FINAL PROJECT REPORT

Project Title: Brown marmorated stink bug control in Washington

PI: Elizabeth H. Beers
Organization: WSU-TFREC
Telephone: 509-293-8755
Email: ebeers@wsu.edu
Address 1100 N. Western Ave.
City Wenatchee
State/Zip WA 98801

Cooperators: N/A

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Notes:

Total Project Funding: $254,793

Budget History:

Item	Year 1: 2016	Year 2: 2017	Year 3: 2018	Year 4: 2010
WTFRC expenses				
Salaries	44,564	59,716	62,104	0
Benefits	9,435	14,973	15,572	0
Wages	8,042	8,364	8,699	0
Benefits	431	448	467	0
Equipment	0	0	0	0
Supplies	3,000	1,500	1,500	0
Travel	3,326	3,326	3,326	0
Plot Fees	2,000	2,000	2,000	0
Miscellaneous				
Total	70,798	90,327	93,668	0

Objectives:

- *Determine distribution of Trissolcus japonicus in Washington*. The samurai wasp, *Trissolcus japonicus*, was discovered in Vancouver, Washington in 2015. This exotic parasitoid is the most promising biological control agent for brown marmorated stink bug (BMSB). In preparation for re-distribution within Washington, a survey of its distribution in the state was necessary.
- *Maintain a laboratory culture of T. japonicus in preparation for release.* We started a laboratory culture from the original find in Vancouver and refined the rearing methods. As we determined that re-distribution would be allowable/advisable, this objective was re-directed to redistribution within the state and measuring the success of such releases.
- *Evaluate IPM-friendly management strategies for BMSB.* As it became clearer that BMSB would be established and become a pest requiring management in Washington tree fruits, we began preparing for implementation of integrated pest management (IPM) control tactics versus broad-spectrum pesticide use.
- *Document the spread of BMSB within the state.* This pest was first documented in the state in 2010 (Vancouver), and is slowly spreading into the agriculturally intensive area of eastern Washington. Tracking its spread provides an unparalleled opportunity to look at patterns and speed of invasion of an exotic species.
- *Determine suitability of native shrub-steppe plants as hosts for BMSB.* The semi-arid shrub-steppe presents a novel environment for BMSB, and its ability to successfully establish and reproduce on native flora is unknown. Proactively investigating this will help determine landscape risk of pest outbreaks.

Significant Findings

- BMSB has been detected in 27 counties in Washington State as of December 2018, with the highest numbers of reports from the urbanized areas of western Washington. All of the major fruit-growing counties have reported BMSB finds.
- *T. japonicus,* an exotic parasitoid of BMSB, was first found in Washington in 2015 in the Vancouver area; since then, all Vancouver sites surveyed have been positive for *T. japonicus*, with high levels of parasitism.
- *T. japonicus* from a laboratory colony was released at 8 sites in eastern Washington (2 sites each in Prosser, Walla Walla, White Salmon, and Yakima); it was recaptured at both Walla Walla sites and one of the Yakima sites.
- Cages made from shade net reduced the amount of stink bug damage inside the cage compared to a spray-only treatment. Codling moth numbers and damage were also reduced.
- Woolly apple aphid densities were consistently higher inside cages, likely due to exclusion of macropredators such as syrphids and lacewings; releasing insectary lacewings did not correct the problem.
- Stink bugs migrate in and out of orchards over an extended period. There is a consistent trend for barriers to reduce immigration into orchards.
- The majority of stink bugs fly into orchards between 4 and 9 ft, showing potential for exclusion using single-wall net barriers.

- There is preliminary evidence that BMSB can complete development on an assemblage of native host plants; preliminary results of gut content analysis indicate a signal persistence of at least 2 weeks, setting the stage for future experiments.

Objective 1. Determine distribution of *Trissolcus japonicus* in Washington.
The first adventive *Trissolcus japonicus* (Fig. 1) population on the west coast was discovered in Vancouver, WA in 2015. These detections prompted a more extensive survey in both eastern and western Washington. Surveys were conducted in 2016-2018 in the Vancouver area (in the same general region as the original find) and four regions in eastern Washington. The 2018 SEM survey also served as an indicator of the success of releases made in 2017 and 2018 (see Objective 2).

Egg masses for the survey were taken from a colony of BMSB maintained in small insect cages on a diet of sunflower seeds, peanuts, carrots, and potted bean plants. Egg masses were removed daily, as previous research has shown that they become less attractive to parasitoids as they age. The egg masses were cut from the leaf, leaving a small portion of leaf tissue, and were glued to card stock (Fig. 2). The card stock was labeled and transported to the survey sites the same day. The pieces of card stock were pinned to the lower surface of known BMSB host plants (deciduous trees). The masses were retrieved 3-4 days after deployment, returned to the laboratory and held at 22 °C (72 °F) until host or parasitoid emergence was complete.

2016 SEM: A total of 134 sentinel egg masses were deployed in the field from 1 June to 19 August at six sites around Vancouver, WA. *Trissolcus japonicus* was found at five out of six sites surveyed (26 egg parasitized egg masses); only Site 2 was negative. Site 6 was particularly productive, with over half of the 13 egg masses yielding *T. japonicus*. A total of 451 adult wasps were recovered from the 26 egg masses, of which 86% were females. This survey confirms the widespread presence of *T. japonicus* in the Vancouver area. The 2015 find, confined to a single site (Site 3) may have been indicative of only low levels of this parasitoid. To date, this represents one of the more evident *T. japonicus* populations in the nation.

2017 SEM: A total of 173 egg masses from the BMSB colony were deployed and monitored in 2017, or a total of 4,435 eggs. Of the egg masses deployed, 16 were attacked by *T. japonicus*; the majority (14) of positive finds were from the Vancouver area (Sites 3 and 6), which were also positive in 2016. The other positive 2 egg masses were deployed on the same card in Pioneer Park, Walla Walla, and yielded 6 males and 36 females. **This collection of *T. japonicus* constitutes the first detection of this species in eastern Washington**. The location of this find is about 7 miles from a release site in Milton-Freewater, OR, made by David Lowenstein and Nik Wiman in 2016, raising the question of whether the Walla Walla population arose from the nearby release or by natural spread with the host.

In addition to the sentinel egg masses from the colony. a single wild type BMSB egg mass was collected on 4 August on the same *Paulownia tomentosa* tree used for most of the SEM deployments at Site 6 (Vancouver). From the 26 eggs, 25 *T. japonicus* emerged (data not shown).

One encouraging aspect of these results is the relatively high rate of attack in the Vancouver sites, where *T. japonicus* appears to be established. In Site 3, 6 of 32 egg masses (19%) were parasitized, and at the Site 6, 8 of 15 egg masses (53%) were parasitized. In contrast, a single group of 2 egg masses was attacked in Pioneer Park (Walla Walla) out of the 71 deployed. Further survey efforts may reveal whether establishment is in its early phases, or *T. japonicus* is less well adapted to the climate of eastern Washington.

Other parasitoids. Two of the sites (Pioneer Park, Walla Walla and Site 3, Vancouver) yielded

Trissolcus euschisti, with a total of 10 egg masses attacked and 34 adults produced. *Anastatus reduvii* was found only at a single site (Site 3, Vancouver), with 3 egg masses attacked and 17 adults produced. While the overall number of egg masses (13) attacked by other parasitoids was similar to that attacked by *T. japonicus* (16), emergence of adults was much lower. The egg masses parasitized by *T. euschisti* averaged 13.9% successful adult emergence, and the egg masses parasitized by *A. reduvii* averaged 22.2%. In contrast, the successful rate of SEM attack by *T. japonicus* was 79.5% (17.9 – 100%) and 96.2% for the wild egg mass.

2018 SEM: A total of 232 BMSB sentinel egg masses plus two wild egg masses, were deployed/found in in 2018. *T. japonicus* was found at two locations in Walla Walla (one a repeat find from 2017), and a single location in Yakima (the site of the sole 2017 release). This shows the likely persistence of the population in Walla Walla, and the success of release efforts made in Yakima in 2017. The BMSB egg masses deployed in Vancouver yielded *T. japonicus*, bringing it to four consecutive years being detected in Vancouver. Other parasitoids detected by the survey were *T. brochymenae*, *T. euschisti*, and *Trissolcus* sp.

Objective 2. Maintain a laboratory culture of *T. japonicus* in preparation for release. Re-distribute this species in eastern Washington and perform follow up monitoring of the success of establishment.
Adult *T. japonicus* found in the Vancouver sites were returned to the laboratory to rear for release. Adults were kept in Petri dishes with honey water. When BMSB egg masses were available, a pair of *T. japonicus* was transferred to a small plastic cup containing the egg mass, and the female allowed to oviposit. After oviposition was complete, the egg mass was removed and incubated at 20 C (72 °F) for three weeks until new adults emerged. Adults held with only honey-water were quite long-lived, and the colony could be perpetuated whenever egg masses were available.

In 2017, *T. japonicus* was released at a single site (Franklin Park, Yakima) in mid-October. A total of 21 parasitized egg masses (507 eggs) on card stock were pinned to host trees. In 2018, two parasitized egg masses were placed in a small closed container with honey-water until all adults had emerged. They were transported to the selected release sites, and the lid removed to allow adults to escape (Fig. 3). Releases were repeated 4-5 times during the growing season (late June-early October). A total of 1,827 adults were released (112 males, 1,715 females) in eight sites (two sites each in Prosser, Walla Walla, White Salmon, and Yakima).

Objective 3. *Evaluate IPM-friendly management strategies for BMSB.*
We examined physical exclusion as an alternative, non-insecticidal means of suppression of BMSB. The outbreak of BMSB in the mid-Atlantic area in 2010 caused a widespread increase in broad-spectrum insecticide use, which provided only mediocre control of the target pest and flare-ups of secondary pests from disruption of biological control. While some level of insecticide use may be inevitable in the control of BMSB, all available tactics that minimize the unwanted side effects should be explored. Washington State fruit growers are making a substantial investment in overhead nets for sunburn control, and these structures provide an opportunity to enclose orchards against pests (both vertebrate and invertebrate). We used native stink bugs as a proxy for BMSB to determine the efficacy of nets, in preparation for the future when BMSB populations expand throughout the state. We also examined the micro-ecosystem changes that occur due to the use of nets as barriers to pest entry into the orchard, including the effect on other apple pests and non-target effects on natural enemies. We used two sizes of complete cages, and for orchards where building a cage is not feasible, we examined the efficacy of a single-wall net barrier as an

alternative.

Obj. 3a. Large-cage exclusion tests (2016-2017). The plots used in this experiment were built over mature trees in a 1.2-acre apple orchard at the WSU Sunrise Orchard (Fig. 4). Cages were built over mature trellised apple trees planted at 3 x 10 ft spacing. Cage frames were built from dimensional lumber supported with posts and guy wires and covered on all sides and top by white shade net (pearl leno 20% shade, Green-Tek West, Dinuba, CA). Each cage was 40 x 50 ft and enclosed 4 rows x 12 trees (total 48 trees/cage). Each row was a different apple cultivar (Jonagold, Gala, Granny Smith, and Golden Delicious).

Three treatments were tested: 1) cages made from shade netting (plus supplemental sprays), 2) uncaged, conventional management (routine airblast sprays), and 3) an uncaged, unsprayed check. Treatment 2 received routine sprays for codling moth, but none specific to secondary pests (mites, aphids). All plots received routine applications of herbicides, fungicides and fertilizer. With the exception of stink bugs, all pest and beneficial populations were naturally occurring in the block. Because the orchard had no history of stink bug damage, artificial pressure was created by collecting consperse stink bug, *Euschistus conspersus*, and releasing them in the block. The ability of pests and natural enemies to penetrate the cage barrier was measured with visual observations (timed counts), traps baited with pheromones or kairomones, or behavioral traps (yellow sticky cards, earwig shelters). Counts were made every 2 weeks throughout the season, and a single index of the seasonal counts for each insect was calculated (cumulative insect days, or CID). This index is the average of two successive counts multiplied by the number of intervening days and summed over the season. The CID were analyzed using analysis of variance (SAS 2017, PROC GLIMMIX). Fruit damage was sampled in mid-summer and again just prior to harvest and analyzed using logistic regression with a binomial distribution (PROC GLIMMIX).

Very few stink bugs were recaptured in pheromone traps, and stink bug fruit damage was correspondingly low in both years (Fig. 5); however, the damage was lowest in the cage treatment in both 2016 and 2017. Other direct pests such as codling moth and leafrollers (data not shown), were also excluded to a marked degree by the cages. Woolly apple aphid reached outbreak levels inside the cages (Fig. 6A) but were present in very low numbers outside the cages. The specialist parasitoid of woolly apple aphid, *Aphelinus mali*, was similarly high inside the cages (Fig. 6B), likely in response to the high aphid populations. It is probable that the cage netting is permeable to this tiny wasp, but also possible that the parasitized aphids present when the trees were caged simply continue to reproduce inside the cages. Despite the high numbers of *A. mali*, the populations of woolly apple aphid were not sufficiently suppressed. Unlike the parasitoid, the macropredators of woolly apple aphid (lacewings and syrphids) were much lower inside the cages, indicating the winged adults were prevented from entering the cage. Earwigs, another woolly apple aphid predator, tended to be lower inside the cages (2016), but this effect was less pronounced than with the winged predators.

Obj. 3b. Small-cage exclusion tests. The experimental design of the small cage (Fig. 7) experiment was similar to the large one, except that the plots were three 'Golden Delicious' trees (single row), and the cages were 10 x 10 x 5 ft. The same treatments were used, but each had 10 replicates in a randomized complete block design. All cages had a pheromone trap for the three tortricids, but the other two treatments were sampled with 2 traps/ species placed in buffer rows to avoid inter-trap competition. Sampling and analysis were done as in the large cage experiment, except that stink bug releases were not made (on the assumption that the large cages represented a more realistic commercial scale).

Woolly apple aphid densities were 100- to 400-fold higher inside the cages than in the airblast and check treatments, respectively. Spider mites were significantly higher inside the cages (2016 only); 95% of the mites found were brown mite. Earwigs were not significantly different between the treatments. Lacewing and syrphid adults were effectively excluded by the cages. Codling moth pheromone trap captures were greatly reduced inside the cages (1.8-15 moths/trap) vs outside (123-240 moths/trap). Likewise, fruit damage by codling moth was 2.0-8.4% inside the cages, vs. 15.9-19.4% (airblast) and 58.2-59.8% (untreated). Sunburn was significantly reduced inside the cages; however, as in the large cages, the airblast treatment also reduced sunburn relative to the check.

In 2018, we tested augmentative release of a woolly apple aphid predator to determine if we could correct the macropredator deficiency inside the cages. We purchased green lacewing, *Chrysoperla rufilabris* (Fig. 8) from Rincon-Vitova Insectaries (Ventura, CA) in the adult stage. The adult of this species is not predacious, thus only eggs and resulting larvae were assessed. We divided the 10 cages into two groups of five each, counted the woolly apple aphid colonies, and used that as a blocking factor to assign treatments (1 – Lacewings released; 2 – check, no release). Ten adults were placed into cups and released into the small cages on 31 May. Lacewing eggs and woolly apple aphid densities were counted weekly for 3 weeks. A second release at a higher predator density was performed on 25 July, using 100 adults/cage. After the first (low density) release, no lacewing eggs were found in the release or check cages. After the second (high density) release, 3.6 eggs/cage were found in the release cages. Woolly apple aphid densities did not differ between the two treatments, indicating the predator release had no measurable effect.

Obj. 3c. Physical exclusion, single-wall barriers. For the single-wall exclusion study, three commercial apple orchards in the Manson, WA area with a history of stink bug damage were used. We constructed net barriers in 2016 and assessed the results in 2016-2018. The barriers (150 ft long x 15 ft high) were made of commercial shade netting (20% pearl leno [white] net; Green-Tek West, Dinuba, CA) fastened to 16 ft posts at the edge of the sagebrush/native vegetation that bordered the orchards. The nets had 6-inch flaps of net sewn at three heights facing the native vegetation. In 2018, approx. one-half the length of the three flaps (75 ft) was retro-fitted with deltamethrin-infused netting (ZeroFly netting, Vestergaard-Frandsen, Washington, DC) (Fig. 9). This resulted in three treatments replicated at each site: 1) net barrier with deltamethrin flaps; 2) net barrier with shade net flaps; and 3) a no-barrier check.

Stink bug populations were assessed with a beating tray in the natural vegetation and in the orchard throughout the field season. Fruit damage was determined through visual inspection in late August/early September. Stink bug damage levels were extremely low with none of the treatments reaching levels higher than 0.2%. Beat tray samples of the surrounding vegetation behind both net treatments resulted in substantially higher counts than samples in the check, and there was no significant difference in the amount of stink bugs found in the orchard between the three treatments (Fig. 10). While the orchard counts were not significantly different, the netted plots received a much higher pressure of stink bugs migrating from the natural vegetation. There is a greater difference in the amount of stink bugs in the orchard compared to the vegetation in the netted plots (>80% reduction) than in the control plot (40% reduction), indicating the barrier prevented most of the stink bugs from reaching the orchard.

A complimentary study was conducted from June - September to determine the height at which stink bugs migrate into orchards. A sticky barrier (13 ft high x 6 ft wide; Fig. 11) was constructed using dimensional lumber and clear sticky panels, 1 x 6 ft (Alpha Scents, West Linn OR), at the orchard boarder in five locations in Manson. Stink bugs on the sticky panels were removed and their height of interception was recorded every week. A single index of the seasonal counts for adult counts was calculated (CID) as the average of two successive counts multiplied by the number of intervening days and summed over the season, and a cumulative trap-day CTD index was used for the sticky panel traps. All data were analyzed using a generalized linear mixed model (PROC GLIMMIX, SAS 2018).

Height of immigration/emigration. An additional question for the single-wall barrier concept is how tall the barriers needed to be to intercept stink bugs. Observations in the mid-Atlantic states indicated that barriers were more effective if they were interposed between the orchard and a field crop (corn, ca 6 ft high) versus a deciduous woodlot (ca. 40 ft high). Vegetation bordering eastern Washington orchards (with the exception of riparian zones) is composed of shrubs, forbs, and small trees, with heights ranging from 1-10 ft. We hypothesized that height of immigration would be related to the height of the surrounding donor vegetation. To test this, we built wooden frames onto which double-sided sticky panels (AlphaScents, West Linn, OR) were attached at 8 (2017) or 11 (2018) heights.

In 2017, there were no statistical differences in stink bug capture among heights over 2 ft; very few stink bugs were caught below this level. The maximum height in 2017 was 8 ft, and it was apparent that much of the immigration was at that height (or higher), so the maximum height was increased 12.5 ft in 2018. In the latter test, there were no statistical differences in interception height for the sticky barriers for stink bugs moving *out* of the orchard (data not shown). However, there was a significant difference in height of movement *into* the orchard (from of the surrounding vegetation). The highest counts were at 4.5 ft (7.20 stink bugs) and the lowest at 12.5 ft (1.20 stink bugs), with the majority captured between 4 and 9 ft (Fig. 12). Similarly, a significantly higher number of stink bugs were caught in the middle flap (6 ft) of the single-wall barriers than in the lower (1 ft) or upper (9 ft) flaps, a further indication that the majority of stink bugs migrate into the orchard ~6 ft.

Obj. 3 Conclusions. The preliminary information from both the large and small cage experiments indicates there is potential for a substantial degree of exclusion of codling moth. The wild moth pressure in the large cage research blocks was considerable, and late season pheromone trap captures indicate a high degree of success in excluding adults. The small cages trials show that shade netting can be an effective barrier to external moth populations Interestingly, a larger proportion of moths were able to escape the cages than enter them.

Sampling stink bugs captured on the net barriers, in the orchard, and in the surrounding vegetation in the Manson trials provided insight into stink bug seasonal migration habits. Stink bugs densities were higher in the surrounding vegetation for both net treatments than the check, yet numerically fewer stink bugs were found in the orchard behind the net treatments than the check. This indicates that the net barrier may be preventing a large portion of the migrating stink bugs from reaching the orchard. The sticky barrier trial further confirms that the majority of adult stink bugs fly into orchards below 12 ft, which implies a barrier may only need to be 12 ft to provide successful control.

Objective 4. Document the spread of BMSB within the state.
As BMSB continues to spread, efforts in 2018 focused on sampling in location gaps between

known populations and in new cities within important fruit growing regions. We used direct surveys (beating trays, pheromone traps) to determine the presence and relative abundance of BMSB in the state. We also solicited input from homeowners and Master Gardeners. Verified finds were recorded in a database available to BMSB researchers, and the results available in map form (http://tfrec.cahnrs.wsu.edu/beers-tfentomology/bmsb/bmsb-wa/).

Obj. 4. Results and Discussion. A total of 27 counties have reported detections of BMSB (Fig. 13). The highest numbers of BMSB reports came from Clark, King, Pierce, Snohomish, and Thurston counties in 2018 likely reflecting the large amount of vehicular traffic that could help spread this invasive species. Conversely, Chelan county has a (relatively) smaller human population and vehicular traffic, but a keen degree of awareness and interest in this species. Trapping studies and citizen reports discovered the first BMSB in both Okanogan county, and the city of Chelan in early October of this year. This is a major concern, as those are both major fruit growing areas. In general, it appears that the arid climate of eastern Washington will not effectively limit the establishment of BMSB, given the growing number of reports in this part of the state, and establishment to the north in the Okanagan Valley of British Columbia.

Objective 5. Determine suitability of native shrub-steppe plants as hosts for BMSB
BMSB has usually been associated with humid temperate environments such as the deciduous hardwood forests in northeast Asia, the Mid-Atlantic States in the American Northeast, and the Pacific Northwest west of the Cascades. However, BMSB has shown itself to be remarkably adaptable. It rapidly colonized many areas of Washington State, including population centers in the arid Columbia Plateau. As its range continues to expand and its populations grow, there is legitimate concern that this highly mobile landscape-level pest will be able to build up in the shrub-steppe habitats that border Washington's tree fruit orchards. Knowledge of the landscape ecology of BMSB in the arid PNW is critical for effective scouting, risk assessment, and area-wide IPM efforts.

Obj. 5a. Laboratory feeding studies. In a preliminary experiment, we noted that BMSB could develop from egg to adult on an assemblage of plants native to eastern Washington (Fig. 14). In 2018, we evaluated more fully the relative suitability of two common shrub-steppe plants for BMSB feeding and oviposition in comparison to Lima bean seedlings (a standard BMSB colony diet), which served as the check. Cuttings of sagebrush and bitterbrush (or bean seedling in potting soil) were placed in acetate cages over 6-in pots. Using a randomized complete block design (3 treatments, 10 reps), we placed a male-female BMSB pair in each arena. Insects were weighed at Day 0, 7, 15, and 22 to monitor weight loss or gain as a metric of host plant suitability. At the same intervals, adult mortality was assessed, eggs were counted, and plant material was replaced. Due to high mortality in our check treatment, no significant differences were observed in any fitness parameters measured. This mortality is likely due to insufficient ventilation in the arenas and the stress experienced by the insects during handling from the frequent weight measurements. In addition, future studies will use an assemblage of native plants, which is consistent with its known near-obligate polyphagy and the conditions it will likely experience in the field.

Obj. 5b. Gut content analysis. Gut content analysis has been used extensively in biological control studies to determine which prey are being used by predators, by screening gut contents with various prey primers. Screening with primers asks the question '*Did you eat species X*?', with the understanding the species screened would likely be pests of interest. This concept may be extended to examining the feeding habits of highly mobile and polyphagous pests (such as

BMSB) to better understand their ecology at the landscape level.

Recent developments in "deep sequencing" technology for genetic analysis enables detailed, plant species-level feeding histories of individual insects to be determined by extracting and sequencing plant genetic material from their gut. This method effectively asks the question '*What did you eat*?', and as such, is a more comprehensive look at feeding history. This approach has been successfully used on various psylla species by Dr Rodney Cooper (USDA-ARS, Wapato, WA) who demonstrated its use for the first time for Hemiptera. We are refining the technique for BMSB in collaboration with Dr. Cooper.

The first step in assessing this methodology is to determine signal persistence, or how long plant DNA is detectable in the gut after feeding has ceased. We used two known hosts of BMSB, Lima bean plants and carrots. We allowed BMSB to feed on Lima bean for 7 days, then switched them to carrots. Insects were killed and their gut dissected (Fig. 15) at 5 time points (0, 1, 3, 7, and 14 days) post-host switch. DNA was extracted using kits (DNeasy, Qiagen, Hilden, Germany) and amplified with two universal plant primers (*trnF* and *ITS*). Amplified plant DNA was quantified with a NanoDrop and submitted for deep sequencing on the PacBio platform at WSU's Core Facility. Each sample included a barcoded primer to identify it post-sequencing. Sequence data from the Core Facility was sorted and compared to known plant sequences (GenBank) using the Geneious Prime software.

The *trnF* primer yielded satisfactory results; however, the *ITS* primer failed to yield sufficient sequences for host plant identifications. The original host plant (Lima bean) was recovered from BMSB guts at all time points post switch, indicating a signal persistence of at least 2 weeks. The sequence composition remained predominantly that of Lima bean at 0 days (pre-switch) and shifted to carrot at all subsequent time points. However, Lima bean DNA was still detected at all post-switch time points, but in decreasing amounts. Sequences identified as plants not included in this study and/or not present locally suggest that this method is sensitive to contamination and that ground truthing is necessary to confirm the presence of identified putative hosts at the site of insect collection. A third universal primer, *trnL*, will be tested in the future to verify the results of *trnF*.

Executive Summary

Project Title: Brown marmorated stink bug control in Washington

Keywords: invasive pest, biological control, parasitoid, host plant suitability

Abstract: Brown marmorated stink bug (BMSB), an invasive pentatomid from Asia, continues to expand its range in Washington State. Its ability to use native host plants may be a factor in its success in the arid interior. An exotic parasitoid, *Trissolcus japonicus*, was found in Vancouver and may help regulate populations.

BMSB has continued to expand it range in Washington State since it was first found in 2010. Homeowner complaints (and likely densities) are higher in the populous urban/suburban areas of western Washington, but there has been substantial incursion into the arid areas east of the Cascades. As these populations continue to spread and grow, they threaten the intensive agricultural production in this region, with tree fruits being among the most vulnerable crops. If broad-spectrum pesticides are the only remedy to prevent damage, we can expect widespread disruption of integrated pest management (IPM) programs resulting in the reduction of natural enemies and outbreaks of secondary pests. To mitigate this outcome, we need to explore non-insecticidal tactics and gain a better understanding of landscape ecology as it relates to risk.

Initial assessments of native natural enemy impact were disappointing, with generally very low levels of predation and parasitism. The native species that attacked native stink bugs were apparently poorly adapted to use BMSB as a host and may have played a role in outbreaks in the eastern US. The discovery of adventive populations of an Asian parasitoid, *Trissolcus japonicus*, provided an opportunity to exploit biological control more fully. One such population was found in Vancouver, WA in 2015, which allowed the re-distribution within the state's borders (inter-state movement is regulated). These efforts were initiated, along with monitoring establishment in the release sites. Due diligence also demands that the non-target effects of this exotic parasitoid on native stink bugs be assessed; to date, there appears to be a small to moderate impact on native pest stink bugs, as they are poor hosts for *T. japonicus*. While not species-specific, *T. japonicus* has promise to moderate BMSB densities, especially in unmanaged habitats.

While biological control will always be helpful, it is likely that direct protection of orchard crops will be necessary at some point. To this end, we have begun exploring two promising tactics: physical exclusion and attract-and-kill. Our physical exclusion experiments have taken two forms: a full orchard enclosure made of shade netting, and a single-wall barrier between the native vegetation harboring stink bugs and the orchard, where topography, orchard architecture, or expense does not permit full enclosure. Both approaches have shown some promise of reducing pest pressure, although to a higher degree with full enclosure. Full enclosure has multiple benefits, including sunburn control, shading orchard workers, and minimizing multiple vertebrate and invertebrate pests. However, they are not without non-target effects, as they appear to exclude natural enemies and promote certain secondary pests. Similarly, single-wall barriers, especially those that incorporate insecticide-infused netting (IIN) may kill some beneficial insects along with the target stink bug pests.

Lastly, understanding the landscape ecology of BMSB in the shrub-steppe will help inform risk analyses for fruit damage. It has been speculated that arid growing regions will not support

BMSB, but its rapid establishment in eastern Washington belies this. BMSB appears to be sufficiently adaptable to a wide range of suboptimal environmental conditions, and we have preliminary evidence that it can complete development on (at least) the same host plants that support native stink bugs. Its ability to expand and pose a threat beyond these habitats remains to be determined.