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Research

Behavior

Factors That Influence Flight Propensity in *Anoplophora glabripennis* (Coleoptera: Cerambycidae)

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Abstract

The effects of mating status, sex, beetle age, host quality, temperature, and wind speed on the propensity of *Anoplophora glabripennis* (Motschulsky) (Coleoptera: Cerambycidae) to take flight were evaluated using a free flight test in the laboratory. Time to initiate flight, the angle of flight, and flight capability (when beetles were enticed to take flight) were also evaluated. Host quality, mating status, beetle age, sex, temperature, and the interactions between one or more of these were all found to be significant predictors of flight in *A. glabripennis* in one or more of the experiments. Female flight propensity peaked at sexual maturity and declined thereafter. Both sexes had a higher propensity to take flight from a stem section of dry host material than from a fresh one. Most (78%) males flew at least once during the four mating status/ages tested from a fresh host stem section, while only 43% of the females flew at least once after chewing an oviposition pit. Flight propensity and distance flown increased with temperature and there was no voluntary flight at 15°C. Flight propensity did not increase with wind speeds 0.0–1.0 m/s, but no ascending flight was observed at 0.5 or 1.0 m/s. Time to flight initiation did not vary with the factors evaluated. Implications these results could have on the success of eradication programs are discussed. Specifically, what factors increase the propensity of mated females to disperse, effectively expanding the infestation zone.

Key words: Asian long-horned beetle, invasive species, flight

The adult stage of insects is specialized for dispersal and reproduction. Dispersal, which is defined as the spreading of individuals away from each other (Begon et al. 2009), is primarily accomplished through flight. Flight that results in dispersal can be of two types: routine to seek and exploit resources, or specialized for longer distance displacement and settlement away from the natal site (Van Dyck and Baguette 2005). In insect species that do not migrate, the frequency distribution for flight distance is right-skewed and long distance fliers make up a small percentage of the population (Steyn et al. 2016). Longer distance fliers would likely be the colonizers of new habitats and important to a species ability to survive in fragmented landscape where host patches are far apart. Routine flight is associated with daily activities like finding food, seeking a mate, finding shelter, finding an oviposition site, etc. and results in a small net displacement. Flight propensity changes with 1) the insect's biology in response to sensory inputs and level of sclerotization for example, 2) external environmental factors such as resource availability or risks of finding them, and 3) internal time-dependent factors like deprivation and sexual maturity/receptivity (Bell 1990, Arnold et al. 2016). In addition, insect flight is the most costly physiological process in the animal kingdom and the adult may use stored reserves from the larval stage or have to acquire additional resources before flight (Candy et al. 1997, David et al. 2015). In general, insect flight is low in immature adults, peaks at sexual maturity, and then slowly declines with age (Dingle 1972).

Anoplophora glabripennis (Motschulsky) (Coleoptera: Cerambycidae) is a xylophagous beetle native to China and the Korean Peninsula (Lingafelter and Hoebeke 2002). It is a pest on its preferred host genera: *Acer* L. (Aceraceae), *Populus* L. (Salicaceae), *Salix* L. (Salicaceae), and *Ulmus* L. (Ulmaceae) both in native and introduced habitats (Hu et al. 2009, Meng et al. 2015). Introduced populations are under eradication in both North America and Europe (Haack et al. 2010). One of the most challenging aspects of the eradication efforts is locating all the infested trees, so they can be destroyed. This is primarily because this beetle spends most of its life cycle inside the tree, so identifying infested trees depends on spotting signs of its presence like oviposition pits or adult exit holes, which are often high in the tree. The beetles walk around their host trees to find mates, feed, and oviposit; only flying when necessary. Another factor that complicates locating all infested trees is an incomplete understanding of this beetle's flight behavior in the diverse habitats and under different environmental conditions in which the infestations exist. The programs have to use the best data available on potential flight distance and direction obtained from mark–release–recapture tests, flight mills, reconstructions of how beetles infested trees in invaded landscapes, and modeling.

The speed and distance at which this *A. glabripennis* moves have been estimated using both the laboratory and field methods already mentioned. The reconstructions used dendrochronology to age beetle exit holes and oviposition pits, locations of infested hosts on the landscape, and modeling to estimate distances moved. The distance *A. glabripennis* will fly varies greatly with the landscape in which they are moving. Based on several mark–release–recapture studies carried out in China beetles move shorter distances when the trees are smaller or closer together (106 m in 20–28 d, Wen et al. 1998) than when they are larger or when gaps between trees are greater (266 m in a season, Smith et al. 2001). These distances moved are similar to reconstructed actual distances moved in infested urban and industrial areas averaged 260 m in 5 yr in warmer areas (United States: Sawyer et al. 2011) and 234–300 m in 9 yr in colder areas (Canada, United Kingdom: Turgeon et al. 2015, Straw et al. 2016). Beetles have moved longer distances in more open landscapes with sparse trees 3.2 km in 5 yr and in woodlots 200 m in 9 yr (Sawyer et al. 2011). Maximum distances beetles traveled in a season were reported to be 2,644 m for females and 2,394 for males (Smith et al. 2004).

The average speed of flight in the field using harmonic radar was 1.8 m/s and the maximum observed speed was 5.3 m/s (Williams et al. 2004). Flight mill data point to slightly slower average speeds (1.1 m/s), but shown it varies with age and mating status (Lopez et al. 2017). The same flight mill study showed that maximum daily distance *A. glabripennis* can travel was much higher (13.7 km/d) than in the mark–release–recapture studies, but that only 5% of the beetles traveled >8k m/d (Lopez et al. 2017). Another flight mill study that flew beetles 2 hr twice a week for life showed they could fly 14 km in their life span (Javal et al. 2018). Therefore, the maximum distance a beetle could travel if it does not come in contact with a landing site is much longer than what field trials suggest but more in line with what the reconstructions suggest. Modeling efforts that employed data on infested trees in the eradication zones suggests that rarer longer distance dispersal events, 8 km in Worcester, Massachusetts and 2.2 km in Cornuda, Italy, have apparently occurred when beetle population densities were higher (Favaro et al. 2015, Trotter and Hull-Sanders 2015).

All of the existing information on *A. glabripennis* movement has been used by eradication programs to set survey boundaries around infested trees and, when conditions warrant, to determine how large an area around an infested tree to remove all potential host trees. Removal of all hosts can potentially remove asymptomatic infested trees that are missed in visual surveys and reduce the need for repeated surveys in high-risk areas. Understanding what conditions cause the beetles to take flight, and how those conditions might influence flight distances will be instrumental in refining the program boundaries and eradication protocols for this pest. Keena and Sánchez (2018b) have documented that the probability of flight for *A. glabripennis* males significantly increases when sex ratios are male skewed, especially after aggressive male–male encounters. Models have provided evidence that dispersal is density dependent, and that tree characteristics, temperature, and humidity correlate with dispersal propensity (Bancroft and Smith 2005). This study will directly assess the effects of mating status, sex, beetle age, host quality, temperature, and wind speed on *A. glabripennis* propensity to take flight under laboratory conditions. Time to initiate flight, the angle of flight, and flight capability (after enticing the beetles to take flight) are also evaluated. Implications of the results for eradication programs will also be discussed.

Materials and Methods

Insects and Rearing Conditions

In these experiments, beetles originating from different geographic locations were used (Table 1). The beetles were either just emerged from wood or from the first four generations of laboratory colonies. The flight bioassays were conducted in 2000–2002. Infested branch sections were transported under permit to the USDA-Forest Service quarantine facility in Ansonia, Connecticut. Voucher specimens of each population were deposited at the Entomology Division, Yale Peabody Museum of Natural History, New Haven, Connecticut.

General colony methods for rearing larvae on artificial diet and obtaining eggs for the next generation are given in Keena (2005). Newly emerged adults that had been reared on artificial diet were held in the dark for 4–5 d to allow their exoskeleton to sclerotize before being weighed and fed. Virgin adults, both diet-reared and emerged from wood, were fed fresh *Acer saccharum* Marshall (sugar maple) twigs (3–7 mm diameter with leaves removed) and held individually in 950 ml glass jars until needed for the flight assays. Adults were held, both before and after the behavioral assays, at either 20 ± 2°C or 25 ± 2°C and 60 ± 5% RH and subjected to a photoperiod of 16:8 (L:D) h. Once mated, pairs were held in a 3.8-liter glass jar and weekly provided fresh *A. saccharum* twigs for food and a bolt (3–7 cm diameter and 20 cm long) with both ends waxed as an oviposition substrate. Fresh *A. saccharum* twigs and bolts were obtained bi-weekly and monthly, respectively, and stored at 10°C and ≥80% RH until used.

General Bioassay Setup and Procedures

All flight assessments were conducted in a white room measuring 8 × 7 m with a ceiling at 2.5 m (Fig. 1). The temperature and humidity were maintained at desired test levels using space heaters and a humidifier. A large pedestal fan was used to help evenly heat the room before trials and to create appropriate wind speeds for each experiment. A single spot light (100 W grow light bulb) was mounted

Table 1. Approximate location (latitude and longitude) of source populations and designations for populations of *A. glabripennis* evaluated in this study

Population	State	City	Latitude	Longitude	Collection date	Total individuals from wood to start colony
C	Illinois	Ravenswood	41.58 N	87.42 W	February 12, 1999	1,450
B1	New York	Bayside	40.45 N	73.45 W	April 5, 1999	384
B2	New York	Astoria/Long Island City	40.76 N	73.92 W	May 21, 1999	204
F	New York	Flushing	40.74 N	73.85 W	August 3, 2000	711
S	New York	Sunnyside	40.74 N	73.92 W	June 21, 2000	118
M1	New York	Maspeth	40.73 N	73.91 W	February 17, 2000	181
M2	New York	Maspeth	40.73 N	73.91 W	December 12, 2001	135

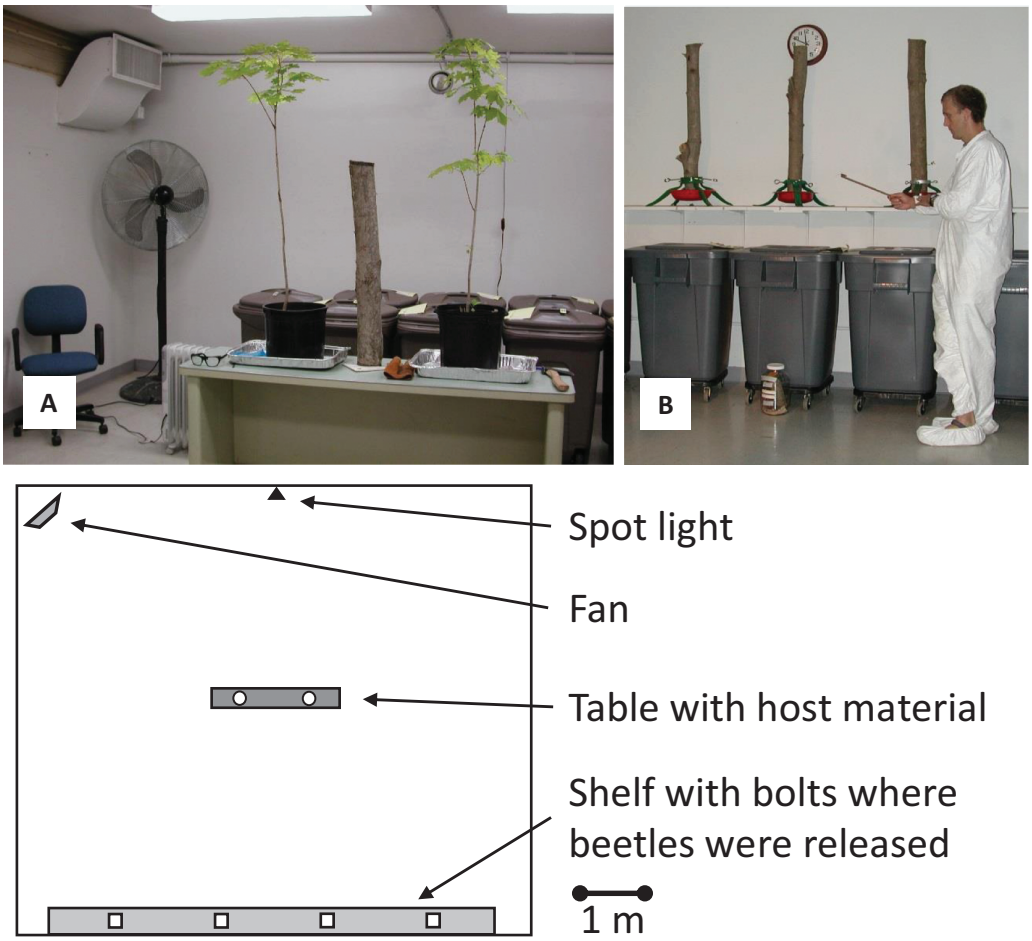

Fig. 1. Flight bioassay room diagram and photos of: (A) the wall opposite the beetle release points showing the positions of the pedestal fan, spot light, heater and table containing the fresh maple stem section and potted Norway maple saplings and (B) shelf with three fresh maple stem sections where beetles were released with experimenter enticing a beetle to fly using a wooden ruler.

at ceiling height across the room from the release point to simulate the sun. A Kestrel 3000 pocket weather meter (Nielsen-Kellermans, Boothwyn, PA) was used to measure the conditions in the room at the beginning of each flight trial. Along one wall, a 40-cm-wide by 7-m-long shelf, 1 m from the mottled white floor, was used as the release point for all flights (Fig. 1b). On the shelf were placed four, 90-cm-tall *A. saccharum* sapling stem sections with bark. Two types of stem sections were used: dry logs (neither end waxed and allowed to dry for >3 wk) mounted on 20 cm^2 pieces of 2 cm thick plywood and fresh logs with one end waxed and the other end in a tree stand with water. In the middle of the room a fresh section of *A. saccharum* sapling (1 m tall 10 cm diameter) and two *Acer platanoides* L. (Norway maple) potted trees (1.5 m tall) were placed on a table (Fig. 1a). Each adult that was flight-tested was identified either by its unique elytral spot pattern or by using a marker to color one white spot on one elytron. The beetles were moved slowly from the glass jar to the bolts at the release point using a twig, to minimize disturbance. Each bioassay consisted of one to four adults, with opposite sexes separated by at least one bolt without beetles. Beetles were given 45 min to voluntarily take flight. After the observation period had ended or after a voluntary flight had been completed, the beetle was enticed to fly to determine its flight capability. This was done by allowing the adult to walk on to a wooden ruler and holding it at shoulder height facing a sidewall while standing near the release point (Fig. 1b). The beetle was allowed to walk on the twig. The twig was turned as necessary to keep the beetle from walking on to the observer. If after 5 min the beetle had not taken flight, it was nudged using another twig and/or the ruler was bounced. This type of disturbance often resulted in a takeoff. The following behavioral responses and durations were recorded: 1) initial direction of movement on the bolt (up or down), 2) did the beetle chew any pits in the bolt during the trial, 3) time the beetle started walking on the bolt, 4) time flight was initiated, 5) the flight angle (ascending, level, descending), 6) distance flown (as a straight line displacement) from the bolt in half-meters, 7) description of the general flight path, and 8) the substrate on which the beetle landed.

Mating Status and Age Experiment

Twenty females from strain M2 (Table 1) that emerged from wood were tested eight times during their life: 1) the day of emergence; 2) 1 wk after emergence; 3) 1 d after mating; and 4) at weekly intervals for 5 wk. Following testing at 1 wk after emergence, females were mated within 2 d with similar aged males from the same population. Pairs were observed to ensure that they copulated for at least 3 min, which would ensure insemination when the beetles were sexually mature (Keena and Sánchez 2018a). The room was maintained at 24 ± 1°C and 50 ± 5% RH. Fresh *A. saccharum* sapling stem sections were used as the release point and wind speed at 0.0 m/s where the beetles were released.

Host Quality and Mating Status Experiment

About half the beetles tested were from populations F, M, and S (Table 1) and had emerged from fresh host material. The remainder were from the second laboratory generation on artificial diet of populations B, C, M, and S (Table 1). Thirty-two males and 30 females each were flight tested using dry and fresh sapling stem sections (496 beetles total) at the release point. Dry and fresh stem sections simulated host material assumed unsuitable and suitable (Meng et al. 2015), respectively, for oviposition and larval development. Beetles from each combination of population and larval food source type (wood or artificial diet) were sequentially assigned to the two host treatments. Each adult was flight tested, always on the same type of host log, at four times during its life: 1) newly emerged (5 d after emergence if artificially reared), never fed, and unmated; 2) 1 wk after the first test, unmated, and well fed; 3) 1 d after mating (beetles were mated right after the second test) and well fed; and 4) 5 wk after mating and well fed. "Well fed" beetles had more fresh maple twigs to feed on than they could use in the time between jar changes. The room was maintained at 24 ± 1°C and 58 ± 8% RH. The fan was not running resulting in wind speed of 0.0 m/s at the release point.

Wind Velocity Experiment

The beetles used in this experiment were from the third laboratory generation of populations B and C (Table 1) reared on artificial diet. Ten to 12 mating pairs from each population being held at 20 or 25°C (part of another study) were assigned to each of the wind treatments when first mated. However, several adults had wing deformities or died prior to testing. Because no additional pairs were available, we tested only 9–24 beetles per sex per wind speed (97 total beetles). The beetles were flight tested 1 wk after being mated. The fan was placed along the wall opposite the host stem sections and running at a medium setting, low setting, or turned off to achieve the desired wind speeds of 1.0 ± 0.05, 0.6 ± 0.08, and 0.0 m/s, respectively. The wind speed was measured at the fresh host stem section where the beetle was released. The room was maintained at 25 ± 1°C and 54 ± 6% RH. Fresh *A. saccharum* sapling stem sections were used.

Temperature Experiment

The beetles used in this experiment were from the fourth laboratory generation of the B and C populations (Table 1) reared on artificial diet, 80 beetles from each population. Ten (coldest temperature only) or 15 mating pairs from each population were sequentially assigned to each of the temperature treatments when first mated and both sexes were flight tested 1 wk later. The three temperatures evaluated were: 16 ± 1°C, 23 ± 1°C, and 30 ± 1°C. The relative humidity was 50 ± 5% and the wind speed was 0.0 m/s. Fresh *A. saccharum* sapling stem sections were used as release points.

Statistical Analyses

The fit of the data on the time to take flight and distance flown to a normal, gamma, and lognormal distribution was evaluated using PROC UNIVARIATE (SAS Institute 2015). The Shapiro–Wilk test was used to assess normality. These two continuous variables were analyzed in PROC GLIMMIX (SAS Institute 2015) using a gamma distribution with a log link. Potential predictors of the probability of beetle flight, a dichotomous variable, were evaluated using a binary distribution with a logit link. Data from the mating status and age experiment were evaluated using a repeated measures design with an unstructured covariance matrix (UN) and treatment (combination of mating status and age) was assessed as a potential predictor of flight. A repeated measures analysis with a heterogeneous autoregressive covariance matrix (ARH[1]) and a random effect of larval food source was used in analyzing the data on host quality and mating status. In the host quality and mating status experiment, the following were evaluated as potential predictors of flight: host quality, mating status, sex, and the two- and three-way interactions between the three. In the temperature and wind speed experiments treatment (temperature or wind speed), sex, and the interaction between the two were evaluated as potential predictors of flight. Note that initially strain was included as a random effect in each experiment where more than one strain was used and in each case the variance component associated with it was estimated to be zero, so it was removed from the model. For each model, residuals were evaluated using Levene's test to assess homogeneity of variance. Differences among means were determined by the least-squares means test with $\alpha = 0.05$ and a conservative Tukey–Kramer grouping (SAS Institute 2015). Chi-square tests were conducted using Statistix 10 software (Statistix 2013) to determine if the proportion of individuals that took flight versus those that remained on the maple stem sections was significantly different >50% and for comparing other proportion data from the experiments. Multilevel chi-square tests were run using PROC FREQ (SAS Institute 2015) for proportion data using the same factors that were evaluated in the previous analyses for each experiment. When a significant difference was found, the proportions for individual treatments were compared using chi-square tests (Statistix 2013). A McNemar test for agreement (McNemar 1947) was done using PROC FREQ (SAS Institute 2015) to compare the distance flown and flight angle observed for individuals that flew both voluntarily and after enticement.

Results

Takeoff, Flight, and Landing Behaviors

To take off, the beetles wagged their antennae (Fig. 2a), opened their elytra (Fig. 2b), unfurled their wings (Fig. 2c), began to fan (Fig. 2d), and leapt off the substrate (Fig. 2e and f). During flight, beetles held their legs to the side and back toward their dorsum. The flight path the beetle took after leaving the substrate was descending, level or ascending, and occasionally started out level and then became ascending. At landing, the sterna were first to hit the substrate and the in some cases, the beetle bounced off the substrate.

Female Mating Status and Age

The combination of mating status and age was a significant predictor of female Asian long-horned beetle flight ($F = 3.73$; df = 7, 19; $P = 0.0105$). Significantly less than 50% of the females that had been mated 14–35 d earlier took flight (Fig. 3). In addition, significantly more 7 d virgin females took flight than did 21 or 28 d mated females (Fig. 3).

Host Quality and Mating Status

Host quality ($F = 29.64$; df = 1, 120; $P < 0.0001$), sex ($F = 5.00$; df = 1, 120; $P = 0.0272$), mating status ($F = 2.78$; df = 3, 360; $P = 0.0408$), the interaction between host quality and sex ($F = 6.41$; df = 1, 120; $P = 0.0127$), and the interaction between host quality and mating status ($F = 3.21$; df = 3, 360; $P = 0.0233$) were all significant predictors of female Asian long-horned beetle flight, but the interaction between the three was not ($F = 1.81$; df = 10, 360; $P = 0.058$). Significantly less than 50% of both sexes on fresh bolts took flight regardless of mating status (Fig. 4). In addition, significantly more 7 d virgin and 21 d mated individuals on dry bolts took

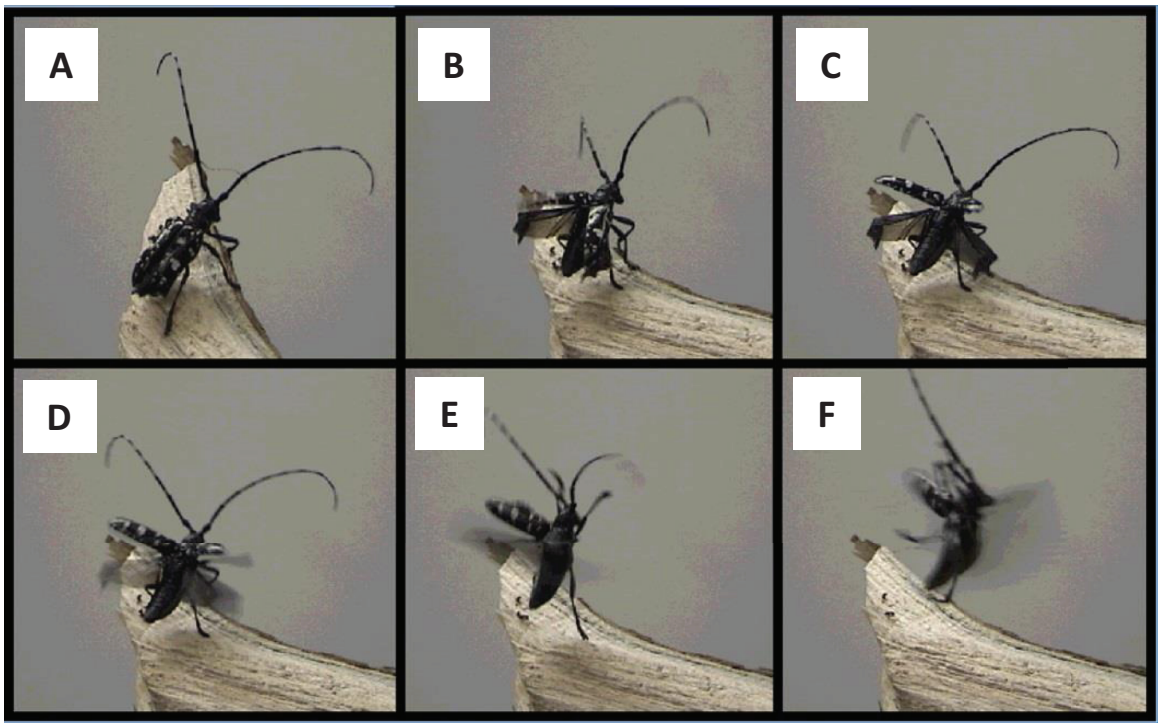

Fig. 2. Photos showing the *A. glabripennis* beetle preflight and launch from a piece of wood. The beetle wagged its antennae (A), opened its elytra (B), unfolded the under wings (C), fanned briefly (D) and jumped off holding its legs to the side and back toward the body (E and F).

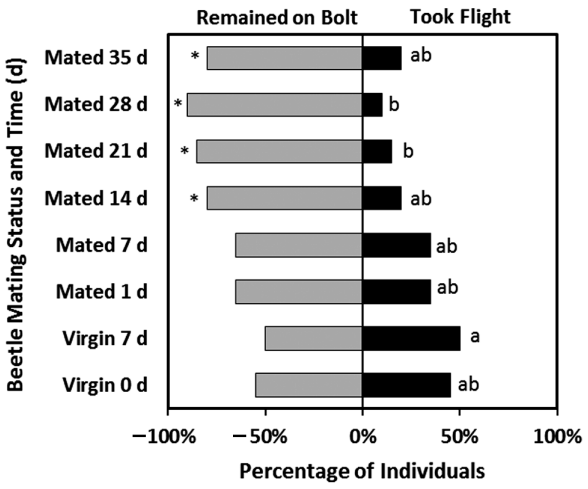

Fig. 3. Comparison of the percentage of *A. glabripennis* females of different mating statuses that responded by taking flight or by remaining on the bolt. Bars preceded by "*" indicate significantly different responses within a mating status (χ^2 tests at $\alpha = 0.05$). Bars followed by different letters indicate significant differences among mating statuses.

flight than did mated (1 and 21 d) and virgin 7 d individuals on fresh bolts (Fig. 4b).

The flight angle observed was not significantly different for individuals that flew both voluntarily and after enticement (S = 5.64, df = 3, $P = 0.1305$). The only significant variation in flight angle was on the fresh host where more descending flight occurred in both sexes than did level flight in females (Fig. 5).

Host quality ($F = 0.10$; df = 1, 113; $P = 0.7472$), sex ($F = 1.18$; df = 1, 113; $P = 0.2799$), mating status ($F = 2.21$; df = 3, 85; $P = 0.0924$), the interaction between host quality and mating status ($F = 1.40$; df = 3, 85; $P = 0.2495$), the interaction between host quality and sex ($F = 1.79$; df = 3, 113; $P = 0.1831$), and the interaction between the three ($F = 1.11$; df = 6, 102; $P = 0.3639$) did not have a significant effect on the time it took Asian long-horned beetles to take flight. Host quality ($F = 0.49$; df = 1, 121; $P = 0.4860$), sex ($F = 0.04$; df = 1, 121; $P = 0.8429$), mating status ($F = 1.97$; df = 3, 88; $P = 0.1242$), the interaction between host quality and mating status ($F = 0.52$; df = 3, 88; $P = 0.6681$), the interaction between host quality and sex ($F = 0.03$; df = 1, 121; $P = 0.8637$), and the interaction between the three ($F = 0.4$; df = 6, 106; $P = 0.8796$) also did not have a significant effect on the distance Asian long-horned beetles flew. The flight angle observed was not significantly different for individuals that flew both voluntarily and after enticement (S = 5.64, df = 3, $P = 0.1305$). There was however a significant difference in the distance flown when beetles flew voluntarily and after being enticed to fly (S = 66.78, df = 21, $P < 0.0001$). When the distance flown voluntarily was subtracted from that flown after enticement significantly more beetles flew a longer distance after enticement than voluntarily (Yate's corrected $\chi^2 = 104.07$, $P = 0.0000$; Fig. 6). Also, most beetles that did not fly voluntarily were capable of ≥1 m of flight.

Significantly more females chewed an oviposition pit (Yate's corrected $\chi^2 = 29.57$, $P < 0.0001$) during the 45-min trial on the fresh host stem section (64%) than did those on a dry stem section (28%), while males chewed both stem section types equally (7% dry and 9% fresh). In addition, significantly more females took flight (Yate's corrected $\chi^2 = 16.79$, $P < 0.0001$) after chewing a pit on the dry host stem section (32%) than did on the fresh stem section (3%). Beetles that flew however were not attracted to the host material in the room; only 6% landed on host material and instead most landed on the first surface they contacted.

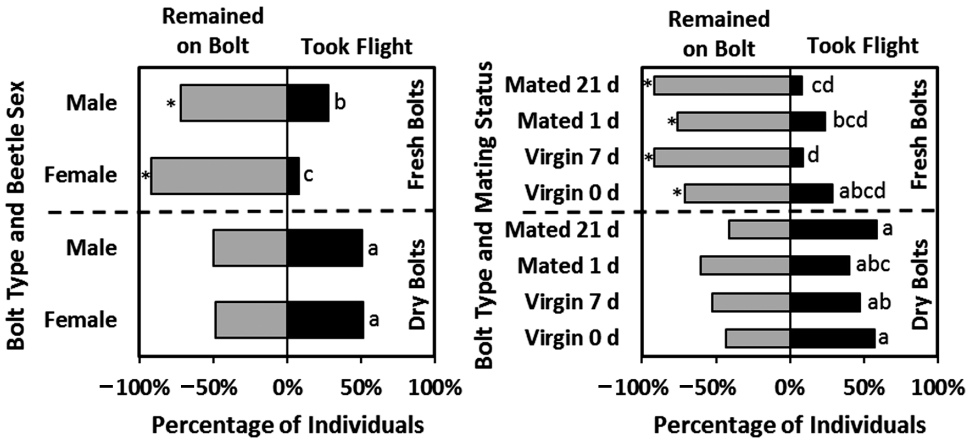

Fig. 4. Comparison of the percentage of *A. glabripennis* males and females (A), and of different mating statuses (B) that took flight or remained on either a fresh or dry *Acer* stem section. Bars followed by different letters were significantly different and for bars preceded by a "*" there were significant responses based on χ^2 tests at $\alpha = 0.05$.

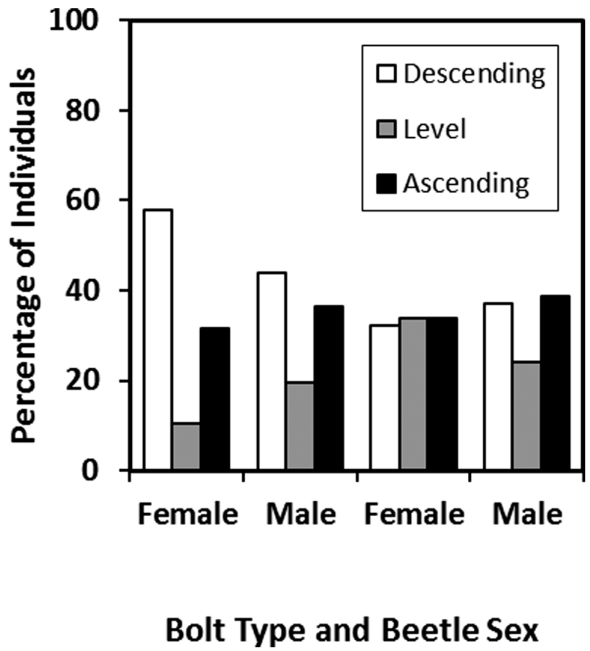

Fig. 5. Percentage of *A. glabripennis* individuals exhibiting each type of flight by sex and type of maple stem section they were placed on. Bars with different letters were significantly different based on χ^2 tests at $\alpha = 0.05$.

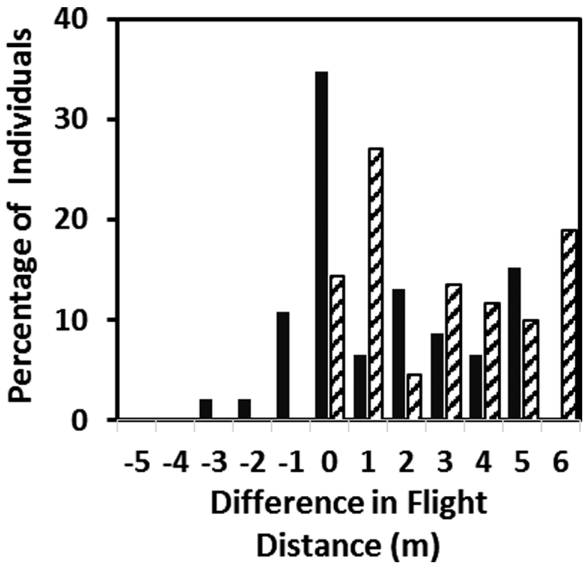

Fig. 6. Percentage distributions of the difference in the distance (m) flown (enticed distance minus voluntary distance) for *A. glabripennis* individuals that flew both voluntarily and after being enticed (solid bars) and only flew after being enticed (striped bars, using zero for the voluntary distance).

Temperature and Wind Velocity

Temperature ($F = 29.64$; df = 1, 120; $P < 0.0001$) was a significant predictor of Asian long-horned beetle flight, but sex ($F = 1.81$; df = 10, 360; $P = 0.0058$) and the interaction between temperature and sex ($F = 1.81$; df = 10, 360; $P = 0.0058$) were not. Significantly more than 50% of the beetles at each temperature remained on the fresh host stem section (Fig. 7). As temperature increased from 15 to 30°C significantly more beetles flew. Although no beetles voluntarily flew at 15°C, 10% flew after being enticed, significantly fewer than flew after being enticed at 22.5 (72%) and 30°C (95%). For beetles that flew there was no significant difference in flight distance between the voluntary and enticed flights (S = 22.00, df = 21, $P = 0.3995$). The distance beetles flew when enticed increased significantly as temperature increased ($F = 3.46$; df = 2, 97; $P = 0.0354$), but did not vary significantly by sex ($F = 3.39$; df = 1, 97; $P = 0.0686$) or the interaction between temperature and sex ($F = 0.90$; df = 2, 97; $P = 0.4095$). When enticed beetles flew an average (±SE) of 0.97 ± 0.50, 1.85 ± 0.28, and 2.68 ± 0.35 m at 15, 22.5, and 30°C, respectively.

Neither wind speed ($F = 1.85$; df = 2, 91; $P = 0.1631$), nor sex ($F = 2.73$; df = 1, 91; $P = 0.1020$), nor the interaction between wind speed and sex ($F = 0.22$; df = 2, 91; $P = 0.8010$) were significant predictors of Asian long-horned beetle flight. However, significantly more than 50% of the beetles at 0.5 and 1.0 m/s remained on the fresh host stem section for the entire 45 min (Fig. 8). Neither wind speed ($F = 1.96$; df = 2, 29; $P = 0.1585$), nor sex ($F = 0.24$; df = 1, 29; $P = 0.6263$) nor the interaction between wind speed and sex ($F = 0.04$; df = 2, 29; $P = 0.9606$) had significant effects on the distance Asian long-horned beetles flew. Wind speed had a significant effect on the proportion of individuals exhibiting different flight

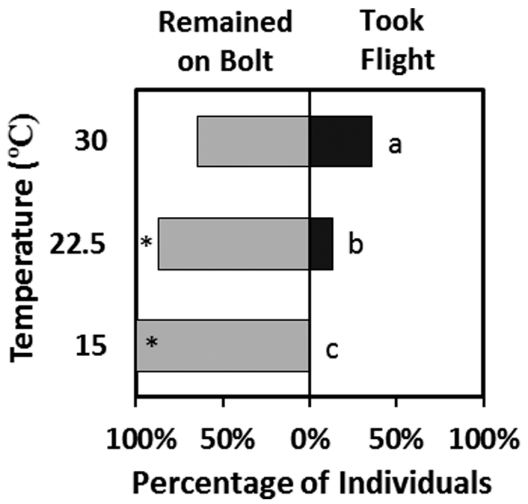

Fig. 7. Comparison of the percentage of *A. glabripennis* males and females mated for 7 d that took flight or remained on a fresh *Acer* stem section when exposed to different temperatures (°C). Bars followed by different letters were significantly different and for bars preceded by a "*" there were significant responses based on χ^2 tests at $\alpha = 0.05$.

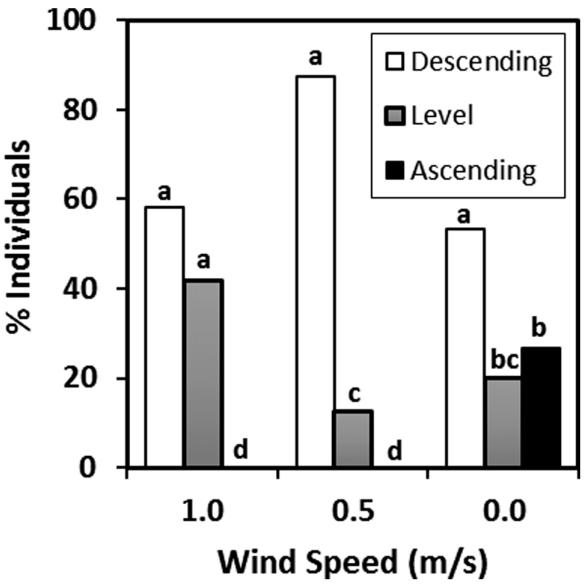

Fig. 9. Percentage of *A. glabripennis* individuals exhibiting each type of flight when exposed to three different wind speeds. Bars with different letters were significantly different based on χ^2 tests at $\alpha = 0.05$.

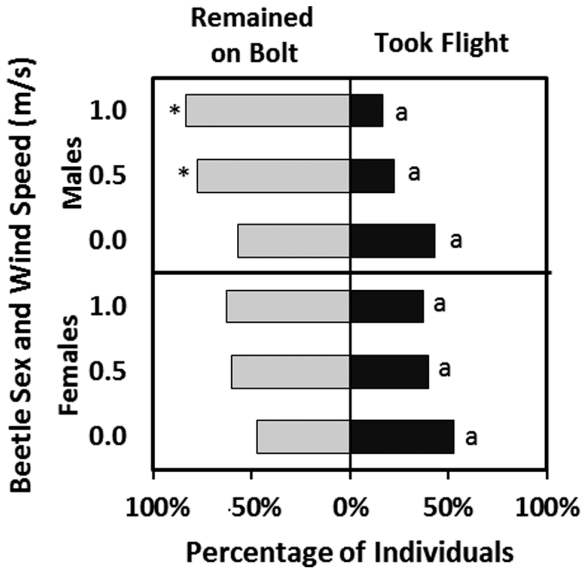

Fig. 8. Comparison of the percentage of *A. glabripennis* males and females mated for 7 d that took flight or remained on a fresh *Acer* stem section when exposed to different wind speeds (m/s). Bars followed by different letters were significantly different and for bars preceded by a "*" there were significant responses based on χ^2 tests at $\alpha = 0.05$.

angles (likelihood ratio $\chi^2 = 9.6107$, df = 4, $P = 0.0475$; Fig. 9). Most notably there were no ascending flights observed at 0.5 or 1.0 m/s wind speeds.

Discussion

Host quality, mating status, beetle age, sex, and temperature were all significant predictors of flight in *A. glabripennis* in one or more experiments. Time to flight was not affected by any of the factors evaluated and distance flown only differed significantly with temperature. Angle of flight was affected by wind speed. More beetles were capable of flight when involuntarily tested than voluntarily took flight. Females assessed the host quality by chewing oviposition-type pits, even on the dry dead host stem sections, before making a choice to stay or fly away. Although wind speed over the range tested was not a significant predictor of flight, there was some indication that flight was affected by wind speed since there was a significant response in males at the 0.5 and 1.0 m/s and no individuals of either sex exhibited ascending flights at these wind speeds.

A decline in female flight propensity after reaching sexual maturity, as was seen in the first experiment, fits with the normal pattern seen in many insects. In other insects, degradation of the flight muscles and the neurons that control flight occur with age (Ridgel and Ritzmann 2005). In *A. glabripennis* significant reduction in flight did not occur until 21 d post-mating (35 d post-emergence from the wood) compared with the 7 d virgin (first time tested after sexual maturity). The same pattern of 21 d mated females flying less than 7 d virgin females, however, was not seen in the second experiment on the fresh host stem section (same host as used in the first experiment). The second experiment used about half of the beetles that emerged from wood and half that were reared on artificial diet, so this may have confounded the results. The artificial diet reared adults would be less sexually mature at each time frame than the ones that emerged from wood since it takes about a week and a half for them to chew out of the wood at the temperatures the bolts were held (Sánchez and Keena 2013). Also when beetles emerge from wood, their exoskeleton has more completely sclerotized, while the 0-d artificial diet reared adults are still soft and more vulnerable to damage. In red flour beetles, *Tribolium castaneum* (Herbst) (Coleoptera: Tenebrionidae), non-reproductive adults that are still sclerotizing move less than more mature beetles, likely to avoid aggression and predation while vulnerable (Arnold et al. 2016).

A higher propensity to take flight from a dry host stem section than from a fresh one was expected. When the current habitat is no longer usable for reproduction or feeding, the beetles would naturally disperse to seek a new habitat. Females made obvious assessments of the host resource by chewing oviposition-type pits, but

male assessments were not obvious. However, recent work on *A. glabripennis* sensilla on the palps has shown that the beetle is sensitive to moisture, so both sexes may have used these sensors in assessing host quality (Hall 2018). The flight angle and distance flown away from an unsuitable habitat will be based on the beetle's perception of habitat patchiness. In more fragmented habitats where host trees are farther apart or scarce, beetle movements would be expected to change from routine resource exploration to specialized directed longer distance movements outside the current habitat to find a suitable host (Van Dyck and Baguette 2005). There was an indication that this may have happened in the host quality experiment because of the differences in flight angles observed on the fresh host stem sections. Also in China, when no trees were close by *A. glabripennis* adults flew spiraling upward (Smith et al. 2004).

When host quality was good, significantly more males flew at least once in the four times they were tested than did females. Seventy-eight percent of males flew, while only 43% of the females took flight. This is consistent with what was found when *A. glabripennis* movement was tracked in the field using harmonic radar, most females stayed on the healthy host they were released on while most males moved from tree to tree (Williams et al. 2004). Since males usually are the sex that makes first contact before mating and they are also likely to fly after aggressive encounters with another male, it makes sense that they would tend to fly more (Keena and Sánchez 2018a,b). The tendency of females to stay on a viable host and oviposit rather than fly is likely why many studies have found that the beetles are slow to disperse from the initial point of infestation until the population builds up (Sawyer et al. 2011, Turgeon et al. 2015, Straw et al. 2016). It is known that virgin females will fly from a viable host when their drive to seek a mate is high enough since they can be captured many meters from the nearest tree with an exit hole in traps baited with a combination of male produce pheromone and kairomones (Nehme et al. 2014). Increasing beetle density also has been shown to increase short range dispersal in both sexes (Bancroft and Smith 2005).

It has been shown that climate (temperature over time) affects *A. glabripennis* generation time and adult emergence timing (Trotter and Keena 2016) and adult activity (Zhou et al. 1984, Keena 2006). *A. glabripennis* adults were active and successfully mated between 15 and 30°C (Keena 2006) and at 28–30°C adult activity greatly increased but declined at temperatures ≥35°C (Zhou et al. 1984). This is consistent with the results presented here where flight propensity increased over the 15–30°C temperature range and that almost no flight occurs, even when the beetles are enticed at 15°C. The absence of voluntary flights at 15°C may also partially explain the slow spread of *A. glabripennis* populations in cooler climates as Straw et al. (2016) hypothesized that temperatures at which the beetle flies would occur less often in colder climates. Tests at higher temperatures would likely have resulted in a decrease in propensity to fly. Field studies in China showed that temperatures around 32°C were associated with decreased dispersal (Bancroft and Smith 2005). Evaluations of the effects of humidity on dispersal by flight are limited to that of Bancroft and Smith (2005), and Zhou et al. (1984), which suggest that increases in relative humidity are associated with increased dispersal, except when it rains. Further work in this area may be warranted.

The average wind speeds measured during mark–release–recapture in China were documented to be about 1 m/s with gusts up to 3.5 m/s (Wen et al. 1998, Smith et al. 2004). Wen et al. (1998) found that wind speeds of 0.3–1.8 m/s correlated with increased flight and that 3.5 m/s correlated with reduced flight. This likely explains the lack of significant differences in flight propensity observed over the range evaluated in this study. If higher wind speeds had been tested, significant differences may have been seen. Despite the lack of differences in flight propensity, there were differences in the flight angle observed: no ascending flights were observed in the 0.5 or 1.0 m/s wind speeds, which may indicate that the beetle adjusts its flight pattern based on the wind. In China, the beetles were observed to fly upwind (Wen et al. 1998), but reconstructions of the spread of *A. glabripennis* in the Worcester, Massachusetts, infestation indicated significantly longer distance spread with the prevailing winds (Hull-Sanders et al. 2017). Further work on *A. glabripennis* flight responses to wind is needed to fully understand how adults orient to the wind and at what wind speeds they cease to fly.

Eradication programs would be most concerned with what increases the propensity of mated females to fly since their movements to new host trees would effectively spread the infestation. Based on the results of this study and previous studies that have also shown that negative female interactions with a conspecific can cause females to take fight (Keena and Sánchez 2018b) programs can expect more mated female flight where beetle densities are higher resulting in lower host quality and more negative conspecific interactions. They can also expect mated females to take flight when temperatures are over 15°C and the tree on which the female is sitting is disturbed or a perceived predator contacts the beetle. For example, I observed at least nine beetles flying away above the tree line from the heavily infested maple tree in Flushing Meadow Corona Park, New York when it was being cut on August 4, 2000 and helped collect an additional 24 live beetles off the ground (that came down with the limbs) before they could take flight (M.K., personal observations). In fact, Favaro et al. (2015) found that the maximum dispersal in the Cornuda, Italy infestation occurred in the year that the population density was high and partial management was implemented. It should also be noted that beetles that take flight when disturbed, as in the host quality study, would be expected to fly further and that distance could further increase if the habitat is fragmented or good hosts are sparse (Van Dyck and Baguette 2005).

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References Cited

Arnold, P. A., P. Cassey, and C. R. White. 2016. Maturity matters for movement and metabolic rate: trait dynamics across the early adult life of red flour beetles. Anim. Behav. 111: 181–188.

Bancroft, J. S., and M. T. Smith. 2005. Dispersal and influences on movement for *Anoplophora glabripennis* calculated from individual mark-recapture. Entomol. Exp. Appl. 116: 83–92.

Begon, M., J. Harper, and C. Townsend. 2009. Ecology: individuals, populations and communities, 2nd ed. Blackwell Scientific Publications, Boston, Oxford, London.

Bell, W. J. 1990. Searching behavior patterns in insects. Annu. Rev. Entomol. 35: 447–467.

Candy, D. J., A. Becker, and G. Wegener. 1997. Coordination and integration of metabolism in insect flight. Comp. Biochem Phys. B 117: 497–512.

David, G., B. Giffard, I. van Halder, D. Piou, and H. Jactel. 2015. Energy allocation during the maturation of adults in a long-lived insect: implications for dispersal and reproduction. Bull. Entomol. Res. 105: 629–636.

Dingle, H. 1972. Migration strategies of insects. Science 175: 1327–1335.

Favaro, R., L. Wichmann, H. P. Ravn, and M. Faccoli. 2015. Spatial spread and infestation risk assessment in the Asian longhorned beetle, *Anoplophora glabripennis*. Entomol. Exp. Appl. 155: 95–101.

Haack, R. A., F. Herard, J. H. Sun, and J. J. Turgeon. 2010. Managing invasive populations of Asian longhorned beetle and citrus longhorned beetle: a worldwide perspective. Ann. Rev. Entomol. 55: 521–546.

Hall, L. 2018. Understanding the sensory basis for *Anoplophora glabripennis* pheromone-mediated behavior. Ph.D. dissertation, Pennsylvania State University, State College, PA.

Hu, J. F., S. Angeli, S. Schuetz, Y. Q. Luo, and A. E. Hajek. 2009. Ecology and management of exotic and endemic Asian longhorned beetle *Anoplophora glabripennis*. Agr. Forest. Entomol. 11: 359–375.

Hull-Sanders, H., E. Pepper, K. Davis, and R. T. Trotter. 2017. Description of an establishment event by the invasive Asian longhorned beetle (*Anoplophora glabripennis*) in a suburban landscape in the northeastern United States. PLoS One 12: e0181655.

Javal, M., G. Roux, A. Roques, and D. Sauvard. 2018. Asian long-horned beetle dispersal potential estimated in computer-linked flight mills. J. Appl. Entomol. 142: 282–286.

Keena, M. A. 2005. Pourable artificial diet for rearing *Anoplophora glabripennis* (Coleoptera: Cerambycidae) and methods to optimize larval survival and synchronize development. Ann. Entomol. Soc. Am. 98: 536–547.

Keena, M. A. 2006. Effects of temperature on *Anoplophora glabripennis* (Coleoptera: Cerambycidae) adult survival, reproduction, and egg hatch. Environ. Entomol. 35: 912–921.

Keena, M., and V. Sánchez. 2018a. Reproductive behaviors of *Anoplophora glabripennis* (Coleoptera: Cerambycidae) in the laboratory. J. Econ. Entomol. 111: 620–628.

Keena, M., and V. Sánchez. 2018b. Inter- and intra- sexual interactions in *Anoplophora glabripennis* (Coleoptera: Cerambycidae) and the impact of different sex ratios. J. Econ. Entomol. 47: 1233–1241.

Lingafelter, S. W., and E. R. Hoebeke. 2002. Revision of the genus *Anoplophora* (Coleoptera: Cerambycidae). Entomological Society of Washington, Washington, D.C.

Lopez, V. M., M. S. Hoddle, J. A. Francese, D. R. Lance, and A. M. Ray. 2017. Assessing flight potential of the invasive Asian longhorned beetle (Coleoptera: Cerambycidae) with computerized flight mills. J. Econ. Entomol. 110: 1070–1077.

McNemar, Q. 1947. Note on the sampling error of the difference between correlated proportions or percentages. Psychometrika 12: 153–157

Meng, P. S., K. Hoover, and M. A. Keena. 2015. Asian longhorned beetle (Coleoptera: Cerambycidae), an introduced pest of maple and other hardwood trees in North America and Europe. J. Integr. Pest. Manag. 6: 4.

Nehme, M. E., R. T. Trotter, M. A. Keena, C. McFarland, J. Coop, H. M. Hull-Sanders, P. Meng, C. M. De Moraes, M. C. Mescher, and K. Hoover. 2014. Development and evaluation of a trapping system for *Anoplophora glabripennis* (Coleoptera: Cerambycidae) in the United States. Environ. Entomol. 43: 1034–1044.

Ridgel, A. L., and R. E. Ritzmann. 2005. Insights into age-related locomotor declines from studies of insects. Ageing Res. Rev. 4: 23–39.

Sánchez, V., and M. A. Keena. 2013. Development of the teneral adult *Anoplophora glabripennis* (Coleoptera: Cerambycidae): time to initiate and completely bore out of maple wood. Environ. Entomol. 42: 1–6.

SAS Institute. 2015. SAS/STAT user's guide, version 9.4. SAS Institute, Cary, NC.

Sawyer, A., W. S. Panagakos, A. E. Horner, and K. J. Freeman. 2011. Asian longhorned beetle, over the river and through the woods: habitat-dependent population spread, pp. 52–54. *In* K. A. McManus and K. W. Gottschalk (eds.), Proceedings, 21st U.S. Department of Agriculture Interagency Research Forum on Invasive Species 2010, 12–15 January 2010, Annapolis, MD. Gen. Tech. Rep. NRS-P-75. U.S. Department of Agriculture, Forest Service, Northern Research Station, Newtown Square, PA.

Smith, M. T., J. Bancroft, G. H. Li, R. Gao, and S. Teale. 2001. Dispersal of *Anoplophora glabripennis* (Cerambycidae). Environ. Entomol. 30: 1036–1040.

Smith, M. T., P. C. Tobin, J. Bancroft, G. H. Li, and R. T. Gao. 2004. Dispersal and spatiotemporal dynamics of Asian longhorned beetle (Coleoptera: Cerambycidae) in China. Environ. Entomol. 33: 435–442.

Statistix. 2013. Statistix for Windows user's manual version 10. Analytical Software, Tallahassee, FL.

Steyn, V. M., K. A. Mitchell, and J. S. Terblanche. 2016. Dispersal propensity, but not flight performance, explains variation in dispersal ability. Proc. Royal Soc. Lond. B 283: 20160905.

Straw, N. A., N. J. Fielding, C. Tilbury, D. T. Williams, and T. Cull. 2016. History and development of an isolated outbreak of Asian longhorn beetle *Anoplophora glabripennis* (Coleoptera: Cerambycidae) in southern England. Agr. Forest. Entomol. 18: 280–293.

Trotter, R. T., and H. M. Hull-Sanders. 2015. Quantifying dispersal of the Asian longhorned beetle (*Anoplophora glabripennis*, Coleoptera) with incomplete data and behavioral knowledge. Biol. Invasions 17: 3359–3369.

Trotter, R. T., and M. A. Keena. 2016. A variable-instar climate-driven individual beetle-based phenology model for the invasive Asian longhorned beetle (Coleoptera: Cerambycidae). Environ. Entomol. 45: 1360–1370.

Turgeon, J. J., M. Orr, C. Grant, Y. Wu, and B. Gasman. 2015. Decade-old satellite infestation of *Anoplophora glabripennis* Motschulsky (Coleoptera: Cerambycidae) found in Ontario, Canada outside regulated area of founder population. Coleopts. Bull. 69: 674–678.

Van Dyck, H., and M. Baguette. 2005. Dispersal behaviour in fragmented landscapes: routine or special movements? Basic Appl. Ecol. 6: 535–545.

Wen, J., Y. Li, N. Xia, and Y. Luo. 1998. Dispersal pattern of adult *Anoplophora glabripennis* on poplars. Acta Ecol. Sin. 18: 269–277.

Williams, D. W., G. H. Li, and R. T. Gao. 2004. Tracking movements of individual *Anoplophora glabripennis* (Coleoptera: Cerambycidae) adults: application of harmonic radar. Environ. Entomol. 33: 644–649.

Zhou, J., K. Zhang, and Y. Lu. 1984. Study on adult activity and behavioral mechanism of *Anoplophora nobilis* Ganglbauer. Sci. Silvae Sin. 20: 372–379.