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RESEARCH ARTICLE

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Divergent host plant utilization by adults and offspring is related to intra-plant variation in chemical defences

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Abstract

1. Adult and juvenile herbivores of the same species can use divergent feeding strategies, and thus may inhabit and consume different parts of the plant. Because the expression of chemical defences often differs between host plant tissues, this variation may result in disparate performance outcomes for adult and juvenile conspecifics that feed on distinct dietary substrates.
2. The goal of this study was to evaluate how host range may differ between adults and juveniles in a generalist herbivore. We addressed the impacts of among- and within-plant defence variation using the wood-feeding Asian longhorned beetle (*Anoplophora glabripennis*) and three host plants having a range of putative resistance.
3. Impacts of host plants on adult and offspring performance were assessed using a series of controlled bioassays. We evaluated adult-feeding and egg-laying behaviours in choice and no-choice experiments using the different hosts, and subsequent offspring establishment. We then evaluated host plant chemical composition related to nutrition and defence.
4. Different plants had strong impacts on adult performance, but these patterns did not extend to effects on offspring. Females were capable of developing eggs when provided *Acer rubrum*, but not *Populus deltoides* or *Populus tomentosa*. Females that produced eggs by feeding on *A. rubrum*, however, deposited eggs into all three plant species. Larvae hatched and consumed tissues in all three hosts. The differences between adult and juvenile utilization of *Populus* spp. were reflected in markedly higher salicinoid phenolic concentrations in bark (>2% dw), while wood had trace quantities.
5. Our results demonstrate that plant resistance mechanisms can differentially act upon adult and juvenile life stages of a polyphagous herbivore when there is differential expression of chemical defences among plant tissue types. *Anoplophora glabripennis* has been a globally successful invader due in part to its broad host range, and our results suggest a mechanism that permits the beetle to exploit marginally resistant plants. This study has implications for how host range differs between insect feeding stages, which is particularly important for invasive, polyphagous species encountering novel food sources.

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KEYWORDS
Asian longhorned beetle, chemical defences, invasive, maple, phenolic glycosides, poplar, salicinoids, wood borer

1 | INTRODUCTION

Plant defences are often differentially expressed among tissues, impacting how resources are utilized by herbivores (Agrawal & Fishbein, 2006; Farrell, Mitter, & Futuyma, 1992; Futuyma & Agrawal, 2009). Expression of defences can be influenced by numerous environmental and physiological mechanisms, and vary across different phenological windows (Aide, 1993; Keith & Mitchell-Olds, 2017; Kursar & Coley, 2003; Meyer & Paul, 1992; Wurst, Van Dam, Monroy, Biere, & Van Der Putten, 2008). Different herbivore species are capable of co-occurring in populations through inter- and intraspecific spatial and successional segregation that ultimately reduces competition (Amarasekare, 2003). However, the partitioning of dietary resources through space and time may also present herbivores with different levels and types of plant defences. Presumably, variation in plant defence expression, at least in part, aims to target herbivores that pose as reproductive or lethal threats (McCall & Fordyce, 2010).

The impacts of intra-plant variation in defence expression may be particularly important for herbivorous beetle species where conspecifics utilize different plant tissues (e.g. foliage, roots, wood). On one hand, this may facilitate host plant utilization by reducing intraspecific competition between herbivore adults and offspring, providing greater resources for the population. However, by exploiting different tissues, adult and larval conspecifics can encounter different concentrations of plant defences and nutrients, and in some cases, plants might produce asymmetric impacts on herbivore performance and, therefore, inferences about plant resistance (Lee et al., 2016; Scheirs, Zoebisch, & De Bruyn, 2004).

Herbivorous insects can have significant impacts on patterns of plant defence expression. Strong selection pressure is common in interactions with subcortical, tree-colonizing insects where defence failures are often fatal (Raffa, Aukema, Erbilgin, Klepzig, & Wallin, 2005). Plants possess sophisticated, multi-tiered defences against subcortical herbivores, which work in concert to reduce tissue loss and increase the likelihood of tree survival. In general, plant defences can be constitutively present or induced, include chemical and physical components, have different mechanisms of action and are energetically demanding (Gershenzon, 1994; Meldau, Erb, & Baldwin, 2012; Mithöfer & Boland, 2012; Schuman & Baldwin, 2016). The impacts of plant defences on herbivores may vary due to several factors, which include the insect's life history, degree of specialization and the phenotype of the host plant.

Due to the catastrophic injury that insect larval stages can have on plant productivity and survival, most of our understanding of plant defence strategies focuses primarily on maternal oviposition and subsequent larval feeding. Far less is understood about how intra-plant distribution of defences impact adult insect behaviour and physiology, and whether they have similar or divergent impacts compared to conspecific juveniles. This point is particularly critical for generalist species with putatively broad host ranges and for invasive insects encountering naïve host plants. Compared to specialists, generalists are more likely to utilize diverse behavioural strategies to exploit host plants and are less likely to utilize highly chemically defended hosts.

Our study aimed to address the influence of intra-plant variation in nutritional quality and chemical defences that are encountered by adult and larval *Anoplophora glabripennis* (Coleoptera: Lamiinae). We assessed how variation in plant defences impacted adult and larval performance, and whether these impacts are asymmetric for the two life stages. Our hypothesis was that differences in beetle performance would be related to differences in tree chemistry, and differences in chemical composition between tissues would alter outcomes between life stages. Our goals were to: (a) assess whether different host plants elicit disparate effects on herbivore adult feeding, oogenesis and oviposition, (b) determine whether the impacts of host plants had similar or divergent impacts on larval performance and (c) determine whether differences in primary and secondary metabolite composition between tissue types and host plant species explained the observed differences.

Anoplophora glabripennis is an invasive, tree-killing insect that utilizes different host tissues through its development (Haack, Hérard, Sun, & Turgeon, 2009; Hu, Angeli, Schuetz, Luo, & Hajek, 2009; Meng, Hoover, & Keena, 2015). In both its native and invasive range, its most preferred hosts include maples and boxelders (*Acer* spp.). In comparison, poplars (*Populus* spp.) have varying degrees of susceptibility (Hu et al., 2009). *Populus* spp. have well-documented defences against generalists, primarily in the form of constitutively present phenolics. *Populus* spp. produce salicinoids (phenolic glycosides), chemicals that have antifeedant and biochemically reactive properties, which typically have negative effects on generalist insects (Boeckler, Gershenzon, & Unsicker, 2011; Hwang & Lindroth, 1997; Lindroth & Hemming, 1990). *Populus* spp. also produce condensed tannins, which can have negative impacts on specialist insects such as leaf beetles (Ayres, Clausen, Maclean, Redman, & Reichardt, 1997; Donaldson & Lindroth, 2004). The roles of salicinoids and condensed tannins in mediating *A. glabripenni*s–host plant interactions are unknown.

2 | MATERIALS AND METHODS

2.1 | Life cycle and laboratory-rearing conditions

Anoplophora glabripennis undergoes a lengthy, multiyear life cycle (1–3 years depending on temperatures). Adults feed on petioles and young twigs about 1–2 cm in diameter. After egg development and mating, females lay a single egg beneath the bark at the phloem–xylem interface (Keena & Sanchez, 2006). Larvae feed entirely

beneath the bark, first utilizing outer vascular tissue before tunnelling into the sapwood.

The *A. glabripennis* colony we utilized in this study was maintained under quarantine at The Pennsylvania State University (PSU) using procedures described previously (Keena, 2002, 2005). Insects were reared at ~22°C, with adults and newly hatched larvae feeding exclusively on red maple (*Acer rubrum*). *Acer rubrum* twigs for feeding adults and bolts for oviposition were obtained weekly from nearby PSU forests. Egg production occurs ~7–10 days after adult emergence. After 20–30 days, male–female *A. glabripennis* mating pairs were moved into 3.8-L jars where they were given feeding and oviposition substrates. Under these conditions, larvae hatch after 2–3 weeks and are harvested after ~6 weeks. Adult beetles in this colony undergo similar mating behaviours and feeding preferences as populations in the field (Haack et al., 2009; Hu et al., 2009; Keena & Sanchez, 2006; Meng et al., 2015).

2.2 | Plant sources

We used three host plants that varied in suitability for *A. glabripennis*. *Acer rubrum* was used as the highly preferred host and was obtained from mixed hardwood stands in PSU forests. Trees were ~6–8 cm diameter at breast height and ~20 years old at the time of harvest. We used *Populus deltoides* as a host that has putative susceptibility and *Populus tomentosa* that has putative resistance (Rui, Guansheng, & Xixiang, 1995; Weilun & Wen, 2005). Both trees were initially obtained from nursery sources (Lawyer Nursery) and maintained in an outdoor nursery at PSU. At the time of harvest, both *Populus* spp. were ~7 years old. Twigs for adult feeding (1–1.5 cm diameter) were collected at the time of tree felling and bolt harvest. *Acer rubrum* twigs were approximately 1–5 years old and *Populus* twigs were 1–2 years old.

2.3 | Experiment 1: How do different host plants impact *Anoplophora glabripennis* feeding behaviour?

We conducted no-choice and choice feeding bioassays with *A. glabripennis* adults using *A. rubrum*, *P. deltoides* and *P. tomentosa*. In no-choice bioassays, individual adults were provided with four ~1.0-cm-diameter twigs of each host. Twigs were removed and replaced after four days, and the experiment was terminated after one week. In choice bioassays, beetles were provided two twigs of *A. rubrum* paired with two twigs of either *P. deltoides* or *P. tomentosa*. Choice experiments had four beetle replicates, and the no-choice experiment had six replicates. Area of tissue removed was determined by tracing onto paper and quantifying in IMAGEJ (Schneider, Rasband, & Eliceiri, 2012).

2.4 | Experiment 2: What are the impacts of host plants on adult *Anoplophora glabripennis* egg development and gut oxidative stress?

To determine the impacts of host plants on egg development, we provided newly eclosed females twigs of *P. tomentosa*, *P. deltoides* or *A. rubrum*. Solitary females were randomly assigned to one of the three tree species and maintained in a 950-mL vessel with a cotton wick in water. Plant tissues were replaced every 6–7 days, and the assay was completed after 30 days. Beetles were dissected to quantify the number of eggs in the abdomen and collect tissues.

We evaluated the impacts of host plants on oxidative stress in the gut. Midguts were removed and partitioned into anterior and posterior regions (Mason et al., 2017). Dissected guts were maintained on ice, weighed and homogenized in ice-cold 10 mM sodium phosphate buffer (pH 7.0). Cells were pelleted, and supernatant was used to assess protein and lipid oxidation. Protein oxidation was determined by analysing carbonyls according to Levin et al. (1990). Samples were treated with 10 mM 2,4-dinitrophenylhydrazine in 2 N HCl; proteins were precipitated, washed in 50:50 ethyl ether and ethanol, and resuspended in 6 M guanidine. Absorbance was read at 390 nm on a Spectramax 250 plate reader (Molecular Devices), and concentrations were determined using an extinction coefficient ε = 22,000 M^{-1} cm^{-1}. Lipid peroxides were assayed by measuring the oxidation of ferrous iron–xylenol orange complex in methanol (Jiang, Hunt, & Wolff, 1992), using the modifications described in Summers and Felton (1994). Peroxides were analysed by absorbance at 560 nm using t-butyl hydroperoxide as the standard (r^2 > .95).

2.5 | Experiment 3: How do different host plants impact *Anoplophora glabripennis* oviposition behaviour?

We performed no-choice and choice experiments to determine how the three host plants influence *A. glabripennis* egg laying. The experimental set-up utilized a single arena that contained a water pick, *A. rubrum* twigs for feeding and an oviposition substrate in the form of a single 5- to 7-cm-diameter, ~25-cm-long bolt. Five identical replicate chambers were assigned for each of the three tree species, and for the combination of *A. rubrum* and *P. tomentosa*.

We randomly selected male–female pairs from a newly eclosed cohort of *A. glabripennis* adults. Adults were provided *A. rubrum* twigs for ~20 days prior to mating. At the onset of the experiments, female *A. glabripennis* were 17–28 days old. Bolts were replaced after seven days, and the experiment was concluded after 14 days. We observed no adult mortality. Four weeks after the bolt was removed from the jar and five weeks after the initiation of the experiment, we enumerated the number of chew marks, and then removed the bark and quantified the total number of eggs and hatched larvae. Larvae removed from each bolt were weighed.

2.6 | Phytochemical analyses of plant tissues

Branch segments were randomly collected from replicate trees at the time of Experiment 3 and immediately transported to the laboratory. Bark was carefully removed with a sharp razor blade, and both bark and wood were flash frozen. Tissues were lyophilized and then ground through a 1-mm^2 mesh screen. Nutritional content was

estimated as soluble sugars, starches and soluble protein. We analysed two different types of phenolic-based defences: salicinoids, which are specific to *Populus* spp., and condensed tannins, which are present in all three species.

Water-soluble sugars and total starch were quantified as described in Chow and Landhäusser (2004). Sugars were extracted using hot ethanol and measured at 490 nm after reacting with 2% phenol and concentrated sulphuric acid. Starch was solubilized with sodium hydroxide and digested enzymatically. The colouring reagent peroxidase–glucose oxidase/o-dianisidine was combined with the resultant glucose hydrolysate (Sigma Glucose Diagnostic Kit 510A; Sigma-Aldrich) supernatant and 80% sulphuric acid, and measured at 525 nm. Glucose was used to produce standard curves for both carbohydrate assays (r^2 > .97). Proteins were extracted in hot 3% SDS Tris buffer (pH 6.8) and quantified using a non-interfering protein quantification kit (G-Biosciences).

Salicinoids are not produced by *Acer* spp., so these defences were analysed only for *P. deltoides* and *P. tomentosa*. Salicinoids were extracted in ice-cold methanol with sonication and analysed by ultra-high performance liquid chromatography (UPLC; Abreu, Ahnlund, Moritz, & Albrectsen, 2011). Samples were injected onto a Waters Acquity CSH C-18 column (Milford, MA) (2.1 × 100 mm, 1.7 μm) and separated with a Waters integrated Acquity I-Class UPLC at 40°C on a gradient of acidified water and acetonitrile (0.1% formic acid). The mass spectrometer was operated in negative ionization mode with selective ion recording of the salicinoid-formate adducts. Calibrations were based on internal standardization by salicylic acid-d_6 (Sigma-Aldrich) using authentic salicin, salicortin and tremulacin standards.

Condensed tannins were analysed in each plant species using hot HCl–butanol as described by Porter, Hrstich, and Chan (1986). Condensed tannins were extracted in 70% acetone containing 10 mM ascorbic acid as an antioxidant. We used condensed tannins that were extracted from *P. tremuloides* to create a standard curve (Hagerman & Butler, 1980) and analysed the samples at an absorbance of 550 nm.

2.7 | Statistical analyses

Statistical analyses were performed using R v. 3.1.2 in Rstudio (R Team, 2015). We conducted parametric and nonparametric analyses depending upon whether data were normally distributed. Female egg production was analysed with Kruskal–Wallis rank sum. Midgut carbonyls and lipid peroxides were analysed using a two-way analysis of variance (ANOVA) with plant species and midgut tissue segment treated as explanatory variables, with individual included as a random effect. Adult-feeding responses were analysed using Kruskal–Wallis and Wilcox tests. No-choice oviposition responses were analysed using a one-way ANOVA and choice tests with a Wilcox test. Transformations were unable to achieve the assumption of normality for most of the plant metabolites. Therefore, we used a two-way permutation ANOVA to analyse the plant metabolites with plant species, tissue type and their interaction as explanatory variables. Post hoc tests were performed using the R package agricolae (de Mendiburu, 2014).

3 | RESULTS

Host plant species had strong effects on *A. glabripennis* feeding preferences. In no-choice tests (Figure 1a), feeding declined by ~80% when *A. glabripennis* were provided either *Populus* species compared with *A. rubrum* (H = 7.42; p = .024). Comparable results emerged for choice tests (Figure 1b), where *A. glabripennis* strongly preferred *A. rubrum* over *P. deltoides* (V = 0; p = .032) and *P. tomentosa* (V = 21; p = .034).

Host plant dramatically influenced egg production (Figure 2a). When adults were provided *A. rubrum* twigs for feeding, their egg loads ranged from 10 to 15 eggs per female. However, when *A. glabripennis* was maintained on either of the two *Populus* species, egg loads were reduced by 90%–100% (H = 6.72; p = .034).

The oxidative status of the adult *A. glabripennis* gut was impacted by host plant species. Compared to *A. rubrum*, lipid peroxides in beetle guts were 70% and 90% greater in adults fed *P. deltoides* and *P. tomentosa*, respectively (Figure 2b, F = 9.1, p = .003). Lipid peroxides were twofold greater in the anterior compared to the posterior midgut (F = 24.3, p < .001) in beetles fed on all three tree species. Carbonyls followed similar trends as the lipid peroxides (F = 30.6, p < .001). Carbonyls were threefold greater in midgut tissues of

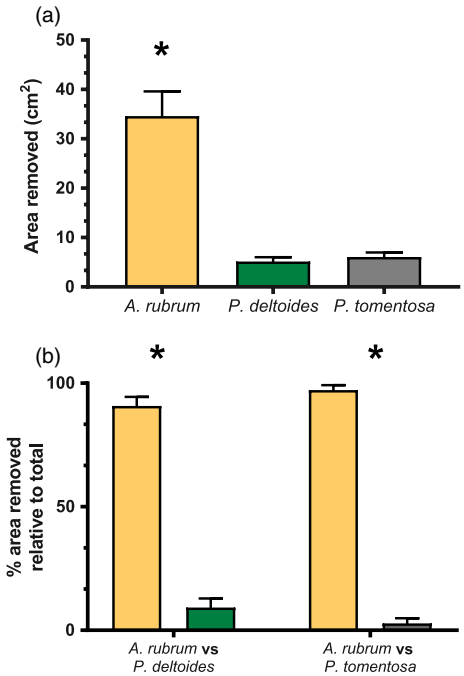

FIGURE 1 Amount of feeding by adult *Anoplophora glabripennis* on twigs of *Acer rubrum*, *Populus deltoides and Populus tomentosa*. Insects were provided twigs in no-choice (a) and choice (b) feeding bioassays. Bars represent mean ± *SE*. *Anoplophora glabripennis* consumed more *Acer rubrum* than either *Populus* in both experiments. Asterisks represent statistically significant differences (p < .05)

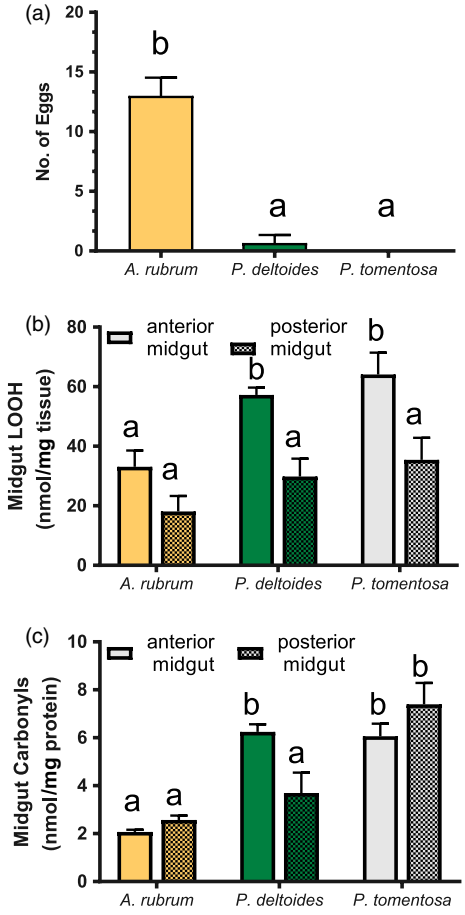

FIGURE 2 Impacts of maturation feeding on different host plants by adult female *Anoplophora glabripennis* on egg load (a), midgut lipid peroxides (b) and midgut protein oxidation (c). Bars represent mean ± *SE*. Statistically significant differences between bars are represented by different letters ($p < .05$). Insects were provided fresh diets weekly over a 30-day period

insects consuming *P. deltoides* and *P. tomentosa* compared to *A. rubrum* (Figure 2c). However, elevated carbonyls in the posterior gut regions were only observed for beetles feeding on *P. deltoides*.

While host plant species produced strong effects on *A. glabripennis* feeding preferences, egg production and gut biochemistry, a different pattern emerged for oviposition behaviour. When females were allowed to feed on *A. rubrum*, they subsequently initiated oviposition behaviours on all three hosts (Figure 3a; $F = 0.79$, $p = .473$). There was likewise no effect of tree species on the number of eggs laid ($F = 2.15$, $p = .152$), or the number of hatched larvae ($F = 3.34$, $p = .07$). There was a strong positive correlation between the number of eggs laid and the number of eggs hatched ($p < .001$, $r^2 = .93$), with no effect of the host tree species on larval eclosion. However, in choice tests, female *A. glabripennis* again overwhelmingly selected *A. rubrum* over *P. tomentosa* ($p < .001$). The influence of host plant on *A. glabripennis* larval mass diverged from their impact on adults. At the time of harvest, the larvae obtained from *P. deltoides* and *P. tomentosa* were 60% and 90% greater in mass, respectively, than larvae from *A. rubrum* (Figure S1, $F = 24.6$, $p < .001$).

Nutrient and water content varied among the three host plant species and between bark and wood (Table 1, Figure 4). Bark contained greater carbohydrate, protein and moisture content compared to the woody tissues for all three species. The sole exception was for starches in *A. rubrum*, which did not differ between bark and wood. Both *Populus* species had similar concentrations of starches and soluble sugars in the bark, being ~60% greater in concentration than in *A. rubrum* (Figure 4a,b). No differences were observed in concentrations of water-soluble sugars in the wood between the three species. Compared to *A. rubrum*, bark protein concentrations were 50% greater in *P. tomentosa* and 40% lower in *P. deltoides*. Wood protein quantities did not differ between tree species. Compared to *A. rubrum*, bark and wood moisture content was greater in both *Populus* spp. ($F = 95.8$, $p < .001$).

Salicinoids and condensed tannins occurred in greater concentration in the bark than in the wood. Salicinoid concentrations were greater in *P. tomentosa* bark compared to *P. deltoides* bark, but in both species only trace concentrations were found in the wood (Figure 4e). *Populus deltoides* was comprised primarily of salicortin (~90% of total) and salicin (~10%), while the salicinoids present in *P. tomentosa* included salicortin (~47%), salicin (~4%) and tremulacin (~49%). *A. rubrum* and *P. deltoides* bark had similar concentrations of condensed tannins (Figure 4), and *P. tomentosa* bark possessed negligible concentrations. Both *A. rubrum* and *P. deltoides* possessed detectable concentrations of condensed tannins in the wood, but concentrations were 3 × higher in *A. rubrum*. In contrast, *P. tomentosa* had only trace amounts of condensed tannins in the wood.

4 | DISCUSSION

Intraspecific utilization of host substrates is common among holometabolous insects, where adult and larval life stages exploit fundamentally different host resources. Insect utilization of different tissue components of the same host presents plants with challenges in mounting defence strategies. Conversely, the strategies that plants employ pose significant challenges to herbivores, which can alter host selection and attack behaviours. Our results illustrate asymmetry in host plant defences encountered by adult and juvenile *A. glabripennis*. Females were incapable of developing eggs when feeding on *Populus*, likely through a combination of lower consumption and oxidative stress in their midguts. In contrast, defences failed to protect against egg laying and juvenile establishment when only a single host was available. Collectively, our results demonstrate that host plants can have divergent impacts on insect conspecifics, related to feeding strategies and variation in defences. These effects extend to potential field-level processes, as adults *A. glabripennis* are mobile and known to exhibit varying attack patterns.

Maternal choice can vary between herbivore species and populations, where some may select either optimal or suboptimal diet choices for their offspring (Clark, Hartley, & Johnson, 2011; Friberg, Posledovich, & Wiklund, 2015; Garcia-Robledo & Horvitz, 2012; Gripenberg, Mayhew, Parnell, & Roslin, 2010; Handley,

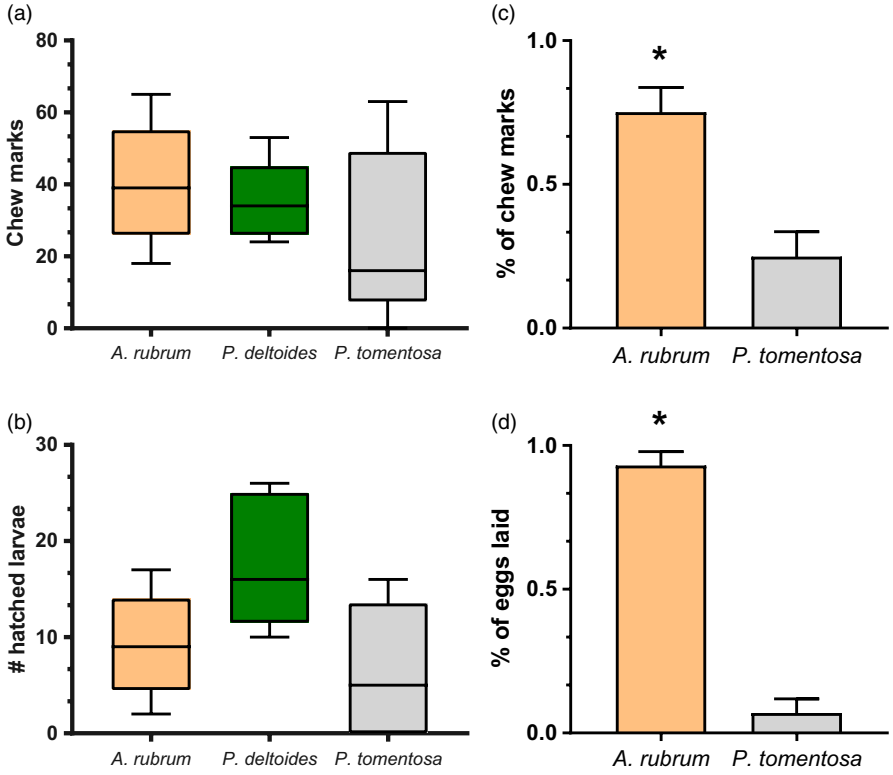

FIGURE 3 Oviposition behaviour and number of hatched larvae when female *Anoplophora glabripennis* were provided twigs for feeding and a bolt for oviposition of *Acer rubrum*, *Populus deltoides* or *Populus tomentosa*. *Anoplophora glabripennis* produced chew marks (a), eggs and larvae (b) in all three hosts. There were no effects of host plant on number of laid eggs or larval eclosion

TABLE 1 Effects of host species (*Acer rubrum*, *Populus deltoides*, *Populus tomentosa*) and tissue (bark vs. wood) on primary and secondary metabolite concentrations (Figure 4). Samples used for water content were separate from other tissues. Salicinoids are not present in *Acer rubrum*. Data were analysed using permutation ANOVA

	Species		Tissue		Interaction	
Metabolite	*F*-value (*df*)	*p*-value	*F*-value (*df*)	*p*-value	*F*-value (*df*)	*p*-value
Water-soluble sugars	1.1 (2, 28)	.361	74.7 (1, 28)	<.001	2.5 (2, 28)	.103
Starches	4.9 (2, 28)	.015	10.6 (1, 28)	.003	2.1 (2, 28)	.146
Protein	18.6 (2, 28)	<.001	150.0 (1, 28)	<.001	9.3 (2, 28)	<.001
Water content	95.8 (2, 24)	<.001	109.2 (1, 24)	<.001	10.1 (2, 24)	.001
Condensed tannins	64.5 (2, 28)	<.001	238.0 (1, 28)	<.001	28.0 (2, 28)	<.001
Salicinoids	78.8 (1, 20)	<.001	452.2 (1, 20)	<.001	81.3 (1, 20)	<.001

Ekbom, & Ågren, 2005; Hufnagel, Schilmiller, Ali, & Szendrei, 2017). Alternatively, the inverse has been observed, where host plant dietary constraints on adults impacted reproduction and fitness, yet did not completely extend to negative impacts on brood performance (Lee et al., 2016; Scheirs, Bruyn, & Verhagen, 2000; Scheirs et al., 2004; Smith, Johnson, Davidowitz, & Bronstein, 2018). In some instances, such as in *Trichobaris* weevils, adult beetles avoid host plants that produce toxins (Lee et al., 2016), but these toxins have no apparent effect on juveniles infesting host plants. The work we report here mirrors that of the *Trichobaris*–tobacco interactions.

Herbivores commonly encounter chemically diverse dietary landscapes that impact their ability to exploit specific plant resources. All animals have ideal nutritive intakes, but the ability to identify and exploit the optimal resource can vary depending upon herbivore life history, geography and available diets (Barrett & Heil, 2012). Herbivores may be able to overcome these barriers through the selection of plants and/or tissues that are palatable, or though mixing of diets to optimize intakes and development of the population (Behmer, 2009). Importantly, optimized diet resources can differ for the same herbivore throughout development (Stockhoff, 1993; Unsicker, Oswald, & Weisser, 2008). These factors contribute to the realized dietary breadth of a particular species. In addition, depending upon feeding patterns, digestive processes and nutritional requirements, the breadth of suitable diets can differ between adults and juveniles (Altermatt & Pearse, 2011; Garcıa-Robledo & Horvitz, 2011). In the system we explored here, we show the importance of defence expression in altering realized host ranges (Ludwig et al., 2002; Rui et al., 1995; Yan & Qin, 1992).

Successful establishment of invasive insects involves multiple intersecting variables (Brockerhoff & Liebhold, 2017; Liebhold

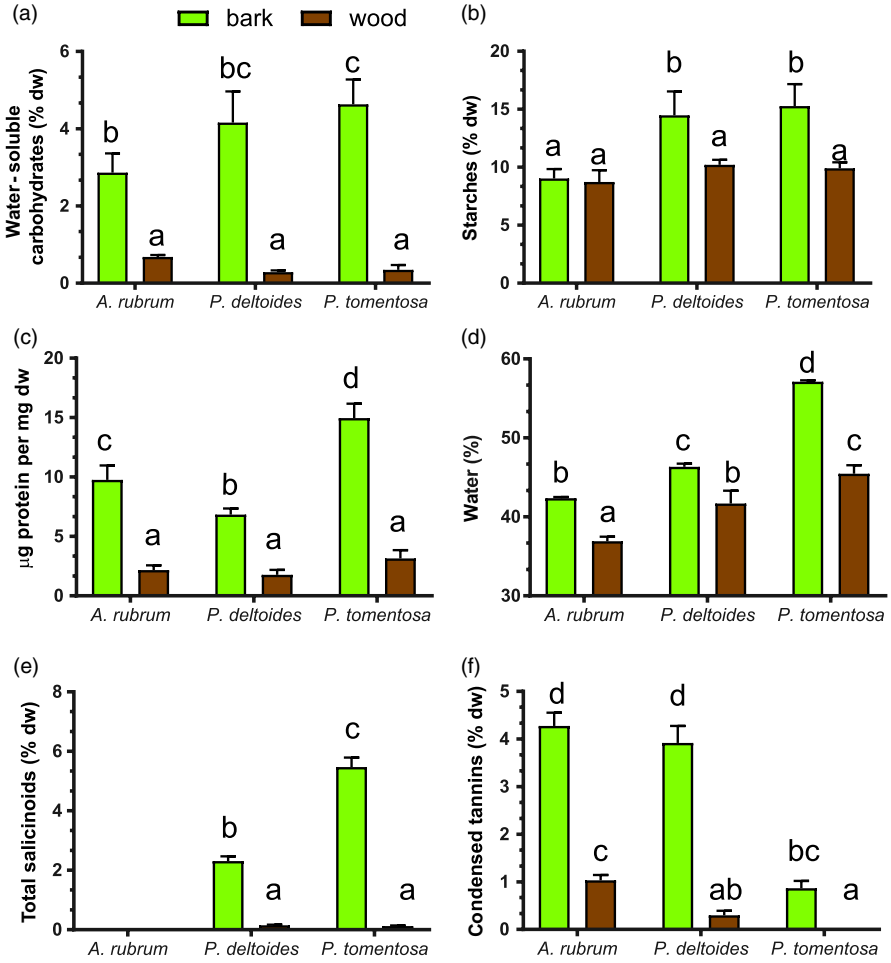

FIGURE 4 Primary metabolites (a–c), water content (d) and secondary metabolites (e and f) in bark and wood of *Acer rubrum*, *Populus deltoides* and *Populus tomentosa*. Letters represent statistically significant differences ($p < .05$) between the plants. *Acer rubrum* does not contain salicinoids

et al., 2017), which include the ability to exploit poorly defended hosts. Host plant exploitation can occur through various mechanisms. Plants may be unable to mount adequate defences (Herms & McCullough, 2014), insects may possess symbionts with elevated virulence (Fraedrich et al., 2008), or the herbivore may possess diverse metabolic and/or behavioural abilities to avoid plant defences. This third scenario seems most in line with *A. glabripennis*, as not only does this species have massive expansions of genes encoding detoxification-related genes in its genome (McKenna et al., 2016), but, as we demonstrate here, it also possesses behaviours that allow its brood to avoid high levels of chemical defences. This pattern seems consistent with generalist invaders; while they have some preference for certain hosts, they will readily utilize other, less preferred plants when populations are at high densities and optimal resources have been exhausted. By using behaviours that avoid host plant defences, *A. glabripennis* is able to expand its larval dietary substrates, a trait that likely contributes to its successful establishment in novel ecosystems.

Salicinoids were the metabolites that clearly distinguished the differences between adults and offspring in the different hosts. These compounds have broadly acting properties on both arthropod and mammalian herbivores (Lindroth, Donaldson, Stevens, & Gusse, 2007; Lindroth & St. Clair, 2013). Despite the patterns we observed, *A. glabripennis* has been documented to use *Populus* in Northern China for reproduction (Weilun & Wen, 2005; Yan & Qin, 1992; Yang, 2005) and was reported as an important pest of *Populus* in China decades prior to its invasion of North America and Europe (Kang-Jou, 1982). Our results here do not seem to be due to a potential genetic bottleneck of a laboratory maintained population, as bioassays conducted with *A. glabripennis* populations in southern China illustrated similar feeding responses (Figure S2). While there are several possibilities that explain this divergence, the most likely scenario is that the *A. glabripennis* in these different populations have different strategies to either detoxify or avoid various defences. Currently, behavioural, physiological and/or biochemical mechanisms that might distinguish discrete native and invasive *A. glabripennis* populations are unclear and require further exploration.

By consuming different substrates and avoiding plant defences, host range can differ for adults and their offspring. Plants possess suites of traits that reduce and alter herbivory, and understanding how defensive and nutritive traits of host plants alter interactions through herbivore ontogeny is unclear in most systems. While we focused here on the decoupling of host suitability between adult and juveniles, the juvenile experience may alter adult processes. The interplay between adult and juvenile host ranges through ontogeny

and across generations, as well as the impacts and mechanisms of plant defences in altering plant–herbivore relationships, is a crucial knowledge gap for many important herbivore species.

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AUTHORS' CONTRIBUTIONS

C.J.M. and K.H. developed the overall questions and hypotheses; C.J.M. and D.C.L. designed and performed the experiments; C.J.M. and R.L.L. performed the chemical analyses; C.J.M. analysed the data; C.J.M. and K.H. led the writing of the manuscript; all authors contributed critically to the manuscript drafts and gave final approval for publication.

DATA AVAILABILITY STATEMENT

Data have been archived in the Dryad Digital Repository under: http://doi.org/10.5061/dryad.g7n0k6n (Mason, Long, Lindroth, & Hoover, 2019).

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