
Bug density per 0.3 row m	Destroyed pod percentage	Damaged seed percentage	Dry soybean yield (g)	Seed number per cage
0	5.4 ± 0.8	12.9 ± 2.1	26.4 ± 3.3	205.6 ± 22.7
1	4.9 ± 0.9	12.6 ± 1.8	21.0 ± 3.0	166.1 ± 24.3
2	4.2 ± 0.9	14.7 ± 1.9	23.5 ± 4.6	173.6 ± 31.3
4	8.2 ± 1.9	18.9 ± 2.7	22.9 ± 3.2	180.5 ± 23.7
2009 Georgetown, DE *A. hilare* cages				
Bug density per 0.3 row m	Destroyed pod percentage	Damaged seed percentage	Dry soybean yield (g)	Seed number per cage
0	5.4 ± 0.8	5.9 ± 1.0 b	28.3 ± 2.1 a	234.5 ± 17.5
1	4.9 ± 0.9	6.8 ± 0.7 b	28.2 ± 2.0 a	222.9 ± 14.0
2	4.2 ± 0.9	9.2 ± 1.0 b	27.8 ± 1.7ab	237.3 ± 16.4
4	8.2 ± 1.9	23.9 ± 5.9 a	20.8 ± 1.8b	193.8 ± 15.2

Values within the same column and treatment followed by a different letter are significantly different (Tukey's-adjustment $P < 0.05$).

Chapter 3

Evaluation of seed and pod injury and damage by *Halyomorpha halys* and *Acrosternum hilare*

Abstract

The brown marmorated stink bug, *Halyomorpha halys* Stål (Hemiptera: Pentatomidae) is a recently introduced phytophagous stink bug pest in mid-Atlantic soybean. Currently, there is little information indicating how this new pest should be managed in soybean or if economic thresholds should be adjusted. In 2009, field cage studies in Beltsville, MD were used to evaluate seed and pod injury caused by *H. halys* compared with the green stink bug, *Acrosternum hilare* Say (Hemiptera: Pentatomidae) after a three week infestation period. In 2010 and 2011, additional field cage studies were conducted at the same location as well as in Suffolk, VA to evaluate *H. halys* injury to several different soybean development stages by infesting plants with different *H. halys* densities for two weeks. Cage plots were harvested, and subsamples were taken to determine seed quality. Results showed that soybean seed injury caused by *H. halys* was similar to that of *A. hilare*. The full flowering R2 soybean development stage was least affected by *H. halys* feeding, while the full pod R4 stage was the most sensitive, and slightly more sensitive than the full seed R6 stage. While no yield loss was associated with stink bug densities, significant seed injury to R4 caged soybean was noted at even low densities. Current thresholds for *A. hilare* should be applied to *H. halys*.

Keywords: *Halyomorpha halys, Acrosternum hilare (Chinavia hilaris),* soybean, thresholds

The brown marmorated stink bug, *Halyomorpha halys* Stål (Hemiptera: Pentatomidae), is a recently introduced invasive pest from Asia. It was first found in Allentown, Pennsylvania around 1996, but not correctly identified until 2001 (Hoebeke and Carter 2003). *Halyomorpha halys* undergoes between four and six generations in the southern parts of its range in China (Hoffman 1931). In the mid-Atlantic, it appears to have one or two generations a year, and its range is likely to be very similar to that of *Acrosternum hilare* Say (*Chinavia hilaris*) (Hemiptera: Pentatomidae), which also exhibits similar generational biology (Nielsen et al. 2008, Holtz and Kamminga 2010). *Halyomorpha halys'* populations now dominate the native stink bug complex in the mid-Atlantic region (Nielsen and Hamilton 2009). It is now reported from over thirty states, including Oregon and California (Belisle 2011). It was first discovered in Virginia in 2004, and is now considered to be a major pest statewide (Day et al. 2011).

This species' potential for being a severe agricultural pest in the United States was not recognized until very recently. This could be due in part because of its wide host range. It was not until large population abundances had built up that it was found to be economically damaging on various crops, including orchard fruit (apples, peaches, cherries, etc.) small fruit (grapes and berries), field crops (corn and soybean), vegetables, ornamentals, and many native shrubs and trees (Hamilton 2009).

Only a couple of studies have compared injury caused by native stink bug species or by native and introduced stink bug species on field crops. Injury differences have been detected in cotton, with the brown stink bug, *Euschistus servus* Say (Hemiptera: Pentatomidae), causing more damage than *E. quadrator* Rolston (Hemiptera: Pentatomidae) (Hopkins et al. 2009). Miner (1966) concluded that *E. servus* did not injure soybean seeds as severely as *A. hilare* or *Nezara viridula* L. (Hemiptera: Pentatomidae), yet in other studies, *N. viridula* was the most

damaging species, *E. servus* second most, and *A. hilare* less damaging, depending on variety (McPherson et al. 1979). Current economic thresholds combine all three species into a stink bug complex or use the same threshold level for each individual species. *Halyomorpha halys* is often more abundant in soybean than the native stink bugs, and high populations cause severe damage to soybean (Nielsen et al. 2011). Few studies have addressed the direct impact of *H. halys* on soybeans and how it compares to native stink bug species. This study provides results of field cage studies with that objective.

Materials and Methods

2009. Small nylon hexagonal-mesh sleeve cages 69 × 105 cm enclosing 3 plants in 0.3 row m were infested with *A. hilare* and *H. halys*. Southern States 'RT4440N', an indeterminate maturity group (MG) 4.4 variety soybean was planted on 25 June at University of Maryland's Central Maryland Research and Education Center in Beltsville, Maryland (the Beltsville site). Adult and fifth instar *H. halys* and *A. hilare* were collected from local commercial soybean fields using sweep nets, transported to the study site and held overnight to ensure vitality before being released into cages. Densities of zero, one, two, and four *H. halys* adults and *A. hilare* adults or nymphs per 0.3 row m were placed into cages enclosing soybean plants at the R4 (full pod) soybean development stage (Fehr et al. 1971). At the R6 (full seed) soybean development stage (Fehr et al. 1971), *A. hilare* adults and *H. halys* adults or nymphs were introduced into additional cages at the same densities. Cages were arranged in a split plot experimental design with species as the whole plot and density as the subplot factors. After a three week infestation period, stink bugs were removed and cages were resealed until harvest. At harvest, soybean plants were removed from the cages and taken into the laboratory to determine seed quality and yield data. Aborted pods were estimated by collecting pods from the ground within the cage, flat pods were

categorized as such by counting pods that did not contain seed, and normal pods that had seed were tallied. Normal pods were hand shelled and seeds were visually examined for normal seeds, seeds with evidence of stink bug punctures (discolored spots with damage), diseases (including molds and purple stain), and seed coat deformations (including shriveled, crinkled, and immature seeds). Yields were determined from the adjusted dry seed weight and cage seed number.

2010 Suffolk, VA. Due to concerns of cage effect in the 2009 small cages, larger field cages were used at two sites in 2010. At the Virginia Tech Tidewater Agricultural Research and Education Center in Suffolk, VA (the Suffolk site), after wheat harvest, 'Asgrow 5605,' an indeterminate MG 5.6 soybean variety, was planted at a rate of 10 seeds per 0.3 m on 0.91 m row centers on 15 June. Cages measuring 2 × 2 × 2 m enclosing four row m plots were infested with the same densities of *H. halys* as was used in the 2009 study, and cages were placed in the field at soybean development stages R4 and R6 using a split plot design. Soybean development stage was the main plot factor and stink bug density was the subplot factor. The perimeter of the cages was buried 0.3 m to provide stability and to prevent stink bugs from escaping. Cages were infested with field-collected *H. halys* fifth instars, and density treatments were replicated four times. After a 14 day infestation period, stink bugs were eliminated by spraying plots with methyl parathion (Methyl 4 EC, Cheminova Inc., Wayne, NJ) at a rate of 0.56 kg (AI)/ha. Cages were removed once it had been determined that there were no surviving stink bugs. Two additional sprays of β-cyfluthrin (Baythroid® XL, Bayer Crop Sciences, Research Triangle Park, NC) and acephate (Orthene® 97, AMVAC Chemical Corp, Los Angeles, CA) were applied to field plots at rates of 0.03 and 0.55 kg (AI)/ha, respectively, in order to suppress native stink bugs from infesting the field. At maturity, five consecutive plants were removed from each cage

plot before harvest and placed in paper bags, taken back into the laboratory, and hand shelled to obtain data on pods and seed quality. A 0.3 × 1 m area was examined for shed pods. Plants remaining in the cage plots were then removed and threshed with a stationary small plot thresher to determine cage plot soybean yield. Data were collected as previously described.

2010 Beltsville, MD. A similar cage study was conducted at the Beltsville site in Maryland. 'Asgrow 4404' (MG 4.4) soybean, also an indeterminate variety, were planted on 1 July at a rate of four seeds per 0.3 row m, and rows were spaced 0.76 m apart. Cages measuring 2 × 4 × 2 m and enclosing 6.1 row m were infested with field collected fifth instar *H. halys*. Cages were erected at soybean development stages R2 (full flower) (Fehr et al. 1971) and R4. Plot design, stink bug densities, and infestation period were the same as at the Suffolk site. Plots were sprayed three times after cages were removed with lambda-cyhalothrin (Warrior® II, Syngenta Crop Protection, Inc., Greensboro, NC) at a rate of 0.03 kg (AI)/ha. All plots were harvested with a Swanson small plot thresher on 3 November.

2011. At the Suffolk site, soybean variety 'Asgrow 5605' was planted on 15 June. Due to sample variability in 2010, replication was increased to six, and a density of eight stink bugs per 0.3 row m was added to the plot design. Plots were thinned to 7 plants per 0.3 row m after germination, cages contained only 3.1 row m, and cages were erected once soybean plants had reached the R4 and R6 soybean development stage. After the two week infestation period, cages were sprayed with a tank mix of bifenthrin (Brigade®, FMC Corp, Philadelphia, PA) at a rate of 0.11 kg (AI)/ha and methyl parathion at the same rate as the 2010 study at this location. At maturity a five plant subsample was taken from cage plots on 8 November, and the whole plots were harvested on 1 December. Data collection procedures were the same as in 2010. At the Beltsville site, the one bug per 0.3 row m density was not used due to cage availability. 'Asgrow

4404' soybean was planted on 22 June and harvested on 16 October. The field was planted using a seeding rate of 8.6 plants per 0.3 row m. Cages were erected when the field had reached the R4 and R6 soybean development stages. Cages were infested with field collected stink bugs at densities of zero, two, four, and eight bugs per 0.3 row m for two weeks, after which they were removed. Destroyed pod data were not taken, but other data collection procedures were similar to the Suffolk site.

Data analysis. Punctured seed, shriveled seed, and wrinkled seed percentages were combined into a single category: percentage damaged seed. Similarly, flat pods and aborted pods were combined into a destroyed pod percentage category. In 2009, the data from the split-plot designed cage studies were analyzed using PROC MIXED in SAS with species as the whole plot and density the subplot factors. Degrees of freedom were obtained using the Kenwood Rogers method (Schaalje et al. 2001), and replicates and soybean development stage were included as random factors (SAS Institute Inc. 2008). The damaged seed percentage was square root transformed in the 2009 R6 data. In 2010 and 2011, data from the split-plot designed cage studies from both locations were analyzed using PROC MIXED in SAS with soybean development stage as the whole plot and stink bug density as the subplot factors. Damaged seed percentage data from the 2010 Beltsville site were square root transformed. Due to non-normal whole cage soybean yield data from the 2010 Beltsville site, data were analyzed with PROC GLIMMIX using a Poisson distribution (SAS Institute Inc. 2008) fitted with an overdispersion parameter to minimize the conditional Pearson residual (Littell et al. 2006).

Results

Species comparison. The two stink bug species and the two different life stages did not cause significantly different levels of injury to soybean seed or yield in either R4 or R6-initiated

cages (Tables 3.1, 3.2, 3.3). *Acrosternum hilare* nymph infested, R4-initated cages had significantly lower yield (Tables 3.1, 3.2), and an interaction between density and species on the cage seed number (Tables 3.1, 3.4). However, the control cages from *A. hilare* nymph plots had much lower yield than the control cages associated with the other stink bugs. Therefore, it does not seem that that this reduction in yield or cage seed number was caused by stink bug feeding. No significant difference in soybean yield or cage seed number was detected between *H. halys* adults and nymphs and *A. hilare* adults in the R6-initiated cages (Tables 3.1, 3.3).

In the R4-initiated cages, densities of one bug per 0.3 row m and higher were enough to cause a significant increase in the damaged seed percentage (Tables 3.1, 3.2) while densities of two stink bugs per 0.3 row m and higher caused a significant increase in the damaged seed percentage in R6-initiated cages (Tables 3.1, 3.3). Increased density did not result in yield loss or a change in cage seed number at either soybean development stage (Tables 3.1, 3.2, 3.3). Caged soybean plants infested by *A. hilare* nymphs generally had lower seed number than from other cages, but meaningful trends were hard to interpret (Tables 3.1, 3.4). Neither species nor density significantly affected seed number in the R6-initiated cages (Tables 3.1, 3.3).

***Halyomorpha halys* injury to soybean.** All 2010 data from Beltsville and Suffolk are summarized in Tables 3.5 and 3.6. All soybean plants from Suffolk had a high percentage of damaged seed, even in the control cages and damaged seed was not associated with either *H. halys* density or soybean phenology (Table 3.5). The highest stink bug density (four bugs per 0.3 row m) in the R4-initiated cages resulted in the highest percentage of destroyed pods compared with all other densities from the R4 and R6-initiated cages (Tables 3.5, 3.7). Five plant-sample seed weight or seed number were not influenced by either *H. halys* density or soybean development stage (Tables 3.5, 3.6), although the highest stink bug densities tended to reduce

seed weight in the R4-initiated cages. However, whole cage soybean yield was unaffected (Table 3.5).

At the 2010 Beltsville site, where cage infestations were initiated at earlier points in soybean reproductive development, densities as low as one stink bug per 0.3 row m resulted in a significant increase in the damaged seed percentage (Tables 3.5, 3.6); this was most pronounced in the R4-initiated cages, which exhibited 25% more seed injury overall than the R2-caged soybean. Stink bugs infesting R4 stage soybean caused a greater percentage of destroyed pods compared with the R2-initiated cages (Tables 3.5, 3.6, 3.7). Cages with the highest *H. halys* densities had a corresponding decrease in five-plant sample seed weight from the R4-initiated cages (Tables 3.5, 3.6). Cage seed number was not affected (Table 3.5). Yields from the whole cages were significantly reduced in the R4-initiated cages compared with the R2-initiated cages (Tables 3.5, 3.6).

In 2011, stink bugs destroyed pods at the highest densities only in the R4-initiated cages at the Suffolk site (Tables 3.8, 3.9). At Beltsville, R6-initiated cages were heavily impacted by both stink bugs and cage effect (Tables 3.8, 3.10). The highest two stink bug densities (four and eight bugs per 0.3 row m) resulted in significantly increased damaged seed at both 2011 sites (Tables 3.8, 3.10) and a decrease in the subsample seed weight in Suffolk (Tables 3.8, 3.9). At Beltsville, stink bug densities did not result in a definitive subsample seed weight loss. Whole cage soybean yield was lower in the Maryland R6-initiated cages (Table 3.8, 3.10).

Discussion

Results from the 2009 study indicate that *H. halys* does not appear to cause greater injury to soybean seed and pods than the native *A. hilare*. The introduced red banded stink bug, *Piezodorus guildinii* Westwood (Hemiptera: Pentatomidae), on the other hand, is much more

injurious to soybean than native stink bugs, resulting in much lower economic thresholds in states where it is now the predominant stink bug species in soybean (Corrêa-Ferreira and de Azevedo 2002, Baldwin et al. 2010). This increase in damage is probably due to increased salivary secretion (Depieri and Panizzi 2011). Analysis of *H. halys* salivary secretions may provide more definitive insight into this species' injury to other crops beside soybean. *H. halys* nymphs caused similar injury to *H. halys* adults when caged on R6 soybean. Native stink bug fifth instars have been shown to cause significantly greater injury than adults in some studies (Yeargan 1977), but not in others (Young et al. 2008). Currently, no economic thresholds separate late instars from adults, and, based on these data, thresholds probably do not need to separate stink bug life stages or *H. halys* from the native species, as has been done for *P. guildinii*.

A plant maturity delay was observed in Suffolk in 2011 when stink bug densities of four and eight stink bugs per 0.3 row m were initiated at the R4 soybean development stage. Plants from these cages retained green leaves longer than plants from the other cages. Coincidentally, the destroyed pod percentages increased significantly at both of these densities, probably stimulating the soybean plants to direct energy towards increased growth (Daugherty et al. 1964). Russin et al. (1987) found a delay in maturity along with compensation at densities slightly lower than one bug per 0.3 row m density (although populations peaked to nine bugs per row-m in field plots). Short infestation periods with high densities (six bugs per 0.3 row m) at the R3-R5 development stags can also delay maturity (Boethel et al. (2000). There was a small increase in subsample seed weight in the R4-inititated cages in Suffolk, suggesting that plants may have been stimulated to compensate for stink bug injury. Based on the studies presented

here, *H. halys* ability to delay soybean reproductive maturity is comparable to *A. hilare*, and is a function of density and duration of infestation.

Native stink bug populations increase in fields during the reproductive stages of soybean, usually in September (McPherson et al. 1993), when many other crops are no longer suitable for reproduction. Stink bug nymphal development on soybean is dependent on the presence of pods that are producing seed (McPherson et al. 1993, Newsom et al. 1980, Panizzi and Alves 1993, Smith et al. 2009). *Halyomorpha halys* also begins to migrate into soybean fields earlier, at the R3 stage, (between seven and 10 days before R4) (Holshouser 2010) than stink bug species native to the mid-Atlantic (Nielsen et al. 2011). Young et al. (2008) found that yield can be reduced when feeding is initiated at R2-R3, although it is unusual for native stink bugs to infest soybean fields in appreciable numbers this early. They also found that seed quality can be reduced during the early reproductive stages at densities of one stink bug per 0.3 row m. In contrast, Boethel et al. (2000) did not see significant reductions in either seed quality or yield when feeding was initiated at the R2 development stage, but the duration of infestation and stink bug densities varied among studies. At the 2010 Beltsville site, stink bug infestation did not result in significant injury to soybean plants in the R2-initiated cages. Although *H. halys* begins to infest fields primarily at the R3 development stage and later, they may not be of immediate concern if they infest fields earlier, unless they occur at extremely high densities.

The R4 development stage was the most sensitive stage to stink bug feeding. Stink bugs increased seed damage in the R4-initiated cages in four of the five study sites. Stink bug populations exceeding threshold levels have been shown to cause significant seed damage and occasionally result in lower seed weight (McPherson et al. 1993). Yield response to stink bug injury is more difficult to document. Soybean plants are known to compensate for seeds

destroyed early in development by increasing the unaffected seeds' size (Russin et al. 1987). Therefore, even though the R4 development stage is more susceptible to stink bug feeding injury, plants can increase the size of undamaged seeds, resulting in equivalent yield to fields with lower stink bug populations (Boethel et al. 2000). Furthermore, at low densities below the economic threshold, stink bug feeding may stimulate soybean plants to increase seed weights, resulting in slight increases in seed weight and yield (Russin et al. 1987), especially in small seeded varieties (Wada et al. 2006). However, relatively high stink bug densities, even for a short time, can cause yield loss when infesting during the R4 development stage, as demonstrated by the 2010 Beltsville results. *Halyomorpha halys* populations exceeding the threshold of one stink bug per 0.3 row m are capable of causing significant injury to soybean seed, even if present for only a short period of time. Regular soybean scouting for *H. halys* should begin as soon as pod production begins so that populations exceeding thresholds can be detected quickly.

Developing soybean pods and seeds from plants at the R6 development stage were generally less affected by stink bug feeding. At the Beltsville 2009 site, higher stink bug densities were required to inflict significant seed damage compared to the R4-initiated cages. Seed quality can still be affected, and seed weight was lower in R6-initiated cages than in R4-initiated cages in one site. This injury appears to be pretty similar to *A. hilare* injury at this development stage, as 14 day infestations of three stink bugs per 0.3 row m can cause significant seed injury (Young et al. 2008). Yield losses due to stink bugs infesting the R6 development stage are much more inconsistent. *Halyomorpha halys* infesting the R6-initiated cages did not result in significant yield losses in 2009 and 2010, but small losses at the Beltsville site in 2011, and *A. hilare* likewise causes inconsistent yield loss at this development stage (Young et al. 2008). As soybeans continue to mature and transition to the R7 development stage, the seed

becomes harder and less suitable for nymph development for some species (Oliveira and Panizzi 2003) but not for others (Panizzi and Alves 1993). Plants also become less susceptible to stink bug feeding injury and yield loss, which may allow thresholds to be raised at this development stage (Musser et al. 2011). *Halyomorpha halys* also caused less injury to soybean plants at the R6 stage, and probably cause even less injury afterwards. In summary, *H. halys* causes similar injury to soybean reproductive structures and yield loss compared to *A. hilare*. Overall, these results suggest that the current threshold used for *A. hilare* in soybean should be applied to *H. halys*.

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References Cited

Baldwin, J., L. Foil, M. Grodner, A. Hammond, G. Henderson, N. Hummel, S. Johnson, R. Leonard, A. Morgan, D. K. Pollet, J. Pyzner, T. E. Reagan, D. Reed, R. N. Story, and M. Stout. 2010. Louisiana Insect Pest Management Guide. LSU AgCenter. http://www.lsuagcenter.com/NR/rdonlyres/B733BE31-7DEA-4264-93A6-AF6FED7023D3/66760/pub1838InsectPestMgmtGuide2010completebook188pgs.pdf. Accessed 6 April 2011.

Belisle, R. F. 2011. Nearly 400 jam into auditorium to learn about stink bugs. Herald-Mail. http://www.herald-mail.com/news/tristate/hm-nearly-400-jam-into-auditorium-to-learn-more-about-stink-bugs-20110209,0,5317231,full.story. Accessed 7 March 2011.

Boethel, D. J., J. S. Russin, A. T. Wier, M. B. Layton, J. S. Mink, and M. L. Boyd. 2000. Delayed maturity associated with southern green stink bug (Heteroptera: Pentatomidae) injury at various soybean phenological stages. J. Econ. Entomol. 93: 707-712.

Corrêa-Ferreira, B. S. and J. de Azevedo. 2002. Soybean seed damage by different species of stink bugs. Agr. Forest Entomol. 4: 145-150.

Daugherty, D. M., M. H. Neustad, C. W. Gehrke, L. E. Cavanah, L. F. Williams, and D. E. Green. 1964. An evaluation of damage to soybeans by brown and green stink bugs. J. Econ. Entomol. 57: 719-722.

Day, E. R., T. McCoy, D. Miller, T. P. Kuhar, and D. G. Pfeiffer. 2011. Brown marmorated stink bug. Virginia Cooperative Extension Publication Number 2902-1100.

Depieri, F. A. and A. R. Panizzi. 2011. Duration of feeding and superficial and in-depth damage to soybean seed by selected species of stink bugs (Heteroptera: Pentatomidae). Neotrop. Entomol. 40: 197-203.

Fehr, W. R., C. E. Caviness, D. T. Burmood, and J. S. Pennington. 1971. Stage of development descriptions for soybeans, *Glycine max* (L.) Merrill. Crop Sci. 11: 929-931.

Hamilton, G. C. 2009. Brown marmorated stink bug. Amer. Entomol. 55: 19-20.

Hoebeke, E. R. and M. E. Carter. 2003. *Halyomorpha halys* (Stal) (Heteroptera: Pentatomidae): a polyphagous plant pest from Asia newly detected in North America. Proc. Entomol. Soc. Wash. 105: 225-237.

Hoffman, W. E. 1931. A Pentatomid pest of growing beans in South China. Peking Nat. Hist. Bull. 5: 25-26.

Holshouser, D. L. 2010. Days to soybean physiological maturity. Virginia Cooperative Extension Pub. No. 3009-1459.

Holtz, T., and K. Kamminga. 2010. Qualitative analysis of the pest risk potential of the brown marmorated stink bug (BMSB), *Halyomorpha halys* (Stål), in the United States. USDA APHIS PPQ pp. 33.

Hopkins, W. B., A. E. Knutson, J. S. Bernal, M. F. Treacy, and C. W. Smith. 2009. Species composition, damage potential, and insecticide susceptibility of stink bugs in cotton in the lower Gulf Coast region of Texas. Southwest. Entomol. 35: 19-32.

Littell, R. C., G. A. Milliken, W. W. Stroup, R. D. Wolfinger, and O. Schabenberger. 2006. SAS for mixed models, second edition. SAS Institute Inc., Cary, NC.

McPherson, R. M., G. K. Douce, and R. D. Hudson. 1993. Annual variation in stink bug (Heteroptera: Pentatomidae) seasonal abundance and species composition in Georgia soybean and its impact on yield and quality. J. Entomol. Sci. 28: 61-72.

McPherson, R. M., L. D. Newsom, and B. F. Farthing. 1979. Evaluation of four stink bug species from three genera affecting soybean yield and quality in Louisiana. J. Econ. Entomol. 72: 188-194.

Miner, F. D. 1966. Biology and control of stink bugs in soybeans. Ark. Agric. Exp. Stn. Bull. 708: 1-40.

Newsom, L. D., M. Kogan, F. D. Miner, R. L. Rabb, S. G. Turnipseed, and W. H. Whitcomb. 1980. General accomplishments toward better pest control in soybean, pp. 51-98. *In* C. B. Huffaker (Ed.), New technology of pest control. Wiley-Interscience Publ, John Wiley & Sons, Inc., New York, NY. 500 pp.

Nielsen, A. L., G. C. Hamilton, and D. Matadha. 2008. Developmental rate estimation and life table analysis for *Halyomorpha halys* (Hemiptera: Pentatomidae). Environ. Entomol. 37: 348-355.

Nielsen, A. L. and G. C. Hamilton. 2009. Life History of the Invasive Species *Halyomorpha halys* (Hemiptera: Pentatomidae) in Northeastern United States. Ann. Entomol. Soc. Am. 102: 608-616.

Nielsen, A. L., G. C. Hamilton and P. W. Shearer. 2011. Seasonal phenology and monitoring of the non-native *Halyomorpha halys* (Hemiptera: Pentatomidae) in soybean. Environ. Entomol. 40: 231-238.

Oliveira, E. D. M., and A. R. Panizzi. 2003. Performance of nymphs and adults of *Piezodorus guildinii* (Westwood) (Hemiptera: Pentatomidae) on soybean pods at different developmental stages. Braz. Arch. Biol. Technol. 46.

Panizzi, A. R. and R. M. L. Alves. 1993. Performance of nymphs and adults of the southern green stink bug (Heteroptera: Pentatomidae) exposed to soybean pods at different phenological stages of development. J. Econ. Entomol. 86: 1088-1093.

Russin, J. S., M. B. Layton, D. B. Orr, and D. J. Boethel. 1987. Within-plant distribution of, and partial compensation for, stink bug (Heteroptera: Pentatomidae) damage to soybean seeds. J. Econ. Entomol. 80: 215-220.

SAS Institute Inc. 2008. SAS/STAT® 9.2 User's Guide. Cary, NC: SAS Institute Inc.

Schaalje, G.B., J. B. McBride, and G. W. Fellingham. 2001. Approximations to distributions of test statistics in complex mixed linear models using SAS PROC MIXED. SUGI (SAS User's Group International) 26, 262. www2.sas.com/proceedings/sugi26/p262-26.pdf.

Smith, J. F., R. G. Luttrell, and J. K. Greene. 2009. Seasonal abundance, species composition, and population dynamics of stink bugs in production fields of early and late soybean in South Arkansas. J. Econ. Entomol. 102: 229-236.

Wada, T., N. Endo, and M. Takahashi. 2006. Reducing seed damage by soybean bugs by growing small-seeded soybeans and delaying sowing time. Crop Prot. 25: 726-731.

Yeargan, K. V. 1977. Effects of Green Stink Bug Damage on yield and quality of soybeans. J. Econ. Entomol. 70: 619-622.

Young, S. Y., J. K. Greene, and G. M. Lorenz. 2008. Damage to soybean by *Acrosternum hilare* (Say) (Heteroptera: Pentatomidae). J. Entomol. Sci. 43: 257-267

Table 3.1. Effects of stink bug species and population density on soybean seed quality and yield at the 2009 Beltsville, MD site

		Damaged seed percentage			Seed number per cage			Cage yield		
Location	Source	*F*	df	*P*	*F*	df	*P*	*F*	df	*P*
Beltsville, MD R4	species	3.16	2, 17.5	0.067	3.75	2, 17.6	0.044*	3.42	2, 45.6	0.041*
	density	24.80	3, 59	<0.001*	1.15	3, 46.8	0.337	1.53	3, 61.9	0.2149
	species x density	0.46	6, 51.8	0.096	2.56	6, 49.9	0.031*	1.92	6, 51.8	0.096
Beltsville, MD R6	species	0.73	2, 5.44	0.5243	0.39	2, 8.83	0.689	0.74	2, 23.9	0.486
	density	11.07	3, 36.8	<0.001*	0.07	3, 3.84	0.973	1.01	3, 71.2	0.393
	species x density	0.91	6, 43	0.4968	1.20	6, 1	0.603	0.86	6, 71.2	0.529

*denotes significant treatment effect.

Table 3.2. Means (±SEM) of various soybean seed quality and yield factors after exposure to zero, one, two, and four *A. hilare* or *H. halys* stink bugs per 0.3 row m. Soybean was infested at the R4 development stage for three weeks in 2009

Beltsville, MD 2009			
Bug density per 0.3 row m	Damaged seed percentage	Dry soybean yield (g)	Seed number per cage
0	12.2 ± 0.6 c	57.9 ± 3.4	523.1 ± 30.0
1	17.0 ± 0.8 b	61.8 ± 3.4	563.4 ± 28.5
2	18.2 ± 0.9 b	58.8 ± 2.6	571.1 ± 22.4
4	23.4 ± 1.1 a	53.2 ± 2.8	525.7 ± 27.1
Species			
A. hilare adult	16.4 ± 0.9	61.4 ± 3.0 a	567.6 ± 27.2 a
A. hilare nymph	19.2 ± 1.1	53.0 ± 2.3 b	504.8 ± 16.2 b
H. halys adult	17.4 ± 1.0	59.4 ± 2.3 ab	565.0 ± 21.5ab

Values within the same column and treatment followed by a different letter are significantly different (Tukey-Kramer HSD, $p < 0.05$).

Table 3.3. Means (±SEM) of various soybean seed quality and yield factors after exposure to zero, one, two, and four *A. hilare* or *H. halys* stink bugs per 0.3 row m. Soybean was infested using field cages at the R6 development stage for three weeks in 2009

Beltsville, MD 2009			
Bug density per 0.3 row m	Damaged seed percentage	Dry soybean yield (g)	Seed number per cage
0	12.4 ± 0.5 b	67.0 ± 2.7	635.8 ± 24.9
1	13.1± 0.6 ab	70.8 ± 2.3	675.8 ± 22.9
2	15.0 ± 0.8 a	69.2 ± 2.6	684.3 ± 29.5
4	16.9 ± 0.6 a	64.6 ± 2.5	643.7 ± 25.8
Species			
A. hilare adult	13.4 ± 0.8	69.2 ± 3.1	669.6 ± 28.1
H. halys nymph	14.5 ± 0.7	69.3 ± 3.1	673.8 ± 34.2
H. halys adult	15.1 ± 0.5	65.2 ± 1.5	636.3 ± 14.3

Values within the same column and treatment followed by a different letter are significantly different (Tukey-Kramer HSD, $p < 0.05$).

Table 3.4. Means (±SEM) of the interaction between stink bug species and density on caged soybean seed number in R4-initiated cages in 2009

Beltsville, MD 2009			
Bug density per 0.3 row m	*A. hilare* adult	*A. hilare* nymph	*H. halys* nymph
0	573.7 ± 68.0 ab	484.9 ± 33.8 b	510.6 ± 45.1 ab
1	532.4 ± 67.4 ab	549.4 ± 29.8 ab	608.3 ± 40.7 ab
2	605.7 ± 32.9 ab	462.9 ± 17.9 b	644.7 ± 31.8 a
4	558.6 ± 61.3 ab	522.1 ± 42.8 ab	496.4 ± 35.8 ab

Values followed by a different letter are significantly different (Tukey-Kramer HSD, $p < 0.05$).

Table 3.5. Effects of stink bug population density on soybean seed quality and yield at the 2010 sites

		Destroyed pod percentage			Damaged seed percentage			5 plant sample weight (g)			Seed number per cage			Dry soybean yield (g)		
Location	Source[a]	*F*	df	*P*	*F*	df	*P*	*F*	df	*P*	*F*	df	*P*	*F*	df	*P*
Beltsville,	density	3.20	3, 18	0.048*	6.77	3, 6.53	0.020*	4.47	3, 76.5	0.042*	1.77	3, 51	0.1957	2.36	3, 17.46	0.106
MD 2010	R stage	90.76	1,3	0.003*	60.73	1, 4.43	0.001*	30.97	1, 11.6	<0.001*	0.82	1, 8.91	0.388	22.63	1, 5.55	0.004*
	R stage x density	3.86	3, 18	0.027*	1.76	3, 6.53	0.248	0.55	3, 7.65	0.661	0.09	3, 15.1	0.964	1.04	3, 17.46	0.398
Suffolk,	density	2.88	3, 21	0.060	1.24	3, 21	0.321	2.21	3, 18	0.122	1.28	3, 7.44	0.350	1.57	3, 21	0.227
VA 2010	R stage	16.5	1, 21	0.001*	0.85	1, 21	0.366	2.96	1, 6	0.136	0.87	1, 12.2	0.369	1.14	1, 21	0.298
	R stage x density	6.68	3, 21	0.002*	1.82	3, 21	0.175	1.12	3, 18	0.368	0.41	3, 7.44	0.751	0.66	3, 21	0.584

*denotes significant treatment effect.

[a]R stage denotes soybean development stage whole plot factor

Table 3.6. Means (±SEM) of various soybean seed quality and yield factors after exposure to zero, one, two, and four *H. halys* nymphs per 0.3 row m. Soybean was infested using field cages at the R4 and R6 development stage for two weeks in 2010

Suffolk, VA 2010					
Bug density per 0.3 row m	Destroyed pod percentage	Damaged seed percentage	5 plant sample weight (g)	Sample seed number	Dry soybean yield (g)
0	10.2 ± 1.1	47.5 ± 6.8	119.8 ± 13.7	1365.5 ± 177.0	708.6 ± 37.3
1	11.3 ± 1.7	53.2 ± 4.8	95.9 ± 10.5	1021.3 ± 102.5	658.8 ± 34.3
2	10.5 ± 1.6	44.3 ± 4.6	93.7 ± 4.8	954.8 ± 46.8	664.5 ± 17.7
4	14.7 ± 2.7	56.3 ± 4.3	83.9 ± 13.4	1036.9 ± 110.4	720.5 ± 23.8
Development stage					
R4	14.2 ± 1.5 a	52.6 ± 2.6	88.1 ± 9.5	1034.9 ± 110.5	700.0 ± 16.6
R6	9.2 ± 0.9 b	48.1 ± 4.5	108.6 ± 6.0	1154.3 ± 61.8	676.3 ± 25.1
Beltsville, MD 2010					
Bug density per 0.3 row m	Destroyed pod percentage	Damaged seed percentage	5 plant sample weight (g)	Sample seed number	Dry soybean yield (g)
0	25.1 ± 6.5 ab	10.8 ± 3.9 b	115.1 ± 6.3 a	222.4 ± 10.7	1,282.0 ± 97.6
1	23.4 ± 4.3b	22.9 ± 7.5 ab	99.6 ± 5.8 ab	235.1 ± 11.3	1,193.9 ± 127.0
2	25.1 ± 4.9 ab	23.3 ± 8.0 ab	116.3 ± 10.8 a	258.9 ± 27.1	1,179.1 ± 103.1
4	33.3 ± 7.5 a	28.9 ± 8.1 a	91.1 ± 9.9 b	205.8 ± 10.9	1,091.7 ± 131.1
Development stage					
R2	14.2 ± 2.0 b	7.2 ± 1.2 b	122.1 ± 5.4a	222.1 ± 8.7	1,444.8 ± 28.7 a
R4	39.3 ± 3.1a	35.8 ± 4.9 a	89.0 ± 4.0 b	239.0 ± 14.8	928.6 ± 58.2 b

Values within the same column and treatment followed by a different letter are significantly different (Tukey-Kramer HSD, $p < 0.05$).

Table 3.7. Means (±SEM) of the interaction between stink bug density and soybean development stage on percentage of destroyed pods from the 2010 cage studies

Suffolk, VA Destroyed pod percentage		
Bug density per 0.3 row m	R4	R6
0	9.6 ± 1.7 b	10.8 ± 1.7 b
1	12.1 ± 2.2 b	10.6 ± 2.9 b
2	13.8 ± 2.1 ab	7.3 ± 0.8 b
4	21.3 ± 2.1 a	8.2 ± 1.0 b
Beltsville, MD Destroyed pod percentage		
Bug density per 0.3 row m	R2	R4
0	8.5 ± 0.9 d	41.8 ± 3.1 ab
1	16.0 ± 4.5 cd	30.8 ± 5.4 bc
2	15.8 ± 1.0 cd	34.5 ± 7.2 ab
4	16.5 ± 6.4 cd	50.0 ± 5.8 a

Values followed by a different letter are significantly different (Tukey-Kramer HSD, $p < 0.05$).

Table 3.8. Effects of stink bug population density on soybean seed quality and yield at the 2011 sites

		Destroyed pod percentage			Damaged seed percentage			5 plant sample weight (g)			200 seed sample weight (g)			Dry soybean yield (g)		
Location	Source[b]	*F*	df	*P*	*F*	df	*P*	*F*	df	*P*	*F*	df	*P*	*F*	df	*P*
Beltsville,	density	3.12	3, 18	0.052	5.13	3, 21	0.005*	3.01	3, 24	0.05*	0.58	3, 17.8	0.635	1.06	3, 21	0.386
MD 2011	R stage	92.23	3, 18	0.002*	5.13	1, 21	0.034*	1.23	1, 24	0.278	0.37	1, 8.16	0.560	16.99	1, 21	0.001*
	R stage x density	3.81	3, 18	0.028*	1.28	3, 21	0.307	2.33	3, 24	0.100	4.90	3, 17.8	0.012*	0.10	3, 21	0.956
Suffolk,	density	5.99	4, 42.1	0.001*	8.02	4, 20.7	0.001*	0.53	4, 40	0.711	3.32	4, 42.1	0.019*	0.84	4, 50	0.505
VA 2011	R stage	19.90	1, 9.93	0.001*	1.21	1, 37.8	0.278	0.13	1, 10	0.730	18.50	1 , 9.92	0.001*	0.01	1, 50	0.922
	R stage x density	5.28	4, 42.1	0.002*	0.84	4, 20.7	0.514	1.84	4, 40	0.140	3.20	4, 42.1	0.022*	0.73	4, 50	0.574

*denotes significant treatment effect.

[a]200 seed samples taken from the five plant sample to obtain damaged seed data

[b]R stage denotes soybean development stage whole plot factor

Table 3.9. Means (±SEM) of various soybean seed quality and yield factors after exposure to zero, one, two, four, and eight *H. halys* adults and nymphs per 0.3 row m. Soybean was infested using field cages at the R4 and R6 development stages for two weeks in 2011

Suffolk, VA 2011					
Bug density per 0.3 row m	Destroyed pod percentage	Damaged seed percentage	200 seed sample weight (g)[a]	5 plant sample weight (g)	Dry soybean yield (g)
0	7.6 ± 0.9 bc	15.4 ± 1.3 b	23.5 ± 0.7 ab	55.4 ± 5.4	764.8 ± 27.8
1	6.8 ± 0.6c	15.6 ± 1.3 b	24.3 ± 0.7 a	51.9 ± 3.7	703.7 ± 36.0
2	7.6 ± 1.1 bc	19.9 ± 2.0 ab	23.4 ± 0.8 ab	56.7 ± 5.3	785.1 ± 57.6
4	10.7 ± 1.8 ab	24.3 ± 2.1 a	23.2 ± 0.5 ab	49.2 ± 4.2	785.1 ± 42.6
8	12.1 ± 1.9a	29.0 ± 2.7 a	21.9 ± 0.5 b	53.0 ± 5.3	746.2 ± 44.0
Development stage					
R4	11.5 ± 1.1 a	21.8 ± 1.6	24.5 ± 0.4 a	54.4 ± 3.3	740.1 ± 30.7
R6	6.4 ± 0.4 b	19.9 ± 1.5	22.0 ± 0.3 b	52.1 ± 2.6	736.3 ± 23.1
Beltsville, MD 2011					
Bug density per 0.3 row m	Destroyed pod percentage	Damaged seed percentage	200 seed sample weight (g)[a]	5 plant sample weight (g)	Dry soybean yield (g)
0	25.1 ± 6.4	8.4 ± 0.6 c	32.2 ± 0.6	42.2 ± 2.3	1,089.9 ± 48.5
2	23.4 ± 4.3	9.6 ± 1.3 bc	32.8 ± 0.8	50.7 ± 2.8	1,084.9 ± 41.0
4	25.1 ± 4.8	16.4 ± 2.7 ab	31.9 ± 0.7	49.8 ± 2.9	1,088.2 ± 39.8
8	33.2 ± 7.5	20.4 ± 2.5 a	31.8 ± 0.8	43.4 ± 2.7	1,023.8 ± 34.7
Development stage					
R4	14.2 ± 2.0 b	15.7 ± 2.1 a	32.4 ± 0.6	47.9 ± 1.7	1,135.7 ± 25.3 a
R6	39.2 ± 3.1 a	11.7 ± 1.4 b	31.9 ± 0.4	45.1 ± 2.4	1,007.8 ± 22.5 b

Values within the same column and treatment followed by a different letter are significantly different (Tukey-Kramer HSD, $P < 0.05$).

[a]200 seed samples taken from the five plant sample to obtain damaged seed data

Table 3.10. Means (±SEM) of the interaction between stink bug density and soybean development stage on destroyed pod percentage and seed weight from the 2011 cage studies

Suffolk, VA 2011 destroyed pod percentage		
Bug density per 0.3 row m	R4	R6
0	8.1 ± 1.6 b	7.0 ± 0.9 b
1	7.9 ± 0.4 b	5.7 ± 1.0 b
2	9.1 ± 2.0 b	6.1 ± 0.5 b
4	15.4 ± 2.3 a	6.0 ± 0.8 b
8	17.2 ± 2.1 a	6.9 ± 0.9 b
Suffolk, VA 2011 200 seed sample weight		
Bug density per 0.3 row m	R4	R6
0	25.1 ± 0.9 abc	21.9 ± 0.5 cd
1	25.8 ± 0.7 a	22.7 ± 0.7 abcd
2	25.5 ± 0.8 ab	21.2 ± 0.6 d
4	23.8 ± 0.9 abcd	21.5 ± 0.3 bcd
8	22.0 ± 0.4 d	21.9 ± 0.9 cd
Beltsville, MD 2011 destroyed pod percentage		
Bug density per 0.3 row m	R4	R6
0	8.6 ± 0.8 d	41.7 ± 3.0 ab
2	16.1 ± 4.6 cd	30.7 ± 5.4bc
4	15.7 ± 1.0 cd	34.5 ± 7.0ab
8	16.4 ± 6.4 cd	50.0 ± 5.9 a
Beltsville, MD 2011 200 seed sample weight		
Bug density per 0.3 row m	R4	R6
0	32.0 ± 1.1 ab	32.4 ± 0.5 ab
2	33.4 ± 1.3 ab	32.1 ± 1.2 ab
4	30.5 ± 0.9 ab	33.2 ± 0.2 a
8	33.6 ± 0.8 ab	30.0 ± 0.5 b

Values followed by a different letter are significantly different (Tukey-Kramer HSD, $p < 0.05$).

Chapter Four

Effects of temperature and relative humidity on the vertical distribution of stink bugs (Hemiptera: Pentatomidae) within a soybean canopy and implications for field sampling

Abstract

The influences of temperature and relative humidity on the vertical distribution of stink bugs (Hemiptera: Pentatomidae) in soybean canopies are not well understood. Stink bug position in the canopy could influence sweep net sampling accuracy. Our study examined the effects of both ambient and within-soybean canopy temperature and relative humidity on stink bug vertical distribution in two commercial, full-season soybean canopies in Virginia, one in 2010 and one in 2011. Temperature and relative humidity were continuously monitored in the upper and lower canopy. The within-canopy vertical distribution of a minimum of 20 stink bugs was documented at each of four different observation times: observations were replicated 14 times in the morning between 7:00 and 8:30 a.m., 14 around noon, 15 during the mid-afternoon between 3:00 and 4:45 p.m., and five observations were replicated in the early evening after 6:15 p.m. *Acrosternum hilare* Say was the primary species observed with 88 % of the total in 2010 and 59 % in 2011; the remainder was primarily *Euschistus servus* Say. No significant relationship was observed between the environmental parameters measured and stink bug vertical distribution in the canopy. Regardless of environmental conditions, an average of 15- 20% of stink bugs was located in the lower canopy, below the sweep net sampling zone. Results showed that sweep net sampling accuracy for stink bugs does not appear to be significantly affected by changes in temperature, relative humidity, or time of day, and sweep netting provides accurate estimates of about 80% of the total population.

Keywords: Stink bugs, soybean, temperature, relative humidity, sweep net, vertical distribution

Stink bugs (Hemiptera: Pentatomidae) are one of the most damaging insect pests of soybeans, *Glycine max* (L.) Merrill (Fabaceae) in the southern U.S. (Musser et al. 2010). Growers and crop consultants typically use a 38 cm diameter sweep net to estimate the density of stink bugs to aid in soybean pest management decisions (Turnipseed 1974; Rudd and Jensen 1977). One drawback to using the sweep net is that the sample is restricted to the uppermost portion of the soybean canopy, potentially missing insects in the lower canopy, especially in irrigated or densely planted soybeans which often attain greater plant height and yield than non-irrigated soybeans (Klocke et al. 1989; Chen and Wiatrak 2011). Furthermore, plant height can be influenced by variety as indeterminate varieties are generally taller than determinate varieties (Norberg et al. 2010). Current extension guidelines recommend samplers to pendulum-swing a 38 cm diameter sweep net crossing the tops of rows, and 'burying' the net to just below the top of the canopy. Other methods include sweeping across multiple rows and sweeping up a plant by dragging the tip of the sweep net across the foliage starting near the base of the plant and terminating near the top of the plant. There is a wide degree of variation between the specific sweep method employed by samplers, and research to date has not conclusively identified one method as being superior for all pests; some methods seem better suited for certain pests than others (Turnipseed 1974; Hillhouse and Pitre 1974). In general, sweep netting will intercept the upper canopy area, but will not be as effective in providing a good sample of stink bugs if they are located below the upper 38 cm. Environmental conditions, such as temperature, wind, and humidity can also affect an insect population's spatial distribution in a canopy and, therefore, sweep net accuracy (DeLong 1932).

However, few studies have determined how stink bugs are vertically distributed in soybean canopies, and how the microclimate within the canopy may affect their distribution. In

the southern United States, stink bug scouting in soybeans is conducted during the summer when temperatures often exceed 38 °C during the afternoon. It is unknown whether these high temperature periods cause stink bugs to move deeper into the canopy. Espino et al. (2008) found that sweep net catches of rice stink bug, *Oebalus pugnax* (F.) (Hemiptera: Pentatomidae), on rice varied by time of day, with higher stink bug abundance in one fourth of the samples taken during the morning; however, possible causes of this relationship were not examined. Rashid et al. (2006) also noted significantly fewer catches of *O. pugnax* on rice during the heat of the day on several sampling dates in July. It was suggested to avoid sampling in the afternoon on hot days, as stink bugs moved lower on the rice stems. However, Cherry and Deren (2000) did not see any significant influence of time of day or air temperature on sweep net efficiency of *Oebalus* spp. in flooded rice. They did note that it was possible that environmental conditions could influence stink bug distribution in fields that were not flooded.

The objective of this study was to determine the influence of time of day and both ambient and within-canopy microclimate temperature and relative humidity on the diurnal vertical distribution of stink bugs in soybean. Provided stink bugs move in the soybean canopy throughout the day in response to changing environmental conditions, both the accuracy of sweep net sampling and, perhaps, the efficacy attained by insecticide treatment may be affected. If this movement could be predicted, procedures could be adjusted to improve sampling results.

Materials and Methods

2010. The experiment was conducted in Virginia Beach, Virginia in a field of indeterminate NKS46-U6 soybeans that were planted on 8 May 2010. The field had a stink bug density of 4.5 per 15 sweeps at soybean development stage R5 (beginning seed; Fehr et. al 1971). Canopy height at the time of sampling was 1.2 m, and the row spacing was 0.38 m. The

canopy was full, with well-developed branching and very little defoliation. For both years, fields with a fully developed canopy were intentionally selected for this study because they are typical of non-drought stressed full season soybean grown in Virginia and throughout the region, and this canopy type is also similar to those in double crop systems during years with normal rainfall. Pods were present on all plant nodes 12 cm above the ground. On 11 August, three HOBO® Data Loggers (U10-003, Onset Computer Corporation, Bourne, MA) were suspended from a pole that was placed in the field. A single data logger was placed into the upper canopy about six cm beneath the uppermost trifoliate, another in the middle of the canopy, and the third was placed just above ground surface. They recorded temperature and relative humidity every eight minutes for the duration of each observation period.

Visual observations were made of any naturally encountered stink bug in the soybean canopy at four time periods: six observations were made in the morning between 7:00 and 8:30 a.m., four observations around noon, five in mid-afternoon between 3:00 and 4:30 p.m., and five in the early evening after 6:15 p.m., between 11 and 18 August, and 3 and 4 September. During each observation period, a minimum of 20 stink bugs (irrespective of instar or species) were located by slowly walking or crawling and kneeling between rows while gently pushing the plants back to expose stink bugs. The vertical position in the canopy was measured in cm above the ground surface. Observations were separated into two zones: the top 38 cm (the maximum depth that a sweep net will most often be passed through the canopy) and everything below. Nymphal aggregates were treated as one individual (Linker et al. 1999), so as to not bias the calculated distribution of stink bugs at each observation period.

2011. The experiment was conducted again in Virginia Beach, Virginia in a field of indeterminate Pioneer 94B73 soybean that were planted on 3 May 2011. The field had a stink

bug density of 2.5 bugs per 15 sweeps at development stage R5. Canopy height at the time of sampling was one m, and the row spacing was 0.38 m. As in 2010, the canopy was full with very little defoliation, and pods were distributed along the entire stem to about 12 cm above the ground. The number of data loggers used was reduced from three to two because in 2010, the temperature and relative humidity readings were nearly identical for the bottom and middle data loggers. The upper data logger was placed six cm below the uppermost trifoliate, and the bottom logger just above the ground. The loggers recorded temperature and relative humidity data every five minutes instead of every eight, as in 2010. Observations were not taken during the evening. There were eight observations in the morning between 7:00 and 8:20 a.m., 10 at noon, and 10 in the mid-afternoon between 3:00 and 4:45 p.m. for a total of 28 observations between July 21 and August 11. Observations of stink bug vertical location in the canopy were taken in the same manner as described in 2010.

Statistical analyses. The percentage of stink bugs found below the upper 38 cm of the canopy was calculated. Temperature and relative humidity readings were averaged for each observation time period where stink bugs were observed. The average conditions from the bottom logger were then subtracted from the upper logger to obtain the range of temperature and relative humidity for each time period. Separate linear regressions fit to the 2^{nd} order polynomial of the ambient temperature (approximated by the upper data logger), ambient relative humidity (approximated by the upper data logger), within canopy range of temperature, and within canopy relative humidity vs. the percentage of stink bugs below the upper 38 cm were performed using the SAS JMP v. 8.0 software (SAS Institute, Cary, NC). Species and life stages were pooled due to insufficient sample sizes of each species and life stage and resulting non-normality and non-linearity. Analysis of variance (ANOVA) was performed to determine if time of day had an

influence the percentage of stink bugs below the upper 38 cm using SAS JMP v. 8.0. A t-test was performed on the 2010 and 2011 mean percentage of stink bugs below the upper 38 cm using SAS v 9.2 to determine if the two years' results were different from each other due to slight differences in canopy architecture. Due to unequal variance, the Satterthwaite method was used to calculate the *t*-value and probability that the yearly mean was different (SAS Annotated Output: Proc T-Test).

Results

Stink bug species composition. Of the 551 stink bugs observed in 2010, the green stink bug, *Acrosternum hilare* Say (also *Chinavia hilaris*), comprised an overall average of 88% of the stink bugs, with the remainder identified as brown stink bugs, *Euschistus servus* Say. In 2011, 727 stink bugs were observed; 59% were *A. hilare* and 39% were *E. servus*. Nymphs comprised 46% of the total observed in 2010 and 25% in 2011. Other species identified in 2011 included *Podisus* spp., *Euschistus quadrator* Rolston and *Thyanta* spp.

Yearly mean stink bug distribution. The mean percentage of stink bugs below the top 38 cm from all observations was 20.5 ± 7.6 percent and 15.0 ± 3.9 percent for 2010 and 2011, respectively, but the yearly means were not significantly different (t = 1.35, df = 29.21, P = 0.19).

Time of day. ANOVA analyses of 2010 and 2011 data indicated no significant difference between the times of day that observations were made and the percentage of stink bugs below the top 38 cm of the soybean canopy (F = 1.00; df = 3, 15; P = 0.42; F = 0.27; df = 2, 25; P = 0.76, respectively). In 2010, the percentage below the top 38 cm was 12.8 ± 7.4 in the afternoon, 16.8 ± 7.4 in the evening, 25.5 ± 6.7 in the morning, and 27.3 ± 8.3 at noon. In 2011, the percentage below the top 38 cm was lower and more uniform (the standard error of the mean

was lower), with 12.9 ± 3.6 low in the morning, 15.2 ± 3.2 in the afternoon, and 16.5 ± 3.2 at noon.

Effects of temperature. Temperatures during the experiment ranged from 22.5 to 40.4°C and 22.3 to 44.9°C in 2010 and 2011, respectively (Tables 4.1, 4.2). In 2010, regression revealed no significant relationship between temperature range and the percentage of stink bugs below the top 38 cm of the canopy (*Percentage below* = 10.88548 – 0.2212434 (*Temperature range*) + 4.9844886 (*Temperature range* – 1.78)^2; R^2 = 0.26, *F* = 3.03, df = 19, *P* = 0.07). In 2011, no significant relationship was found between temperature range and the percentage of stink bugs below the top 38 cm of the canopy (*Percentage below* = 0.138158 + 0.0055897 (*Temperature range*) – 0.0011573 (*Temperature range* – 4.4798)^2; R^2 = 0.03, *F* = 0.35, df = 27, *P* = 0.71). Ambient temperature, as measured by the uppermost logger, also did not have a significant influence on stink bug vertical distribution in either 2010 (*Percentage below* = 21.336372 – 0.2848786 (*Ambient temperature*) + 0.2618396 (*Ambient temperature* – 30.096)^2; R^2 = 0.19, *F* = 1.99, df = 19, *P* = 0.17) or 2011 (*Percentage below* = 16.806858 + 0.0218194 (*Ambient temperature*) – 0.078254 (*Ambient temperature* – 33.8128)^2 ; R^2 = 0.09 *F* = 1.19, df = 27, *P* = 0.32).

Effects of relative humidity. Relative humidity ranged from 35.6% to 92.6% and from 39.8% to 91.7% in 2010 and 2011, respectively (Tables 4.1, 4.2). Relative humidity was highest in the morning and lowest in the afternoons. In both years, the relative humidity measured by the bottom logger was higher than that measured by the top logger. However, the relative humidity range did not have a significant influence on stink bug distribution in either 2010 (*Percentage below* = 28.153866 – 0.0877282 (*Relative humidity range*) -0.2395068 (*Relative humidity range* – 9.562)^2; R^2 = 0.11, *F* = 1.01, df = 19, *P* = 0.38) or 2011 (*Percentage below* =

14.054084 – 0.2967638 (*Relative humidity range*) – 0.0410105 (*Relative humidity range* – 11.465)^2; $R^2 = 0.11$, $F = 1.01$, df = 27, $P = 0.38$). Likewise, ambient relative humidity as measured by the uppermost logger, also had no significant effect on stink bug distribution in either 2010 (*Percentage below* = 23.333209 – 0.0222369 (*Ambient relative humidity*) – 0.0042803 (*Ambient relative humidity* – 68.89)^2; $R^2 = 0.00$, $F = 0.04$, df = 19, $P = 0.96$) or 2011 (*Percentage below* = 13.63637 + 0.0571796 (*Ambient relative humidity*) – 0.0089739 (*Ambient relative humidity*-64.1303)^2; $R^2 = 0.05$, $F = 0.60$, df = 27, $P = 0.56$).

Discussion

In this study, time of day, ambient, and within-soybean canopy temperature and relative humidity did not influence the vertical distribution of stink bugs in a predictable manner. The influences of environmental conditions on the behavior of other Hemipterans have been investigated. Romney (1945) found that sweep net catches of beet leafhoppers, *Eutettix tenellus* (Baker) (Hemiptera: Cicadellidae) in pepperwort (*Lepidium alyssoides*, Brassicaceae) varied widely with temperature, wind speed, and time of day. Population density estimates taken by weep net of *Orius insidiosus* (Say) (Hemiptera: Anthocoridae) vary among time of day, temperature, and cloud cover influences. More insects were caught in the evening and on clear, cool days (Dumas et al. 1964). Other predatory bug counts vary with both soil temperature and cloud cover (Dumas et al. 1962, 1964). Shepard et al. (1974) found that not only did *Geocoris* spp. (Hemiptera: Lygaeidae) move to lower cage positions as temperatures rose throughout the day, but that this behavior also affected sweep net catch, with greater *Geocoris* catches during the morning hours.

Temperatures during the study for both years were several degrees above average (average July high temperature for Southeast Virginia is 30.5 °C, (National Climatic Data Center

2008)). Although temperatures during the study were unusually hot for Virginia, these temperatures are more frequently encountered in many other soybean growing states south of Virginia. A previous study of the Southern green stink bug, *Nezara viridula* (L.), in Australia found that basking behavior was greatest between 7 and 9 am. After that time, bugs began to move under leaves or lower in the canopy (Waite 1980). Musser et al. (2007) found that time of day affects whole plant counts of *Lygus spp.* (Hemiptera: Miridae), in cotton, *Gossypium hirsutum*; fewer insects were observed in the terminal and flowers during the heat of the day. However, time of day did not have a significant influence on sweep catches, which suggested that bugs moved under leaves and more interior to the top of the canopy, but they were still in the 38 cm intercept region of the sweep net inside the canopy.

In this study, regardless of climatic conditions, 15-20% of stink bugs were located below the normal sweep net sampling zone (~38 cm from the top of the canopy). Russin et al. (1987) noted that stink bug feeding injury was confined to the upper halves of soybean plants until populations reached high levels of 3.8 or more per row-meter, with populations peaking at 9.5 per row meter. They speculated that overcrowding caused stink bugs to begin feeding deeper in the canopy. However, this study used determinate soybean varieties, whereas the observations in 2010 and 2011 were performed in a field with indeterminate soybean varieties. As a result, stink bugs may have been concentrated around the best food source, which was likely seeds in a certain stage of development, and stink bugs were less likely to move lower in the canopy. In an indeterminate variety, these pods may not be distributed equally in the soybean plant, unlike a determinate variety, where the seed and pod development across the plant is in similar phenological stages.

A second difference between these studies involved stink bug density. Sweep net countes indicated lower densities than were used by Russin et al. (1987). Sampling was not done continuously to determine the stink bug peak population level, which, in Russin et al. (1987)'s study, was three times higher than the mean population density.

Using current recommendations converting a density per row-m to per sweep net sample from across the Southeast, the mean density required to reach a threshold level of stink bugs corresponds to between 4.1 to 6.2 bugs per 15 sweep sample (Reisig and Roberson 2011, Roberts and McPherson 2010, Baldwin et al. 2010, Lorenz et al. 2006). Because of this wide range, there is a need to more sharply define the sweep net thresholds. The study presented here did not address this question, but provides evidence that the sweep net can be promoted as a precise sampling method even though it can only be used to sample the top of the canopy.

One factor that was not addressed by our study was the influence of canopy height on sweep net catches of stink bugs. However, there were differences in canopy height between years and a slightly smaller proportion of stink bugs were below the sweep net intercept zone in the smaller 2011 canopy. Spurgeon and Cooper (2011) found decreasing counts of *Lygus* with increasing plant canopy height. In summary, based on our results, a sweep net sampler would capture the same percentage of a stink bug population in soybeans regardless of ambient temperature, relative humidity, or time of day, and should be able to intercept most stink bugs in a canopy.

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References Cited

Baldwin, J., L. Foil, M. Grodner, A. Hammond, G. Henderson, N. Hummel, S. Johnson, R. Leonard, A. Morgan, D. K. Pollet, J. Pyzner, T. E. Reagan, D. Reed, R. N. Story, and M. Stout. 2010. Louisiana insect pest management guide. LSU AgCenter. http://www.lsuagcenter.com/NR/rdonlyres/B733BE31-7DEA-4264-93A6-AF6FED7023D3/66760/pub1838InsectPestMgmtGuide2010completebook188pgs.pdf. Accessed 6 April 2011.

Chen, G. and P. Wiatrak. 2011. Seeding rate effects on soybean height, yield, and economic return. Agron. J. 103: 1301-1307.

Cherry, R. and C. Deren. 2000. Sweep net catches of stink bugs (Hemiptera: Pentatomidae) in Florida rice fields at different times of day. J. Entomol. Sci. 35: 490-493.

DeLong, D. M. 1932. Some problems encountered in the estimation of insect populations by the sweeping method. Ann. Entomol. Soc. Am. 25: 13-17.

Dumas, B. A., W. P. Boyer, and W. H. Whitcomb. 1962. Effect of time of day on surveys of predaceous insects in field crops. Fla. Entomol. 45: 121-128.

Dumas, B. A., W. P. Boyer, and W. H. Whitcomb. 1964. Effect of various factors on surveys of predaceous insects in soybeans. J. Kans. Entomol. Soc. 37: 192-201.

Espino, L., M. O. Way, and L. T. Wilson. 2008. Determination of *Oebalus pugnax* (Hemiptera: Pentatomidae) spatial pattern in rice and development of visual sampling methods and population sampling plans. J. Econ. Entomol. 101: 216-225.

Fehr, W. R., C. E. Caviness, D. T. Burmood, and J. S. Pennington. 1971. Stage of development descriptions for soybeans, *Glycine max* (L.) Merrill. Crop Sci. 11: 929 - 931.

Hillhouse, T. L. and H. N. Pitre. 1974. Comparison of sampling techniques to obtain measurements of insect populations on soybeans. J. Econ. Entomol. 67: 411-414.

Klocke, N. L., De. E. Eisenhauer, J. E. Specht, R. W. Elmore, and G. W. Hergert. 1989. Irrigation soybeans by growth stages in Nebraska. Biological Systems Engineering: Papers and Publications, Paper 45.

Linker, H. M., J. S. Bacheler, H. D. Coble, E. J. Dunphy, S. R. Koenning, and J. W. Van Duyn. 1999. Integrated pest management soybean scouting manual. North Carolina Cooperative Extension Service pub. no. AG-385. http://ipm.ncsu.edu/soybeans/Scouting_Soybeans/soyfinal.pdf. Accessed 6 April 2011.

Lorenz, G., D. R. Johnson, G. Studebaker, C. Allen, and S. Young, III. 2006. Insect pest management in soybean. University of Arkansas Division of Agriculture Cooperative Extension Service Publication Number MP197-10M-4-00RV.

Musser, F. R., G. M. Lorenz, S. D. Stewart, and A. L. Catchot, Jr. 2010. 2010 soybean insect losses for Mississippi, Tennessee, and Arkansas. Midsouth Entomologist 4: 22-28.

Musser, F., S. Stewart, R. Bagwell, G. Lorenz, A. Catchot, E. Burris, D. Cook, J. Robbins, J. Greene, G. Studebaker, and J. Gore. 2007. Comparison of direct and indirect sampling methods for tarnished plant bug (Hemiptera: Miridae) in flowering cotton. J. Econ. Entomol. 100: 1916-1923.

National Climatic Data Center. 2008. Normal daily maximum temperature, deg F. http://lwf.ncdc.noaa.gov/oa/climate/online/ccd/maxtemp.html. Accessed 8 March 2012.

Norberg, O. S., C. C. Shock, and E. B. G. Feibert. 2010. Growing irrigated soybeans in the Pacific Northwest. Oregon State University Extension Service EM 8996.

Rashid, T., D. T. Johnson, and J. L. Bernhardt. 2006. Sampling rice stink bug (Hemiptera: Pentatomidae) in and around rice fields. Environ. Entomol. 35: 102-111.

Reisig, D. and S. Roberson 2011. Stink bug threshold. http://www.ces.ncsu.edu/plymouth/ent/soysbthreshold.html. Accessed 6 February 2012.

Roberts and McPherson. 2010. Insect management. http://www.caes.uga.edu/commodities/fieldcrops/soybeans/documents/RobertsMcPherson_InsectMgmt.pdf. Accessed 4 February 2012.

Romney, V. E. 1945. The effect of physical factors upon catch of the beet leafhopper (*Eutettix tenellus* (Bak.)) by a cylinder and two sweep-net methods. Ecol. 26: 135-147.

Rudd, W. G. and R. L. Jensen. 1977. Sweep net and ground cloth sampling for insects in soybeans. J. Econ. Entomol. 70: 301-304.

Russin, J. S., M. B. Layton, D. B. Orr, and D. J. Boethel. 1987. Within-plant distribution of, and partial compensation for, stink bug (Heteroptera: Pentatomidae) damage to soybean seeds. J. Econ. Entomol. 80: 215-220.

Shepard, M., V. H. Waddill, and S. G. Turnipseed. 1974. Dispersal of *Geocoris* spp. in soybeans. J. Ga. Entomol. Soc. 9: 120-126.

Spurgeon D. W. and W. R. Cooper. 2011. Among-sampler variation in sweep net samples of adult *Lygus hesperus* (Hemiptera: Miridae) in cotton. J. Econ. Entomol. 104: 685-692.

Turnipseed, S. G. 1974. Sampling soybean insects by various D-Vac, sweep and ground cloth methods. Fla. Entomol. 57: 217-223.

SAS Annotated Output Proc TTest. UCLA: Academic Technology Services, Statistical Consulting Group. From http://www.ats.ucla.edu/stat/sas/output/ttest.htm. Accessed 22 August 2011.

Waite, G. K. 1980. The basking behaviour of *Nezara viridula* (L.) (Pentatomidae: Hemiptera) on soybeans and its implication in control. J. Aust. Entomol. Soc. 19: 157-159.

Table 4.1. Environmental conditions and stink bug position during each observation period, 2010. Averages were taken from data logger readings, and ranges were taken by subtracting the lower sensor from the upper sensor

Date	Time of day[a]	Number of bugs observed	Temp. of upper canopy (°C)	Temp. range (°C)	Relative humidity of upper canopy	Range of relative humidity	Percentage of stink bugs in lower canopy
11 Aug.	noon	21	39.6	3.1	51.4	-14.1	5.0
12 Aug.	morn	26	26.3	0.1	92.6	0.0	3.8
13 Aug.	morn	24	27.1	1.1	90.0	-4.3	25.0
13 Aug.	aft	30	30.4	2.3	67.6	-15.7	3.3
13 Aug.	even	35	25.2	0.3	82.1	-5.0	20.0
14 Aug.	even	29	24.0	0.8	81.7	-3.2	3.4
17 Aug.	morn	25	28.3	2.3	82.7	-9.3	44.0
17 Aug.	noon	26	40.4	4.5	46.5	-15.6	61.5
17 Aug.	aft	30	37.1	3.3	53.7	-13.2	16.7
17 Aug.	even	28	29.9	0.3	76.6	-3.2	14.3
18 Aug.	morn	30	26.0	0.3	89.9	-3.8	26.7
18 Aug.	morn	22	35.3	3.9	58.9	-18.2	13.6
18 Aug.	noon	35	37.0	3.5	56.1	-16.1	42.9
18 Aug.	aft	22	29.7	0.5	81.0	-5.8	18.2
3 Sept.	aft	29	27.2	1.7	74.1	-14.8	10.3
3 Sept.	even	26	24.4	0.5	84.9	-3.9	26.9
4 Sept.	morn	25	22.5	0.3	77.0	-3.7	40.0
4 Sept.	noon	31	33.3	2.4	40.6	-17.5	0.0
4 Sept.	aft	32	34.5	3.6	35.6	-16.7	15.6
4 Sept.	even	26	23.8	0.8	54.8	-7.1	19.2

[a] Morn = morning observations (7:00-8:30 a.m.), midmorn = mid-morning (10:30 a.m.), aft = afternoon observations (3:00-4:15 p.m.), and even = early evening observations (after 6:15 p.m.)

Table 4.2. Environmental conditions and stink bug position during each observation period, 2011. Averages were taken from data logger readings, and ranges were taken by subtracting the lower sensor from the upper sensor

Date	Time of day[a]	Number of bugs observed	Temperature of upper canopy (°C)	Temp. range (°C)	Relative humidity of upper canopy	Range of relative humidity	Percentage of stink bugs in lower canopy
21 July	aft	21	42.5	6.3	51.8	-24.7	4.8
22 July	morn	21	31.1	3.6	74.8	-16.7	19.0
22 July	noon	21	40.5	2.5	54.6	-13.0	0.0
22 July	aft	21	44.9	7.3	41.6	-27.6	19.0
25 July	noon	27	38.5	5.5	56.0	-25.4	29.6
25 July	aft	24	39.2	5.4	56.3	-24.3	0.0
27 July	morn	20	25.0	0.5	92.0	-4.5	15.0
27 July	noon	21	35.8	3.4	46.3	-18.4	14.3
27 July	aft	22	33.9	2.2	55.5	-15.3	13.6
28 July	morn	21	26.8	1.3	91.7	-1.7	4.8
28 July	noon	24	36.8	3.2	60.7	-15.5	20.8
28 July	aft	21	34.4	1.0	71.3	-6.1	42.9
29 July	morn	24	26.7	0.4	85.7	-3.8	20.8
29 July	noon	22	39.2	3.9	59.0	-14.9	31.8
29 July	aft	23	37.9	3.7	60.2	-16.0	13.0
3 Aug.	morn	22	26.3	0.9	85.5	-5.0	13.6
3 Aug.	noon	21	35.3	2.9	64.5	-12.7	14.3
3 Aug.	aft	22	37.7	1.7	55.0	-7.2	18.2
4 Aug.	morn	23	25.8	1.1	86.3	-6.7	26.1
4 Aug.	noon	26	32.7	1.6	74.9	-8.5	15.4
4 Aug.	aft	28	33.7	3.2	66.8	-17.4	17.9
5 Aug.	morn	25	25.9	1.0	84.3	-6.9	4.0
5Aug.	noon	25	31.8	1.8	62.3	-10.0	16.0
10 Aug.	noon	24	38.2	0.6	43.6	0.1	4.2
10 Aug.	aft	20	33.3	0.5	52.2	-2.7	10.0
11 Aug.	morn	24	22.3	0.0	82.5	-2.1	0.0
11 Aug.	noon	27	35.5	1.8	40.5	-8.8	18.5
11 Aug.	aft	23	35.0	2.1	39.8	-5.4	13.0

[a] Morn = morning observations (7:00-8:30 a.m.), aft = afternoon observations (3:00-4:15 p.m.)

Research Summary

Using cage studies, stink bug (Hemiptera: Pentatomidae) population densities at the current threshold level of one stink bug per 0.3 row m were associated with an increase in seed damage over the course of a three week infestation period. This effect was not consistent among sites or years; such inconsistency could be due in part to good growth conditions and to soybean development during the study, especially after R6. Soybean plants at the R4 development stage were more sensitive to cage influence than R6 soybean. R6 soybean that is drought stressed is likely more prone to stink bug injury and yield loss, as evidenced by the results from the 2009 Georgetown site. The injury potential to soybean between *Euschistus servus* Say and *Acrosternum hilare* Say, or between adults and nymphs of *A. hilare* was similar. Based on these data, thresholds should remain at the one stink bug per 0.3 row m level for all combined phytophagous soybean feeding stink bugs that infest R4 stage soybean in double crop soybean systems. If soybean is being grown for grain, and weather conditions are favorable for good yield, thresholds in faster maturing double crop fields could be increased if stink bug populations reach threshold after R6.

The recently introduced *Halyomorpha hal*ys Stål is a pest in soybean. Damage to soybean from *H. halys* adults and nymphs compared to *A. hilare* adults and nymphs was not different using cage studies done at Maryland. Hence, thresholds should combine *H. halys* with the other native members of the phytophagous soybean feeding stink bug complex. Cage study results from 2010-2011 showed that *H. halys* infestations prior to pod development were not associated with increased seed damage or yield loss. However, care should be taken to manage threshold populations once pods are formed (R3-R4). R4 was the most susceptible stage to both stink bugs and cages. Infestations at R4 caused significant yield reduction compared to R2. R4-

stage soybean was slightly more sensitive to stink bug injury than R6, except at the 2011 Maryland site. Densities of four stink bugs per 0.3 row m were required to cause significant seed injury at both 2011 sites when allowed to feed for two weeks. Current threshold recommendations should apply to this stink bug species.

Based on observational studies, stink bugs in tall, fully developed canopies at the R5 development stage are primarily located in the upper 38 cm of the soybean canopy. Their distribution was not influenced by ambient temperature and relative humidity outside of the canopy or by temperature and relative humidity conditions inside the soybean canopy. Time of day did not influence stink bug vertical distribution. Based on these results, a sampler should not be concerned about temperature or relative humidity or time of day when sweeping for stink bugs. Between 15 and 20% of stink bugs are deep enough in the canopy to avoid sweep net capture.