

Host Selection and Feeding Preference of *Agrilus planipennis* (Coleoptera: Buprestidae) on Ash (*Fraxinus* spp.)

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ABSTRACT We studied the host selection behavior and feeding preference of the emerald ash borer, *Agrilus planipennis* Fairmaire (Coleoptera: Buprestidae). *A. planipennis* is an exotic forest insect pest native to Asia that was discovered in North America in 2002 and is causing widespread mortality of ash trees (*Fraxinus* spp.) in southeast Michigan and surrounding states. We compared host selection and feeding behavior on different species of ash including Manchurian ash (*F. mandshurica* Rupr.), green ash (*F. pennsylvanica* Marsh), white ash (*F. americana* L.), black ash (*F. nigra* Marsh), blue ash (*F. quadrangulata* Michx.), and European ash (*F. excelsior* L.). Manchurian ash is native to Asia, whereas the other species (native to North America or Europe) represent novel hosts for *A. planipennis*. Beetles distributed themselves more frequently and fed to a greater extent on green, black, and white ash compared with blue, European, and Manchurian ash. Although beetles consumed every ash species offered to them, Manchurian ash and blue ash were least preferred in feeding bioassays. When we analyzed the volatile content of intact and girdled ash for quantitative variation in 11 compounds that elicited antennal activity, we found that the overall volatile profiles of the six ash species differed significantly in their relative amounts of antennally active compounds. Green ash has lower relative amounts of volatiles compared with Manchurian ash, which might render it more attractive and less resistant to *A. planipennis*. Lower tolerance and resistance of green ash might make it more susceptible to mortality compared with Manchurian ash, which coevolved with the beetle in its native range. Repellent odors, potential antifeedants, and genes for resistance in Manchurian ash could be explored for methods to manage *A. planipennis* populations.

KEY WORDS native versus exotic hosts, olfactory cues, host volatiles

The emerald ash borer, *Agrilus planipennis* Fairmaire (Coleoptera: Buprestidae), is a recently established invasive pest of ash (*Fraxinus* spp.) in North America (Haack et al. 2002, Poland and McCullough 2006). Since its discovery in 2002, it is estimated that >40 million ash trees have been killed in Michigan, and tens of millions in eight other states, as well as in Ontario and Quebec, Canada (Poland and McCullough 2006, EAB Info 2009, CFIA 2009). In its native range (China, Japan, and Korea), the beetle sporadically infests Chinese ash, *F. chinensis* Roxb., and Manchurian ash, *F. mandshurica* Rupr., but does not pose a threat to natural *Fraxinus* stands or plantations (Wei et al. 2004). *A. planipennis* feeds and breeds exclusively on *Fraxinus* spp. (Cappaert et al. 2005, Anulewicz 2006). Adults consume leaf margins throughout their lifetime and must feed for ~2 wk before they mate and lay eggs in the bark (Bauer et al. 2004). In outbreaks,

extensive larval feeding within the phloem and cambium of branches and tree trunks disrupts translocation of water and nutrients, resulting in tree mortality (Poland and McCullough 2006).

There is evidence that *A. planipennis* adults use both visual and olfactory cues in host location. When box traps with different colored panels were suspended among ash trees, *A. planipennis* preferred to land on purple panels compared with black, green, blue, red, silver, white, or yellow, suggesting that visual cues are involved in host or mate finding (Francese et al. 2005). Gas chromatographic-electroantennographic detection analyses (GC-EAD) of volatiles from Manchurian ash showed 16 compounds that elicited antennal responses in both sexes (Rodriguez-Saona et al. 2006). In an olfactometer bioassay, female beetles were attracted to plants that were damaged by feeding or treated with methyl jasmonate (a signal of physiological stress), indicating that olfactory cues might be used to locate host trees under stress (Rodriguez-Saona et al. 2006). Volatiles from ash leaves (de Groot et al. 2008) and bark (Crook et al. 2008) elicit antennal responses by *A. planipennis* and are attractive in the field.

We tested the relative preference of adult *A. planipennis* to foliage from a host in its native range, *F.*

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mandshurica, compared with four North American hosts, *F. pennsylvanica*, *F. americana*, *F. nigra*, blue ash (*F. quadrangulata* Michx.), and one European species, European ash (*F. excelsior* L.), by quantifying beetle distribution and feeding choices on leaves of the six species. We analyzed the volatile profiles of green, black, white, blue, European, and Manchurian ash species for differences that could account for their preferences as hosts in nature. We collected volatiles by aerating foliage and quantified 11 antennally active compounds (Rodriguez-Saona et al. 2006) by GC to determine whether there are differences among the six species that could render North American ash species more preferred than hosts native to Asia.

Materials and Methods

Treatment of Beetles. Green ash trees from Ingham County, MI, that were infested with *A. planipennis* were felled, bucked into 90-cm-long bolts, and stored at 4°C. As beetles were required for experiments, they were allowed to emerge in rearing tubes at 25°C. They were separated by sex and kept in 295-ml plastic beverage containers with an evergreen ash, *F. uhdei* (Wenzig) Lingesh, leaf in a vial of water for feeding. Evergreen ash occurs in the western United States, and *A. planipennis* has not been exposed to it yet in nature. In this way, beetles were fed an ash species that was not used in experiments to avoid possible confounding effects of preconditioning. Containers with beetles were stored in growth chambers at 25°C and >70% RH with fresh food that was replaced twice a week, until they were used in experiments.

Treatment of Plants for Feeding Experiments and Aerations. Two- to 4-yr-old nursery seedlings of the six ash species ranging from 15 to 30 cm in height were stored at 3–4°C until they were planted. In late spring, seedlings were planted in 18 cm long by 18 cm wide by 30 cm tall pots with hi-porosity soil mix (Baccto; Michigan Peat, Houston, TX) and fertilized weekly with a 20–20–20 N-P-K Scotts Peters Professional water-soluble fertilizer (Marysville, OH). They were grown in a greenhouse at ≈25°C, under natural light augmented with 400-W high-pressure sodium lamps. After seedlings flushed and foliage expanded, leaves of approximately the same age from each species were clipped for use in feeding trials. Intact plants from which leaves were not clipped were used for aerations.

Six-Choice Feeding Assay. To determine host selection and feeding preference of *A. planipennis* on six species of *Fraxinus*, we released beetles that were ≈5 d old into metal screen cages (60 by 60 by 60 cm) containing one leaf of each species. Thirty beetles were released into each cage and left there for 48 h. Four cages (two for males and two for females) were set up at any given time, and the experiment was replicated seven times over a period of 7 wk, resulting in 14 replicates for each sex. Foliage was cut from 2- to 4-yr-old greenhouse saplings. Fifteen saplings of each species were used for the study, and care was taken to see that leaves from the same plant were not used more than once, thus avoiding pseudo-replica-

tion. Leaves were placed individually in glass vials containing water and arranged in a row in random order. Because we had 14 replicates (i.e., leaves from 14 different trees of each species) in the study, every treatment was placed in every position in the cage during replication, ensuring that there was no bias because of leaf position in the cage. We had to replicate the experiment over time because of limited availability of emerging beetles and flushed foliage at any particular time. Because the experiment was set up in complete blocks over time, there was no bias resulting from season.

The number of beetles present on the foliage of each species was recorded every 2 h during the day for a 48-h period to assess host selection. Beetles are most active during daytime, particularly in the sun (Rodriguez-Saona et al. 2007), so we performed observations only in the daytime. At night, beetles tend to rest on the walls of the cage. A total of nine daytime observations of beetle distribution were made over 48 h for every replicate. Leaves were scanned before and after feeding (Winfolia software; Regent Instruments 2003) to determine leaf area available to beetles and the amount consumed. Males and females were tested in separate trials, with 14 replicates per sex. We ensured that beetles were well fed before the experiment to enable them to make a satiated choice and avoid ravenous feeding on the first species they encountered.

Collection of Plant Volatiles. Headspace volatiles from entire plants were collected using a push-pull aeration system (Rodriguez-Saona et al. 2001). The stem, including branches and leaves ≈ 20 cm from the soil, were placed inside a glass cylinder (4 liters, 30.5 cm high, 15.2 cm diameter) with a guillotine base (Analytical Research Systems, Gainesville, FL) that closed the cylinder around the stem. Air was generated from a portable 2.0-HP, 20-gal Westward air compressor (Grainger, Northbrook, IL) and pushed through activated charcoal into the top of the glass cylinder and then drawn over the plant at 2 liters/min. Emitted volatiles were captured in adsorbent Super-Q traps (Alltech, Deerfield, IL), 5 cm from the base of the cylinder, by pulling half the air at 1 liter/min with a Gast 1/4-HP vacuum pump (Grainger). The rest of the air was vented out of the bottom of the system through an opening around the stem of the plant that was plugged with cotton. Volatiles were collected at 25 ± 0.5°C and 40% humidity for 10 h during the day.

After volatiles were collected from healthy plants, the stem of each plant was girdled with a razor blade by removing bark and phloem tissue from a 2-cm section 10 cm above the soil. Girdled seedlings were held for 24 h in an attempt to induce plant defenses and possibly stress volatiles. Plants were aerated again as described above. The aerated portion was clipped, and wet and dry weights of aerated tissue were recorded.

GC. Collected volatiles were extracted from the Super-Q adsorbent by flushing with 150 µl of pentane-hexane mixture containing 3.5 ng/µl of heptyl acetate (internal standard) with a stream of nitrogen gas. Two microliters of each sample was injected into a Varian

3400 GC equipped with a flame ionization detector (FID) and a 25-m- by 0.20-mm-ID, 0.33- μ m-thick HP1 column using splitless injections and helium as the carrier gas. The oven temperature began at 60°C for 1 min and increased at 10°C/min to 190°C and then at 35°C/min to 300°C and held for 5 min. Compounds previously found to elicit antennal activity in GC-EAD analyses, hexanal, (*E*)-2-hexenal, (*Z*)-3-hexenol, (*E*)-2-hexenol, butoxyethanol, (*Z*)-3-hexenyl acetate, hexyl acetate, (*E*)- β -ocimene, nonanal/linalool (co-elute on this column), nonatriene and (*Z,E*)- α -farnesene (Rodriguez-Saona et al. 2006), were quantified based on comparison of peak areas with that of the internal standard. Identities of compounds were confirmed by running selected samples on a Hewlett-Packard model 5989 GC-mass spectrometer (GC-MS) and a synthetic blend containing compounds of interest on the gas chromatograph. The temperature program was 40°C for 1 min and then increased at 8°C/min to 300°C. The amount of each compound of interest was calculated in nanograms per gram dry weight of tissue.

Statistical Analyses. All data were assessed for normality and homoscedasticity. For the six-choice feeding assay, the number of beetles observed on each species over a 48-h period was transformed by $\log_{10}(x + 1)$ and analyzed by repeated-measures analysis of variance (ANOVA; PROC GLM) followed by the Ryan-Einot-Gabriel-Welsch (REGW) multiple comparisons procedure (Day and Quinn 1989) with species as the main effect. The size of leaves used in bioassays, the total foliage area consumed, and the arcsine square root transformed proportion of foliage consumed for each species were analyzed by multivariate ANOVA with leaf area as a covariate (PROC GLM) followed by the REGW multiple comparisons procedure. We also performed the following orthogonal contrasts to examine differences in the proportion of foliage consumed by males and females between the following groups of ash species: (1) exotic hosts (green, black, white, blue, and European ash versus native host (Manchurian ash), (2) green, black, white, and European ash versus blue ash, (3) blue ash versus Manchurian ash, and (4) green and white ash versus black ash.

Data from volatile collections were also $\log_{10}(x + 1)$ transformed and analyzed by multivariate ANOVA (PROC MANOVA) to determine differences in the amounts of 11 compounds of interest among the six species. Principal components analysis (PROC PRINCOMP) was performed to summarize multivariate data in two dimensions and graphically determine whether clustering of species occurred based on their overall volatile profiles. Correlation coefficients between the amounts of compounds and the first two principal components were calculated to determine which compounds contribute to the separation of the overall volatile profiles among species. Sequential Bonferroni adjustments were performed to account for the number of tests conducted (Rice 1989).

We then examined the relationship between host preference by *A. planipennis* and volatiles emitted by

ash species by plotting indices of host preference and volatile emission for each species using our empirical data. We performed principal components analyses on data from the landing and feeding experiments, as well as on the volatile profiles of the six ash species, and used the loadings on first principal component to calculate a host preference index and a volatile emission index as described below.

$$\text{Host preference index} = \sum_{i=1}^n PC1(L, F)$$

where *PC1* represents the loadings on the first principal component that account for 99% of the variation in beetle distribution and feeding, *L* is the average number of beetles that were distributed on each of the six species of ash over the course of the experiment, and *F* is the arcsine transformed proportions of foliage of each species that was consumed during the experiment.

$$\text{Volatile emission index} = \sum_{i=1}^n PC1(V)$$

where *PC1* represents the loadings on the first principal component that account for 39% of the variation among the 11 compounds we quantified, and *V* is the amount in nanograms of 11 volatiles emitted by the six ash species that were intact.

All analyses were performed using SAS Institute version 9.1 (SAS Institute 2002–2003) statistical software and $\alpha = 0.05$.

Results

Six-Choice Feeding Assay. Our bioassays could not control for leaf size because saplings of different species were obtained from different nurseries and varied in size. Our experiment was replicated over the season as saplings continued to grow. Hence, there were differences in leaf sizes of the six ash species used in our experiments. *A. planipennis* distributed and fed preferentially on three of the North American hosts (green, black, and white ash) compared with blue, European, and Manchurian ash (Figs. 1 and 2). There was a significant effect of species (repeated-measures ANOVA males: $F_{5,78} = 7.7$, $P < 0.0001$; females: $F_{5,78} = 3.4$, $P < 0.008$) and time (MANOVA males and females: Wilk's $\lambda = 0.62$, $F_{8,71} = 5.3$, $P < 0.0001$) on the number of beetles observed on foliage, but no species $\times$ time interaction in either sex (males: Wilk's $\lambda = 0.6$, $F_{40, 312} = 0.9$, $P = 0.55$; females: Wilk's $\lambda = 0.7$, $F_{40, 312} = 0.66$, $P = 0.94$). Beetles became more discriminatory as time progressed. There was no difference among species during the first three observations (conducted at 20-min intervals), but after the fourth observation, beetles appeared to discriminate among species by spending more time on green, black, and white ash compared with blue and Manchurian ash (Fig. 1).

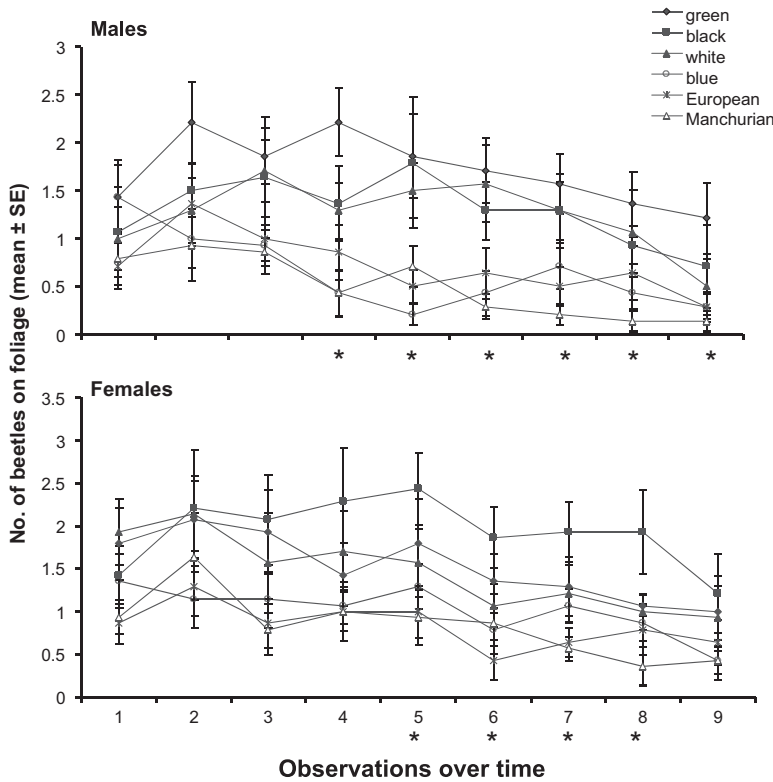


Fig. 1. Number of beetles observed on foliage during daytime hours over a 48-h period. Results of a repeated-measures ANOVA ($n = 14$). Asterisks below x-axis indicate time periods during which the number of beetles observed on foliage differed significantly based on REGW multiple comparisons tests.

There was a significant species effect on the total amount as well as proportion of foliage consumed for both sexes (MANOVA males: Wilk's $\lambda = 0.41$, $F_{15, 207} = 5.3$, $P < 0.0001$; females: Wilk's $\lambda = 0.41$, $F_{15, 207} = 5.2$, $P < 0.0001$; Fig. 2). Leaf area, when examined as a covariate, had no effect on males as far as the amount ($F_{1,77} = 0.28$, $P = 0.6$) or proportion of foliage consumed ($F_{1,77} = 0.58$, $P = 0.4$) were concerned. However, there was a significant effect of leaf area on the amount ($F_{1,77} = 28.4$, $P < 0.0001$) but not proportion consumed ($F_{1,77} = 0.02$, $P = 0.9$) by females. The total area of green and white ash foliage consumed by males exceeded that of any other species (Fig. 2a). The proportion consumed did not differ among green, black, or white or among blue, European, and Manchurian (Fig. 2b). Females consumed significantly more green, black, and white ash than blue or Manchurian ash (Fig. 2c). The proportion of foliage consumed was also not significantly different among green, black, white, and European ash or between blue and Manchurian ash (Fig. 2d). Both sexes consumed less blue and Manchurian ash than any other species. The above results are further confirmed by orthogonal contrasts comparing specific groups of ash species (Table 1).

Volatile Profiles of *Fraxinus* spp. Multivariate ANOVA indicated that the six species differed significantly in the total complement of volatiles that we

quantified (healthy: Wilk's $\lambda = 0.0023$, $F_{55,161} = 7.91$, $P < 0.0001$; girdled: Wilk's $\lambda = 0.0036$, $F_{55,161} = 6.93$, $P < 0.0001$; Fig. 3). In both healthy and girdled trees, green ash had relatively low amounts of volatiles (indicated by low factor loadings on the PC1 axis), whereas Manchurian ash had high amounts (indicated by relatively higher factor loadings on the PC1 axis) compared with the other species (Fig. 3).

Except for some overlap, principal components analysis on the entire quantifiable volatile profile in healthy and girdled trees showed separation of the six species based on relative differences in their volatile profiles (Fig. 3). For healthy trees, PC1 distinguished among species containing high amounts of all compounds except hexanal, (Z)-3-hexenol, and (E)-2-hexenol. PC2 separated species containing high amounts of hexanal, (Z)-3-hexenol, (E)-2-hexenol, butoxyethanol, (Z)-3-hexenyl acetate, and nonanal/linalool and low amounts of (Z,E)- α -farnesene. In girdled trees, PC1 distinguished among species containing high amounts of all compounds except hexanal. PC2 separated out species containing low amounts of hexanal, (E)-2-hexenol, and (Z,E)- α -farnesene and high amounts of (Z)-3-hexenyl acetate (Table 2).

Relationship Between Host Preference by *A. platanipennis* and Volatile Emission by *Fraxinus* spp. When we plotted the indices of host preference and volatile emission for the six ash species using the results of the

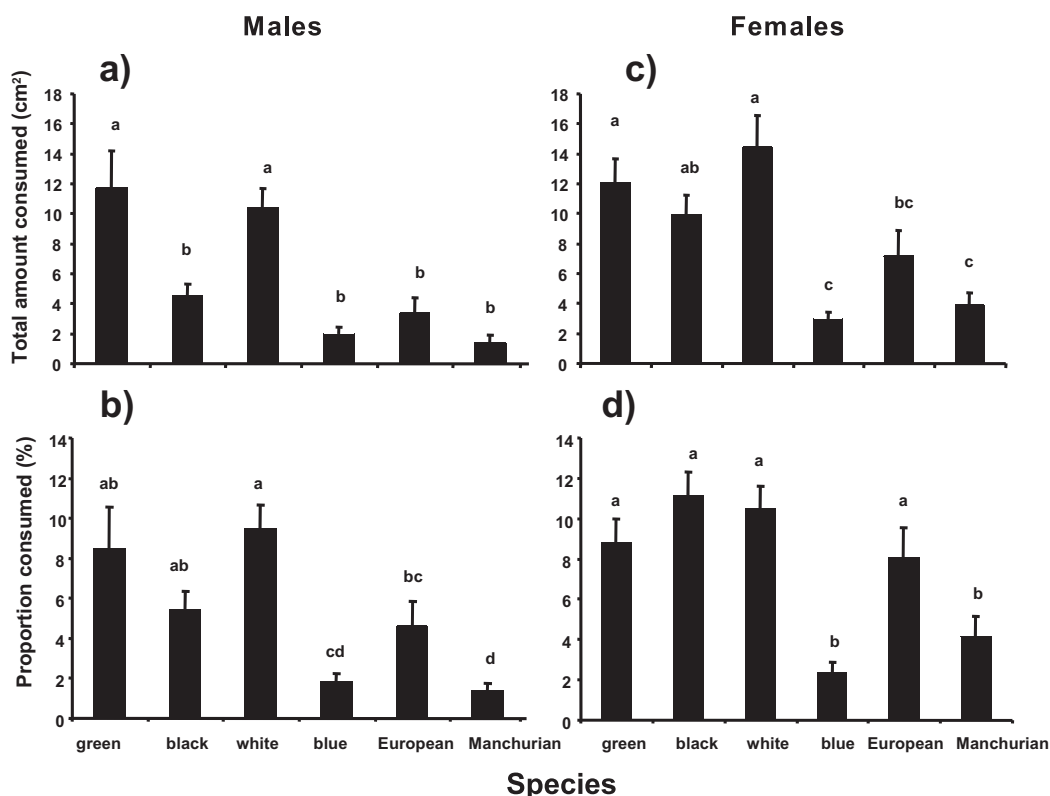


Fig. 2. Total amount (a, c) and proportion (b, d) of foliage consumed ($n = 14$) by *A. planipennis* in a choice bioassay among six species of *Fraxinus*. Bars with the same letter are not significantly different, REGW-multiple comparisons test, $P < 0.05$.

principal components analysis, it became apparent that for the most preferred species (green ash) and least preferred species (Manchurian ash), there is an inverse relationship between host preference and volatile emission (Fig. 4). Green ash, which has the lowest quantity of emitted volatiles is the most preferred, whereas Manchurian ash, with the highest quantity, is the least preferred for feeding. The relationship is not as pronounced for the four other species, whose indices of volatile emission and host preference were intermediate between green and Manchurian ash.

Discussion

Despite the fact that Manchurian ash is a native host of *A. planipennis*, it was clearly the least preferred by both sexes in terms of the feeding choices and the distribution of beetles among the species tested (Figs. 2a–d). Initially, beetles distributed themselves in equal numbers on all species and proceeded to leave blue and Manchurian ash for the more preferred green, black, and white ash (Fig. 1). In another study, females preferred to oviposit on ash species compared with nonash species and chose green and white ash over blue ash (Anulewicz 2006). Similarly, green ash logs had more galleries than white ash, and blue ash had fewer galleries than other ash species, although larvae did successfully complete development on blue ash (Anulewicz et al. 2008). Female host preferences can be correlated with larval performance on hosts (Jaenike 1990, West and Cunningham 2002), and there might be fitness benefits to feeding and ovipositing on less resistant hosts in a new geographic range.

It is estimated that *A. planipennis* was established in the United States for at least 10 yr before its discovery there in 2002 (Cappaert et al. 2005), and it has killed >40 million green, white, and black ash trees (EAB Info 2008). Its preference for and rapid success on North American ashes could result from its ability to

Table 1. Differences in proportion of foliage consumed by *A. planipennis* between the following groups of ash species

Comparisons	Sex	F-value	P value
Exotic hosts (green, black, white, blue, and European ash) versus native host (Manchurian ash)	Males	13.86	0.0004
	Females	13.46	0.0005
Green, black, white and European ash versus blue ash	Males	44.39	<0.0001
	Females	44.10	<0.0001
Blue ash versus Manchurian ash	Males	1.77	0.19
	Females	1.85	0.18
Green and white ash versus black ash	Males	0.87	0.35
	Females	1.03	0.34

Results of orthogonal contrasts ($N = 14$ replicates per host species and sex).

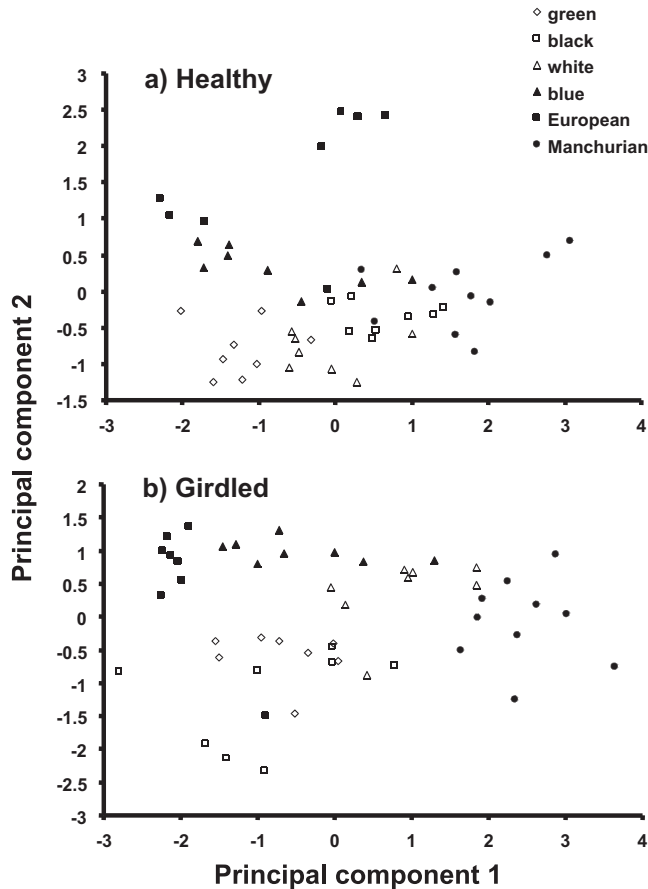


Fig. 3. Principal components analysis of emitted volatiles from healthy and girdled saplings of six species of *Fraxinus*. Volatiles elicited antennal responses from *A. planipennis* in electrophysiological assays (Rodriguez-Saona et al. 2006).

overcome the defenses of novel hosts in a new habitat more easily than the co-evolved defenses in native hosts or from the presence of anti-feedants in ash species with which it has coevolved. Of the North American ashes, blue ash was the least preferred, and the number of *A. planipennis* that were observed on and that fed on blue ash was similar to Manchurian ash (Figs. 2a–d; Table 1). Blue ash was also less preferred than white or green ash for oviposition (Anulewicz 2006, Anulewicz et al. 2008), suggesting that it might have physical or chemical properties that render it less suitable for adult feeding and larval development.

A possible hypothesis to explain the low amount of feeding on Manchurian ash is that this species might possess higher nutritive value than green ash; thus, lower consumption would be required for beetles to achieve similar fitness. Alternatively, Manchurian ash might contain compounds that limit beetle consumption. Comparison of crude phloem extracts of Manchurian ash versus white and green ash showed hydroxycoumarins, phenylethanoids, and calceolariosides unique to Manchurian ash that could confer resistance to herbivory (Eyles et al. 2007). If the alternate hypothesis is true and if larval feeding responsible for tree mortality follows a pattern similar to that of adult feeding, it may explain in

part why Manchurian ash is not as susceptible to herbivory as North American ash species.

In studies of resistance of ash species to attack by *A. planipennis*, Rebek et al. (2006, 2008) found that green and white ash had much higher rates of colonization and mortality than Manchurian ash, which yielded far fewer beetles than North American ashes. It seems that lower relative amounts of volatiles are emitted from green ash than Manchurian ash (Figs. 3 and 4), which may indicate lower host resistance, making green ash more attractive to beetles. When indices of host preference were plotted against ash volatile profiles, green ash, with the lowest volatile emission profile, had the highest host preference index, whereas the reverse was true for Manchurian ash (Fig. 4). It seems somewhat paradoxical that a host species with the highest volatile emission profile is the least preferred, particularly when host selection by the beetle seems to be mediated at least in part by the perception of emitted host volatiles. (*Z*)-3-hexenol, one of the components of the attractive green leaf volatile blend that stimulated beetle antennae in electrophysiological studies (Rodriguez-Saona et al. 2006; de Groot et al. 2008), increased trap catches significantly in the field at a low dose of 48 mg/d but not at a high dose

Table 2. Eigenvectors and correlation coefficients of compounds in foliage from healthy and girdled ash saplings with the first two principal components

State	Compound	Principal component 1		Principal component 2	
		Eigenvector	Correlation coefficient	Eigenvector	Correlation coefficient
Healthy	hexanal	0.13	0.28	0.4569	0.69 ^b
	(E)-2-hexenal	0.23	0.52 ^b	0.1532	0.24
	(Z)-3-hexenol	−0.01	−0.03	0.3392	0.71 ^b
	(E)-2-hexenol	0.06	0.26	0.1283	0.37 ^a
	butoxyethanol	0.16	0.38 ^a	0.4198	0.73 ^b
	(Z)-3-hexenyl acetate	0.25	0.48 ^a	0.3907	0.53 ^b
	hexyl acetate	0.34	0.63 ^b	0.0375	0.05
	(E)-β-ocimene	0.29	0.68 ^b	0.0690	0.12
	nonanal/linalool	0.18	0.47 ^a	0.3133	0.59 ^b
	nonatriene	0.36	0.70 ^b	−0.1876	−0.26
	(Z,E)-α-farnesene	0.69	0.87 ^b	−0.4097	−0.36 ^a
	Percent total variance	39.35		19.89	
Girdled	hexanal	−0.05	−0.14	−0.19	−0.29 ^a
	(E)-2-hexenal	0.10	0.31 ^a	−0.25	−0.45 ^a
	(Z)-3-hexenol	0.14	0.46 ^a	0.02	0.03
	(E)-2-hexenol	0.18	0.64 ^b	−0.02	−0.04
	butoxyethanol	0.05	0.37 ^a	−0.02	−0.08
	(Z)-3-hexenyl acetate	0.23	0.44 ^a	0.73	0.80 ^b
	hexyl acetate	0.28	0.77 ^b	0.15	0.23
	(E)-β-ocimene	0.45	0.91 ^b	0.05	0.06
	nonanal/linalool	0.17	0.55 ^b	−0.02	−0.04
	nonatriene	0.48	0.84 ^b	0.23	0.23
	(Z,E)-α-farnesene	0.59	0.85 ^b	−0.54	−0.45 ^a
	Percent total variance	51.12		16.56	

Sequential Bonferroni-adjusted $\alpha = 0.05$. Statistical significance indicates compounds that are responsible for quantitative variation in volatile profiles among six species of ash.

^a <0.05.
^b <0.01.
^c <0.001.

of 330 mg/d. Similar inverse dose-dependent responses have been seen in other phytophagous beetles (Coleoptera: Curculionidae: Scolytinae). Catches of *Ips latidens* (LeConte) was significantly lower in traps with high doses of 3-carene, and both *I. latidens* and *Ips pini* (Say) responded in lower numbers to high doses of myrcene (Miller and Borden 2000). The inverse dose-responses of beetles to behaviorally active com-

pounds suggest that host preference reflects an assessment by the beetle of both host quality and suitability for larval development. The fact that fewer beetles were observed on Manchurian ash as time progressed in the six-choice assay showed that *A. planipennis* can discriminate among different ash species and that certain exotic ashes are more attractive and palatable than its native host (Fig. 1).

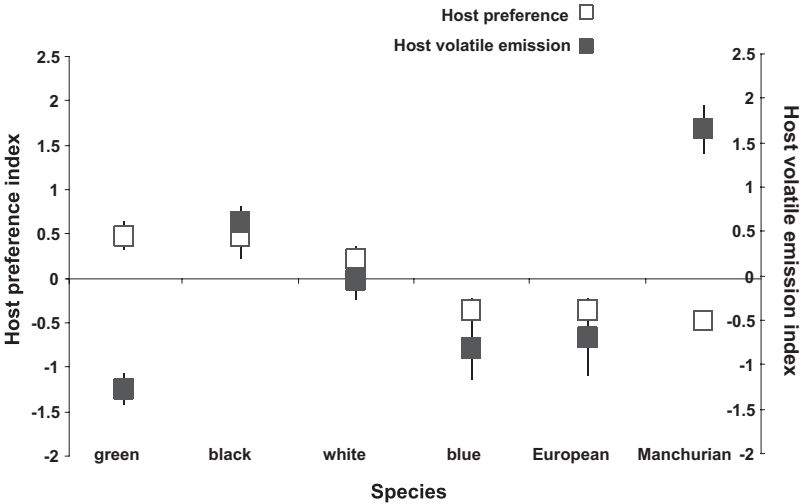


Fig. 4. Plot of indices (mean $\pm$ SE) after principal components analyses of ash volatile emission profile and host selection (landing and feeding) by the emerald ash borer for six species of ash.

It is not uncommon for related tree species to differ in the relative amounts of constitutive or emitted volatiles (Holm and Hiltunen 1997, Faldt et al. 2001, Pureswaran et al. 2004) and for herbivorous insects to discriminate among hosts based on these differences (Abe and Matsuda 2005, Pureswaran and Borden 2005). In another study, stressed Manchurian ash saplings released relatively higher amounts of volatiles when they were subjected to feeding by beetles or sprayed with methyl jasmonate, a stress-induced phytohormone compared with control trees (Rodriguez-Saona et al. 2006). In nature, mature trees that were girdled or treated with herbicide were more attractive to beetles than healthy controls (Cappaert et al. 2005), suggesting that there are attractive semiochemical factors in mature trees under stress that warrant study. Lower relative amounts of emitted volatiles from green ash may make it more attractive to beetles than Manchurian ash, because it may indicate lower host resistance.

Host range may be a versatile feature of plant-insect associations especially in cases where exotic hosts are congeners of native hosts (Janz et al. 2001). Members of the genus *Fraxinus*, even when geographically separated, are phylogenetically closely related (Waller 2001). It seems that it was particularly favorable for *A. planipennis* that the new host species in North America is a closely related congener of its native host, because adult survivorship did not differ when fed either on green or Manchurian ash (unpublished data). The current outbreak of *A. planipennis* in North America and its preference for and dramatic success on ash species evolutionarily novel to it raise questions on the ecological interactions that surround biological invasions resulting from host switching. Manchurian ash seems to be more resistant (possesses physical or chemical traits that lower beetle fitness) and/or more tolerant (able to survive and reproduce after combating adverse effects of an attack) to herbivory by *A. planipennis* than green, black, and white ash.

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