

Effects of Extreme Low Winter Temperatures on the Overwintering Survival of the Introduced Larval Parasitoids *Spathius galinae* and *Tetrastichus planipennisi*: Implications for Biological Control of Emerald Ash Borer in North America

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Abstract

Climate change has been linked to shifts in the distribution and phenology of species although little is known about the potential effects that extreme low winter temperatures may have on insect host–parasitoid interactions. In late January 2019, northern regions of the United States experienced a severe cold wave caused by a weakened jet stream, destabilizing the Arctic polar vortex. Approximately 3 mo later at six study sites in southern Michigan and three in southern Connecticut, we sampled the overwintering larvae of the emerald ash borer, *Agrilus planipennis* Fairmaire (Coleoptera: Buprestidae), and two larval parasitoids, *Spathius galinae* (Hymenoptera: Braconidae) and *Tetrastichus planipennisi* (Hymenoptera: Eulophidae), that are being introduced as emerald ash borer biocontrol agents in North America. At these nine study sites, emerald ash borer-infested ash trees and/or saplings were debarked and each overwintering emerald ash borer and parasitoid larva was then examined for cold-induced mortality, as indicated by a brown coloration, flaccid, and watery consistency. In early spring in Michigan, we found 4.5–26% of emerald ash borer larvae, 18–50% of *S. galinae* larvae, and 8–35% of *T. planipennisi* larvae were killed by cold. In Connecticut where temperatures were more moderate than in Michigan during the 2019 cold wave, <2% of the larval hosts and parasitoids died from cold injury. Our findings revealed that cold-induced mortality of overwintering larvae of emerald ash borer and its larval parasitoids varied by location and species, with higher mortality of parasitoid larvae in most Michigan sites compared to host larvae. The potential impacts of our findings on the management of emerald ash borer using biocontrol are discussed.

Key words: climate change, extreme cold weather, overwintering, invasive, biological control

Insects are poikilothermic organisms that are unable to regulate their body temperature, and therefore, their development and survival are profoundly affected by changes in the temperature of their habitats. Although many insects have evolved effective mechanisms to deal with normal seasonal temperature changes (see review in Tauber et al. 1986), unusual or abnormal seasonal temperatures (relative to the normal range of seasonal changes in temperature) can affect the population dynamics of both insects and their natural enemies (Bale et al. 2002, Hance et al. 2007, Klapwijk et al. 2012). Thus, understanding the population impacts of extreme seasonal weather

events on insect natural enemies, as well as their insect hosts, can be important for the development and implementation of effective biological control programs designed to protect both natural and agricultural ecosystems (Baffoe et al. 2012).

The emerald ash borer, *Agrilus planipennis* Fairmaire (Coleoptera: Buprestidae), is a serious invasive pest of ash (*Fraxinus* spp.) trees in North America that was likely transported to North America in the 1990s as larvae or pupae inside wooden-packaging materials from China (Haack et al. 2002, Bray et al. 2011, Herms and McCullough 2014, Canadian Food Inspection Agency 2019, Emerald Ash Borer

Information 2019). Natural enemies of emerald ash borer (one egg parasitoid and three larval parasitoids) native to Northeast Asia have been introduced into North America for biocontrol of this borer (Federal Register 2007, 2015; Bauer et al. 2015; Duan et al. 2018). Since the emerald ash borer biocontrol program began in 2007, several biocontrol agents such as the larval parasitoids—*Tetrastichus planipennisi* Yang (Hymenoptera: Eulophidae) and *Spathius galinae* Belokobylskij (Hymenoptera: Braconidae)—have successfully established and are spreading and helping to conserve species of ash in Michigan and Connecticut (Duan et al. 2013, 2015, 2017, 2018, 2019; Abell et al. 2014; Bauer et al. 2015; Kashian et al. 2018; J. J. Duan, L. S. Bauer, and R. Van Driesche, unpublished data). In both North America and Asia, emerald ash borer overwinter as larvae ranging from first to mature fourth instars, whereas its larval parasitoids overwinter as mature last-instar larvae inside the feeding galleries of parasitized host larvae between the bark and sapwood (Bauer et al. 2015).

Recent studies demonstrate that *S. galinae* is important in the protection of older, larger ash trees, whereas *T. planipennisi* is important in the protection of younger, smaller ash trees and saplings, due to differences in parasitoid ovipositor length in relation to bark thickness (Abell et al. 2012; Murphy et al. 2017; Duan et al. 2017, 2018, 2019). Determination of the overwintering survival of *S. galinae* and *T. planipennisi* in various regions of the United States may help predict the success rate of their establishment and their potential for the biological control of emerald ash borer as part of the conservation of North American ash trees.

From 21 to 31 January of 2019, the Midwestern states, including the entire state of Michigan, experienced a severe cold wave caused by instability of the jet stream around the Arctic polar vortex (Vaughn et al. 2017), which brought the coldest temperatures in over 20 yr to most locations in the affected region, including some all-time record lows (National Oceanic and Atmospheric Administration 2019). For example, on 31 January 2019, the city of Flint in southern Michigan recorded a low temperature of -26°C (-14°F), breaking the record for that date of -22°C (-8°F) set in 1963. In contrast, states in southern New England, including the state of Connecticut, experienced more typical cold temperatures during this period. Because of this variation in the severity of the 2019 cold wave, we hypothesized that larval mortality in spring 2019 would be higher in Michigan than in Connecticut for both *S. galinae* and *T. planipennisi*, the two larval parasitoid species being introduced as emerald ash borer biocontrol agents north of the 40th parallel in North America.

To test this hypothesis, we sampled the overwintering larval stages of emerald ash borer and the introduced larval parasitoids *S. galinae* and *T. planipennisi* the following spring (approximately 3 mo after the 2019 cold wave) from emerald ash borer-infested ash at long-term emerald ash borer biocontrol study sites in southern Michigan and central Connecticut, both areas where *S. galinae* and *T. planipennisi* are well established (Duan et al. 2013, 2018, 2019; MapBiocontrol 2019). The larval mortality observed for emerald ash borer, *S. galinae*, and *T. planipennisi* sampled from infested ash in early spring 2019 at the Michigan and Connecticut study sites were then summarized and discussed in relation to the temperature data recorded from January to May 2019 at nearby weather stations.

Materials and Methods

Study Sites

Our long-term emerald ash borer biocontrol study sites in Michigan included six, secondary mixed hardwood forests in three southern counties: Ingham (three sites), Gratiot (two sites), and Shiawassee

(one site). The sites in Ingham County consisted of two adjacent Meridian Township parks—Central and Nancy Moore Parks (site CP), Legg and Harris Nature Center Parks (site LP), and one county park—William M. Burchfield Park (site BF). The sites in Gratiot County were the Gratiot-Saginaw State Game Area (site GSW) and the Maple River State Game Area (site MRE), while the remaining site in Shiawassee County was Rose Lake Wildlife Area (site RL). For more detailed information and background on these sites, see Duan et al. (2013). *Tetrastichus planipennisi*, first approved in 2007 for introduction in the United States as an emerald ash borer biocontrol agent, was released from 2007 to 2010 at these Michigan study sites (MapBiocontrol 2019), and established, self-sustaining, and expanding populations were documented during the subsequent decade (Duan et al. 2013, 2015, 2017, 2018). *Spathius galinae*, a third emerald ash borer larval parasitoid approved for introduction in the United States in 2015, was released from 2015 to 2017 at these sites (MapBiocontrol 2019), and by fall 2017, we observed moderate levels of emerald ash borer larval parasitism ($\sim 30\%$) by *S. galinae* at all the sites (J. J. Duan, L. S. Bauer, and R. Van Driesche, unpublished data).

Our long-term emerald ash borer biocontrol study sites in Connecticut include three secondary mixed hardwood forests in two counties: two adjacent state forests in Litchfield County (sites CT1 and CT2) and one in the Cromwell Wildlife Management Area in Middlesex County (site CT3). The GPS coordinates and other habitat characteristics of these three sites are described in Duan et al. (2019). At these sites, *T. planipennisi* was released from 2015 to 2016, *S. galinae* in 2016 and 2017, and both species are confirmed to have established self-sustaining populations at the three sites (Duan et al. 2019).

Sampling Procedures for Parasitism of Emerald Ash Borer Larvae

In Michigan, from late April to early May of 2019 (approximately 3 mo after the late January cold wave), we haphazardly selected and debarked eight pole-sized ash trees (diameter at breast height [DBH] ranging from 8 to 21 cm) and 20 ash saplings (DBH from 2.5 to 5.8 cm) infested with emerald ash borer at each of the six study sites. The results of our earlier research on emerald ash borer biocontrol at these long-term study sites revealed that *T. planipennisi* was more frequently recovered from ash saplings and small ash trees (DBH < 8 cm), whereas *S. galinae* was more frequently recovered in larger (DBH > 8 cm) ash trees (Duan et al. 2017; J. J. Duan and L. S. Bauer, unpublished data). Due to the limited availability of ash saplings at our Connecticut sites, we sampled larval emerald ash borer and parasitoids from 3 to 5 pole-size ash trees (DBH 9–21 cm) at each of the three study sites but did not sample ash saplings. All the ash trees (in both Michigan and Connecticut) or saplings (Michigan only) that were selected for sampling were alive and showed signs and symptoms of recent emerald ash borer infestation (e.g., fresh woodpecker feeding, epicormic shoots, bark splits, and/or D-shaped emerald ash borer exit holes). The sampled ash trees or saplings were also within 0.5–12 km from the parasitoid release plots at each study site.

To observe emerald ash borer parasitism by *S. galinae* and/or *T. planipennisi*, we debarked the entire trunk and all branches that were >2.5 cm in diameter for each selected tree with a drawknife to expose the emerald ash borer larval galleries. Outer and inner bark tissues, as well as layers of outer sapwood as needed, were carefully removed to examine each emerald ash borer larval gallery or pupation chamber for the presence of emerald ash borer larvae and/or parasitoids. Parasitism by *S. galinae* was identified in the field

based on presence of broods of parasitoid larvae, pupae, cocoons, and/or pharate adults, and subsequently in the laboratory, by identification of adults that emerged from individuals reared in the laboratory (Duan et al. 2019). Parasitism of emerald ash borer larvae by *T. planipennisi* was scored in the field based on presence of broods of visible parasitoid larvae, pupae, and/or pharate adults in emerald ash borer galleries (Duan et al. 2013).

For each emerald ash borer larva parasitized by *S. galinae* or *T. planipennisi*, the total number, lifestage, and fate of overwintering parasitoid individuals in each brood were recorded. Each parasitoid larva, pupa, or pharate adult was scored as healthy (whitish in color), dead due to cold injury (brown coloration and watery consistency, Fig. 1A and B) or other unknown factors (e.g., dried up or enveloped by host plant callus tissue). In addition to the status of *S. galinae* and *T. planipennisi* lifestages, we also scored overwintering mortality of the unparasitized emerald ash borer larvae ($\leq 40\%$ were first- to second-instar larvae, whereas the remaining larvae ranged in age from third instars to J-shaped mature fourth-instar larvae) caused by cold using the same criteria as for the parasitoids (Fig. 1C). Mortality of the observed emerald ash borer larvae due to woodpecker predation, North American native parasitoids (primarily native *Atanycolus* species complex [Braconidae] and *Phasgonophora sulcata* Westwood [Chalcididae]), and other undetermined factors such as host tree resistance (emerald ash borer cadaver encapsulated by host tree callus) and pathogens (cadaver appeared dry, liquefied, or sporulated) were also recorded (Liu and Bauer 2006; Duan et al. 2015, 2017), but those data were excluded from analyses in this study.

Temperature Data

Minimum and mean daily temperatures from January to May in 2019 and in the previous 10 yr (2009–2018) were obtained from weather stations closest to our study sites in Michigan and Connecticut (National Oceanic and Atmospheric Administration 2019).

We used temperature data from weather stations at the Capital Region International Airport in Lansing, MI (15–50 km from our Michigan study sites) and the Meriden-Markham Municipal Airport in Meriden, CT (16–22 km from our Connecticut study sites).

Data Analysis

Mean overwintering mortality rates (\pm SE) caused by cold injury for the emerald ash borer larval parasitoid *S. galinae* or *T. planipennisi* at each study site were calculated from the proportion of the parasitoid larvae scored as killed by cold in each observed brood of the parasitoid larvae. Likelihood ratio chi-square tests based on the nominal logistic regression model were used for detection of effects of the study sites and ash-tree size class (pole-size trees vs saplings) on the overwintering mortality rates of *S. galinae* and *T. planipennisi* in Michigan. Similar likelihood ratio chi-square tests were used in analyzing the effect of study sites on the overwintering mortality of *S. galinae* from the three Connecticut study sites. In addition, we also calculated the overwintering mortality rate of emerald ash borer larvae from each site as the proportion of emerald ash borer larvae killed by cold relative to the total number of emerald ash borer larvae (healthy individuals + those killed by cold). Minimum daily temperatures recorded at the Michigan and Connecticut weather stations from January to May 2019 and for the previous 10 yr (2010–2018) were compared by two-sample *t*-tests using R-statistical program (R Core Team 2018). All statistical analyses except the two-sample *t*-tests were carried out with JMP 14.0.0 statistical software (Sall et al. 2017).

Results

Mortality of Overwintering Parasitoids and Emerald Ash Borer Larvae in Michigan

Throughout the sampling period at the six study sites in Michigan, a total of 1,102 overwintering *S. galinae* larvae in 217 broods were

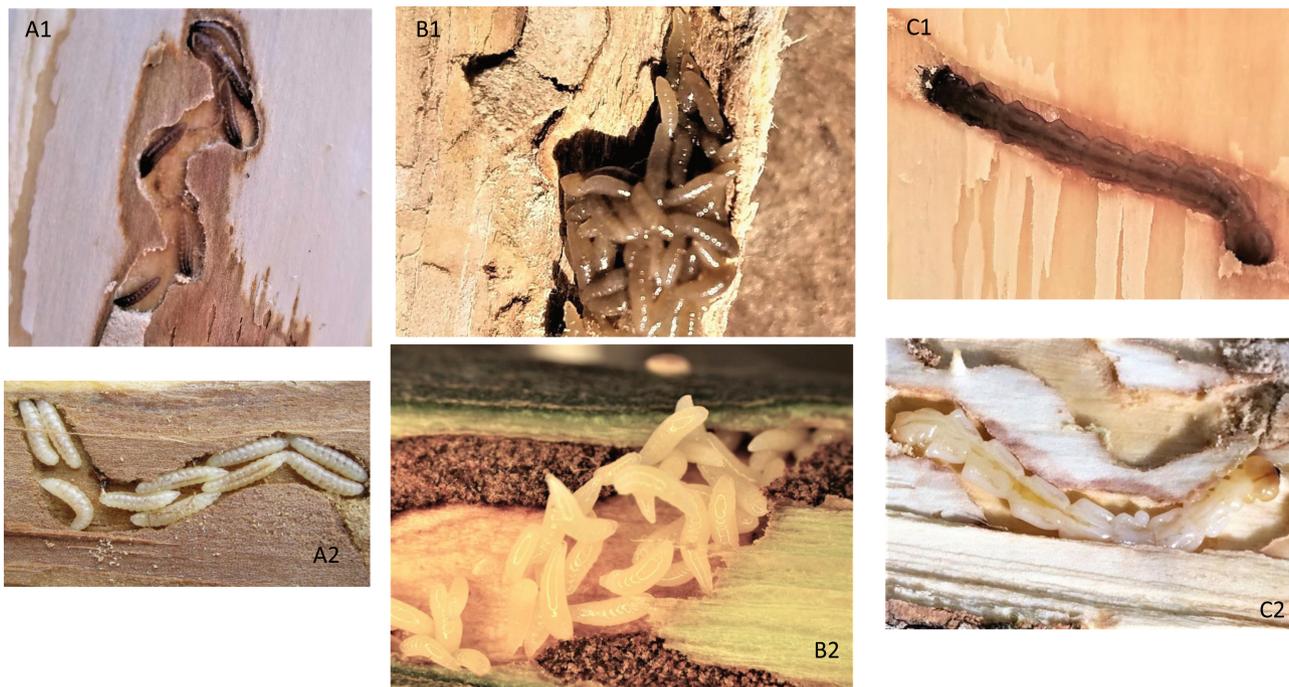


Fig. 1. Dead, brown, and watery overwintering larvae of emerald ash borer and two species of larval parasitoid killed by extreme temperatures versus healthy larvae for each species—*S. galinae* larvae (A): cold-killed (A1) versus healthy (A2); *T. planipennisi* larvae (B): cold-killed (B1) versus healthy (B2); and emerald ash borer larvae (C): coldkilled (C1), versus healthy (C2).

observed: 196 broods in pole-size trees (DBH = 8–21 cm) and 21 broods in saplings (DBH = 2.5–5.8 cm). The mean mortality per brood of the observed *S. galinae* larvae due to cold from all the broods ranged from approximately 18% to 50% across study sites (Fig. 2A). While tree-size category (ash sapling vs pole size) did not have a significant effect on overwintering *S. galinae* larval cold mortality (likelihood ratio chi-square test: $\chi^2 = 0.02918$, $df = 1$, $P = 0.863$), sample site had a highly significant effect (likelihood ratio chi-square test: $\chi^2 = 55.61$, $df = 5$, $P < 0.0001$).

At the same six Michigan study sites, a total of 1,946 *T. planipennisi* larvae in 162 broods were observed, with 30 broods from pole-size trees and 132 broods from saplings. The mean mortality per brood of the observed *T. planipennisi* larvae that was due to cold injury ranged from approximately 8% to 35% across study sites (Fig. 2B). As observed for *S. galinae*, ash tree-size category had no significant effect on overwintering larval mortality for *T. planipennisi* (likelihood ratio chi-square test: $\chi^2 = 1.28$, $df = 1$, $P < 0.2556$), and study site had a highly significant effect (likelihood ratio chi-square test: $\chi^2 = 321.73$, $df = 5$, $P < 0.0001$).

Among all emerald ash borer larvae observed ($n = 1,434$) (excluding those killed by woodpeckers, parasitoids, and other undetermined factors), cold weather caused 4.5–26.2% of emerald ash borer larval mortality across study sites (Fig. 2C). Similar to the cold-injury-induced mortality patterns observed for the overwintering parasitoid larvae, ash tree size did not significantly affect the overwintering mortality of emerald ash borer larvae (likelihood ratio chi-square test: $\chi^2 = 0.1328$, $df = 1$, $P = 0.7155$), and the effect of study site was highly significant (likelihood ratio chi-square test: $\chi^2 = 58.06.73$, $df = 5$, $P < 0.0001$).

Mortality of Overwintering Parasitoids and Emerald Ash Borer Larvae in Connecticut

For *S. galinae*, we observed a total of 843 overwintering larvae in 128 broods at the three study sites in Connecticut, where fewer, larger diameter trees (DBH = 9–21 cm) were sampled than in Michigan. As no ash saplings were available to sample at the Connecticut sites, only three *T. planipennisi* broods (totaling 115 individuals) were observed attacking emerald ash borer in the trees we sampled (Fig. 3). The observed overwintering mortality rate from cold injury in *S. galinae* was low (0–7.3%), and it varied significantly among sites (likelihood ratio chi-square test: $\chi^2 = 321.73$, $df = 5$, $P < 0.0001$). No mortality of the overwintering *T. planipennisi* larvae due to cold was observed in the three broods. The overwintering mortality of emerald ash borer larvae observed ($n = 203$) at the Connecticut study sites was extremely low (0–1.8% per site).

Low Temperatures Caused by the 2019 Polar Vortex Event in Michigan and Connecticut

A record low temperature (minimum $\approx -26^\circ\text{C}$; mean ≈ -18 to -20°C) was recorded in Michigan during the 2019 polar vortex events (21–31 January), approximately -6°C below the normal seasonal low temperatures for this period (Fig. 4A and B) (National Oceanic and Atmospheric Administration 2019). In contrast, more normal low temperatures (minimum $\approx -18^\circ\text{C}$, mean $\approx -14^\circ\text{C}$) were recorded in Connecticut during the same period, approximately -2°C above the normal mean minimum temperature range (Fig. 4A and B) for this period. The daily minimum temperature was an average of 7.1°C ($\pm 4.1^\circ\text{C}$ 95% CI) lower in Michigan compared to Connecticut during this period (two-sample t -test, $t_{10} = -3.87$, $P = 0.003$) for the period from 21 to 31 January 2019.

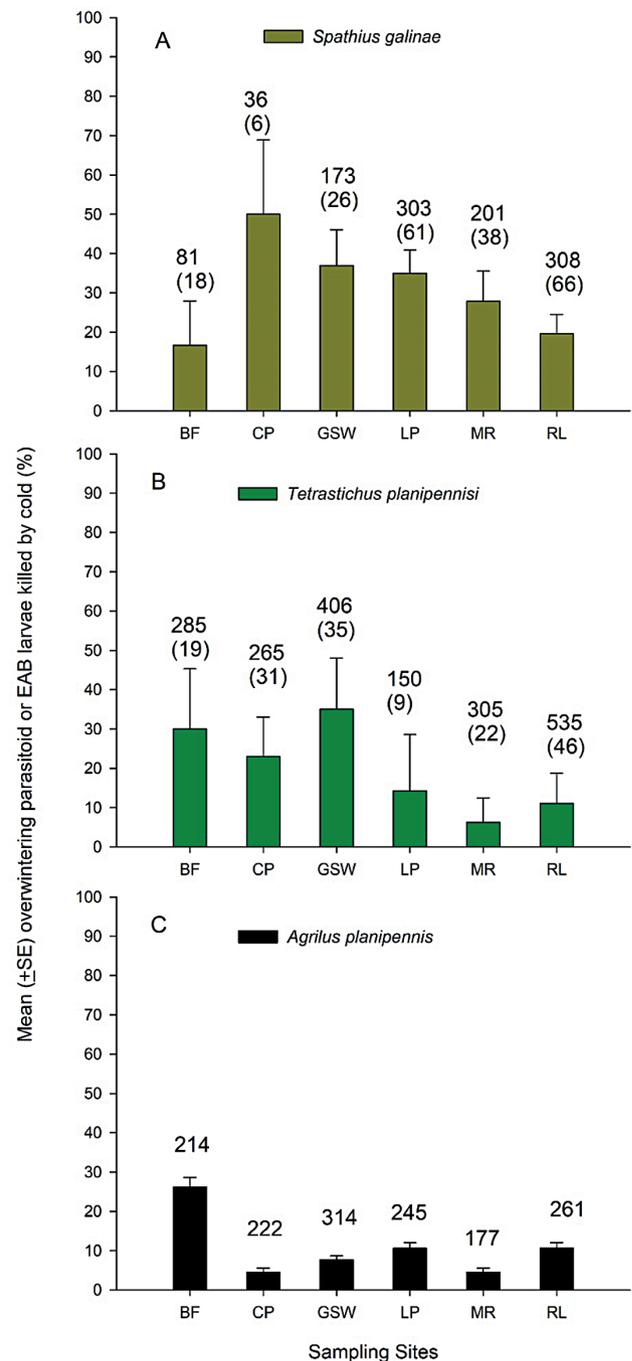


Fig. 2. Mean (\pm SE) percentage of larval mortality of overwintering *S. galinae* (A), *T. planipennisi* (B), and their larval emerald ash borer host (C) due to cold-induced injury at the six sampled study sites in southern Michigan. Numbers above each bar represent the total number of individuals observed or number of parasitoid broods (in parenthesis). Mean percentage and SE were calculated based on data pooled from ash saplings and pole-size trees sampled in early spring 2019.

Discussion

Our field observations from the six emerald ash borer biocontrol study sites in southern Michigan showed that the extreme cold wave that occurred in late January of 2019 resulted in low to moderate levels of overwintering mortality of both *S. galinae* (18–50%) and *T. planipennisi* (8–35%), but relatively less mortality of overwintering emerald ash borer larvae (4.5–26%). However, in

southern Connecticut where the cold wave was less extreme, we observed little or no (<2%) overwintering cold-related larval mortality of *S. galinae*, *T. planipennis*, or emerald ash borer. Moreover, our results from Michigan showed that, although the effect of this cold wave on the overwintering larval parasitoids and emerald ash borer varied significantly among the study sites, larval mortality from low temperature events during winter appeared to be higher for the two larval parasitoids than for their host emerald ash borer larvae in most (five out of six) study sites (Fig. 2A–C). This differential effect of extreme cold suggests that these two parasitoid species may be less cold-tolerant than emerald ash borer larvae. As result, parasitism rates may be lower in the coming 2020 field season, thereby favoring an increase in emerald ash borer population growth in affected areas (i.e., Michigan and other Midwestern states affected by the 2019 polar vortex event).

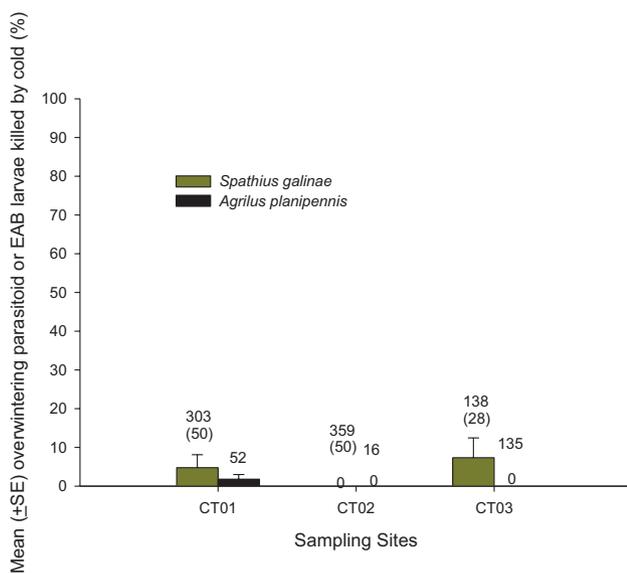


Fig. 3. Mean (\pm SE) percentage of mortality of overwintering larvae of *S. galinae* and its emerald ash borer host (*Agrilus planipennis*) at the three sampled study sites in southern Connecticut. Only three broods of live *T. planipennis* (with no overwintering mortality) were observed when sampling ash trees; thus, data for this parasitoid species were not presented in this figure.

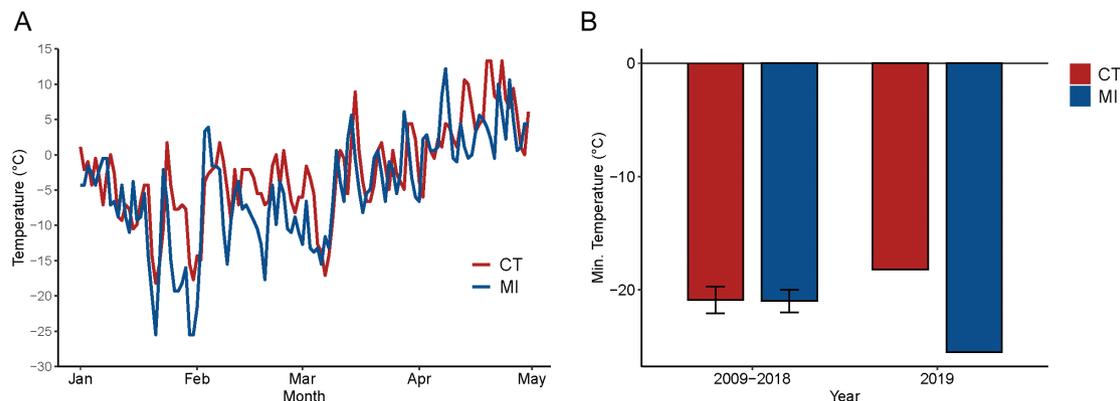


Fig. 4. Minimum temperatures recorded at nearby weather stations (Meriden-Markham Municipal Airport in Meriden, CT and Capital Area International Airport, Lansing, MI) from January to May 2019 (A); mean minimum (\pm SE) temperatures reported for the previous 10 yr (2009–2018) (NOAA 2019), and absolute minimum temperatures reported from the nearby weather stations for January to May 2019 (B).

Several published studies report that the overwintering stages of emerald ash borer and its larval parasitoids *S. galinae* and *T. planipennis* can lower their supercooling points or increase their cold-hardiness in response to below-freezing temperatures (Crosthwaite et al. 2011, Hanson et al. 2013, Christianson and Venette 2018, Chandler et al. 2020). This ability to adjust the degree of cold-hardiness may have prevented the week-long cold wave from causing even higher (>50%) mortality rates of these beneficial insects. Our field observations of larval mortalities exposed to the extreme cold event in Michigan in January 2019 suggest that these overwintering parasitoid larvae might be less tolerant of sudden cold events than overwintering emerald ash borer larvae.

A previous study with the invasive hemlock woolly adelgid, *Adelges tsugae* Annand (Hemiptera: Adelgidae), in eastern United States showed that the induction of cold-hardiness produced much year-to-year variation in cold-hardiness, in addition to genetic differences between northern and southern adelgid populations (Elkinton et al. 2017). Consequently, the cold temperatures required to kill hemlock woolly adelgids vary from year to year and week to week based on prior exposure to low temperatures at particular geographic locations and among different locations (Elkinton et al. 2017). Although we did not measure the cold-hardiness of emerald ash borer and its larval parasitoid populations observed in both Michigan and Connecticut in this study, we did observe significant variations in cold-related mortality of these insects among different study sites. We suspect that exposure to temperatures at our study sites before, during, and after each cold event might be highly variable due to the difference in forest types and/or topographic features, resulting in different mortality rates in these insects.

Atmospheric scientists have recently documented an increase in frequency of extreme winter cold events, especially in eastern North America, in the last decade associated with the rapidly warming Arctic and melting of the ice in the Arctic Ocean. (Francis et al. 2012, Cohen et al. 2018). Explanations of this phenomenon invoke changes to the strength and geographic amplitude of the jet stream that result in more extreme cold events followed by extreme warm events in winter (Kim et al. 2014, Kretschmer et al. 2016). Many studies show that cold-hardiness in insects is inducible by exposure to cold temperatures. For example, a recent laboratory study showed that hemlock woolly adelgids lowered their supercooling point, and thus became more cold-hardy following exposure to cold temperatures for as little as 3 d (Elkinton et al. 2017). They further showed that a cold event in February 2016 caused more mortality

following a warm January in that year, compared to an even colder event the previous year following warmer January temperatures. In other words, cold tolerance can develop in insects in response to cold temperatures as the winter progresses, but it takes time. Thus, the very rapid cycling between warm and cold events in response to global climate change may lead to higher winter mortality, despite the general warming of mean winter temperatures associated with global climate change.

Published data from several studies also show that the supercooling point of the overwintering stages of emerald ash borer (ranging from -28 to -35°C) may be lower than that of overwintering *S. galinae* (mean range: -26 to -27°C) or *T. planipennis* (mean range from -27 to -30°C) (Wu et al. 2007, Crosthwaite et al. 2011, Hanson et al. 2013, Chandler et al. 2020). Differences in supercooling points among these species could explain the disparity in overwintering mortality between emerald ash borer and its parasitoids that we observed in Michigan, given that a record low temperature (minimum $\approx -26^{\circ}\text{C}$; mean ≈ -18 to -20°C) was recorded during the 2019 cold wave (21st to 31st January). In contrast, the low temperature (minimum $\approx -18^{\circ}\text{C}$, mean $\approx -14^{\circ}\text{C}$) recorded in Connecticut during the period of the 2019 cold wave appeared to be well above the supercooling points of overwintering emerald ash borer and its parasitoids, which likely explains the lower mortality observed for overwintering larvae of emerald ash borer and its larval parasitoids at the Connecticut study sites.

Published studies have linked climate change to observed shifts in the distribution and phenology of various species, which pose a threat to global biodiversity (e.g., Parmesan 2006, 2007; Hance et al. 2007; Kistner-Thomas 2019). From the pest management perspective, climate change can also affect the potential for outbreaks of insect pests, either by direct impacts on pest survival or range expansion, or indirect impacts on pest abundance due to altered interactions with their natural enemies (Berggren et al. 2009, Klapwijk et al. 2012, Duan et al. 2014). The observed differences in overwintering larval mortality rates between emerald ash borer and its larval parasitoids (*S. galinae* and to a lesser degree, *T. planipennis*) due to the extreme cold wave in Michigan strongly suggest that climate change could reduce the efficacy of these two biological control agents for managing emerald ash borer populations. However, our study did not include direct, 'normal winter' temperature treatments as controls because of the logistical difficulties in creating such control treatments in natural forests in anticipation of rare weather events. Further studies with simulated extreme weather event using climate-controlled chambers or long-term field studies will help determine the effects of weather events such as that observed on the population dynamics of emerald ash borer and the two introduced larval parasitoids in Michigan after the polar vortex event of January 2019.

The *T. planipennis* populations reared and released for emerald ash borer biocontrol in North America originated in Jilin and Liaoning provinces, China (Liu et al. 2003, 2007; Federal Register 2007), and *S. galinae* from the vicinity of Vladivostok, Russia (Duan et al. 2012, Federal Register 2015). Although the collection sites in Asia for these two emerald ash borer larval parasitoids are similar in latitude (ranging from the 41st to the 43rd parallel) to our study sites in North America, these regions of Asia have a coastal climate, similar to Connecticut. In contrast, Michigan tends to have an inland, continental climate that is subject to bouts of arctic cold during winter when the polar vortex is destabilized. As a result of climate change, extreme low temperature events occurs with increasing frequency and intensity due to a weakened jet stream in the northern hemisphere. Therefore, we recommend expanding foreign exploration to more northerly and inland regions of Asia (e.g., Heilongjiang

province, China) for species and/or strains of potential emerald ash borer biocontrol agents with greater cold-hardiness, capable of surviving bouts of extreme low winter temperatures.

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