

EFFECTS OF GENETIC AND ENVIRONMENTAL VARIATION IN  
*SOLIDAGO ALTISSIMA* ON ASSOCIATED ARTHROPOD COMMUNITIES

A Thesis  
by  
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## **FOREWORD**

The research detailed in this thesis will be submitted to *Oecologia*, an international peer-reviewed journal published by Springer. The thesis has been prepared according to *Oecologia's* author guidelines.

## **ABSTRACT**

EFFECTS OF GENETIC AND ENVIRONMENTAL VARIATION IN *SOLIDAGO ALTISSIMA* ON ASSOCIATED INSECT COMMUNITIES. (August 2012)

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The current unprecedeted rate of biodiversity loss places a growing urgency on the need to elucidate the factors driving community and ecosystem dynamics. Previous studies demonstrate a positive relationship between plant interspecific diversity and insect community diversity, but more recent focus has included the effects of plant intraspecific diversity on associated arthropod communities. Variation among plants in traits important to insects provides potential mechanisms for differential insect response to genetically dissimilar conspecifics. Because a plant's physiology and susceptibility to herbivory are, in part, regulated by environment, it is important to consider the large-scale role of environmental variation in affecting insect community structure. My objective was to determine how genetic and environmental variation within a *Solidago altissima* population affects the structure of the associated insect community and colonization by a dominant herbivore. Additionally, I considered insect responses to variation in biomass production and foliar quality to investigate potential mechanisms driving observed patterns.

I used a common garden approach to test the effects of plant intraspecific variation on insect abundance and community structure. *Solidago altissima* ramets were propagated at

the Appalachian State University (ASU) greenhouse and four genotypes from four elevations (260 m, 585 m, 885 m, 1126 m) were planted in a common garden at the ASU Gilley Research Station. In August 2011, the insect community was quantified using vacuum sampling methods, and the aphid, *Uroleucon nigrotuberculatum*, was quantified visually. Leaves were collected to assess foliar metrics: Nitrogen (N), Carbon:Nitrogen (CN), and volatile terpenes, and aboveground biomass was estimated non-destructively. Insects were assigned to a morphospecies, and community abundance, richness, and evenness were calculated. Insects were also grouped into feeding guilds to examine trophic level effects.

Both host-plant genotype and environment affected insect community structure. Variation in host-plant genotype affected community richness and colonization by the specialist aphid. Though effects of environment are harder to discern because of my experimental design, environmental variation appears to affect abundance, where plants from 885 m supported the highest number of insects. Aphid abundance did not vary between plants from different elevations. Linear regression analysis revealed relationships between insect community measures and foliar water, N, CN, and terpene concentrations. Plants from 885 m had the highest nutritional quality (i.e., lowest CN), water content, and insect abundance. Results of the one-way ANOVA suggest that the environment may regulate plant phenotypic expression, which is reflected in insect community structure.

In conclusion, I found that both host-plant genetic and environmental variation affect insect community structure, where small-scale genetic variation is more influential to specialist insect population dynamics, and large-scale environmental variation is more important to structuring the rest of the insect community.

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## INTRODUCTION

As anthropogenic impacts continue to alter ecosystems through critical losses in biodiversity (Chapin et al. 2000), there is a pressing need to better understand the factors driving community and ecosystem processes. Elucidating the relationship between plant and insect communities is especially important as this interaction can affect a number of ecosystem processes including net primary production, nutrient dynamics, and pollination (Weisser and Siemann 2004). Plant-insect interactions are a function of both bottom-up (i.e., effects of the plant community on arthropod community) and top-down (i.e., effects of the arthropod community on the plant community) forces (Weisser and Siemann 2004). Particularly important is the bottom-up effect plant community species diversity can have on arthropod community structure.

A well-established positive relationship exists between interspecific diversity of plant and insect communities (Crutsinger et al. 2006; Haddad et al. 2009). However, ecologists in the relatively new field of community genetics are testing how genetic diversity within a plant population (i.e., intraspecific diversity) may act as an organizing force on arthropod community structure. Within a plant population, both the number of genotypes (i.e., genotypic diversity; Frankham et al. 2003; Hughes et al. 2008), and the genetic differences between individuals (i.e., genotype identity; Johnson and Stinchcombe 2007), have been shown to have a similar effect on arthropod community structure as does plant community species diversity. Identifying the mechanisms responsible for the link between plant and arthropod community diversity is essential to attaining a broader understanding of the factors

structuring these communities that are so important to ecosystem health (Weisser and Siemann 2004). Proposed mechanisms primarily involve bottom-up forces, and include the effects that differences in plant primary production (Crutsinger et al. 2006) and nutritional quality have on the arthropod community (Basset 1991; Cisneros and Godfrey 2001; Stiling and Moon 2005). Additionally, environmental variation is an essential component in community genetics studies, as both selective forces and an individuals' response to these forces will vary in different environments. Including environmental variation in community genetics studies allows comparison of the relative importance of small- (genetic differences between host-plant patches) and large-scale spatial variation (environmental variation) in structuring arthropod communities (Johnson and Agrawal 2005).

### **Linking plant and arthropod community diversity**

It is well established among community ecologists that plant species diversity is positively related to diversity of the insect community, as well as to ecosystem stability and productivity (Tews et al. 2004; Tilman et al. 2006). Therefore, as plant species diversity increases, one can expect a similar increase in arthropod community diversity and ecosystem functioning. Ecosystem processes such as primary productivity (Tilman et al. 2006) and decomposition rate (Torsvik and Øvreås 2002) have been shown to increase with plant and microbial community diversity, respectively. The community diversity of organisms tightly linked to primary producers (i.e., arthropod communities) has also been shown to respond positively to increases in plant interspecific diversity, though multiple mechanistic explanations exist for this positive correlation. According to the more individuals hypothesis (Srivastava and Lawton 1998), greater plant diversity is expected to directly affect herbivorous insects through increases in plant biomass (Tilman et al. 2006; Haddad et al.

2009). These effects may transcend the herbivore community to higher trophic levels, as predators and parasitoids react to increases in prey abundance (Johnson and Agrawal 2005). For example, in experimental plots with diversity ranging from 1 – 16 plant species, it was found that as the number of plant species increased, cumulative herbivore species richness increased by 43% and cumulative predator richness increased by 35% (Haddad et al. 2009).

Other hypotheses explaining the observed relationship between plant and insect community diversity hinge on the fact that the majority of herbivorous insects exhibit specialized feeding behaviors, consuming only a single or narrow range of closely related host-plant species (Bernays and Graham 1988). The resource specialization hypothesis predicts that a more diverse plant community will support a more diverse arthropod community due to increases in available resources and microhabitats for specialist insect species (Southwood et al. 1979; Haddad et al. 2009). For example, Wenninger and Inouye (2008) found that species diversity of the plant community affected insect abundance and richness at the start of, but not later in, the growing season. Specialist insects dominated the community early on, but as the season progressed, a shift in the relative prevalence of specialist to generalist insects reduced the arthropod communities' dependence on specific attributes of the plant community composition and diversity.

Alternatively, the resource concentration hypothesis (Root 1973) predicts that less specious plant communities will support higher abundances of specialist herbivorous insects, which seek out, and usually remain on, dense clusters of their specific host-plant. Accordingly, communities composed only of a single or few plant species growing in concentrated clusters should sustain higher specialist herbivore abundances compared to diverse plant communities. For example, Koricheva et al. (2000) found that the abundance

of host-specific leafhoppers was highest in monoculture plots and decreased linearly as the interspecific diversity of the plant community increased.

The enemies hypothesis (Root 1973) approaches herbivore community dynamics from a top-down perspective. Arthropod predators may respond positively to increases in primary production and vegetation structural diversity that is characteristic of highly diverse plant communities. This proposed positive link between plant community diversity and predator abundance may act to control herbivore dynamics through increased predation.

### **Community genetics**

Traditionally, ecologists have approached questions about plant-insect community dynamics by focusing on the effects of interspecific diversity, but more recently these studies have considered the influence of intraspecific diversity within a plant population on insect community structure. The emerging field of community genetics integrates ecological and evolutionary processes in effort to gain new insight into the factors driving community structure and ecosystem processes (Whitham et al. 2003, 2006, 2008; Johnson and Stinchcombe 2007; Hughes et al. 2008). The incorporation of a genetic component into community ecology studies may provide a mechanistic approach to disentangling the factors driving plant-insect interactions. Many community genetics studies consider how intraspecific genetic diversity influences associated communities through effects of the extended phenotype (Whitham et al. 2003; Wimp et al. 2005). The concept of the extended phenotype recognizes the effects of genes beyond the population level (Dawkins 1982, 1999; Whitham et al. 2003; Bailey et al. 2009). If a phenotype is to have far-reaching effects in an ecosystem, individuals within a population must vary genetically in ecologically important traits (e.g., growth rate, foliar chemistry; Hughes et al. 2008). When variation exists in

ecologically important traits within a population, it can be predicted that: (1) different genotypes will vary in the species they support, and (2) a positive relationship exists between population genetic diversity and diversity of the associated community (Wimp et al. 2005).

Many community genetics studies focus on the effects of genetic diversity in a plant population on associated arthropod communities. Plant populations provide a model system for investigating effects of extended phenotypes, as variation in a basal resource may act as a strong organizational force on the structure of associated communities; an effect that is predicted to transpire across trophic levels (Johnson and Stinchcombe 2007). Arthropod communities also provide an ideal system to test for community-level effects of genetic diversity, as most arthropod communities are characteristically diverse, representing a range of feeding guilds and functional roles (Wimp et al. 2004). Additionally, arthropods rely on plants for the nutritional resources and microhabitats they provide, with some specialist insect species (e.g., *Uroleucon* species) carrying out entire lifecycles on an individual plant (Pilson 1992). This direct interaction between plant populations and arthropod communities offers a mechanistic approach to testing for a genetic component to community structure.

Many ecosystems are composed of a few plant species that dominate the vegetative biomass. Due to their prevalence in the plant community, the extended phenotype of these dominant, or foundation species, should have considerable effects on associated community and ecosystem dynamics (Whitham et al. 2003, 2008; Ohgushi et al. 2011) and are commonly used in community genetic studies (Wimp et al. 2004; Genung et al. 2010). Additionally, plant species that reproduce clonally are often used in community genetics studies, as clonal reproduction provides an easy opportunity to experimentally manipulate genetic diversity (Hughes et al. 2008). Recognizing each clone as a genetically unique

individual allows one to empirically test for the community-level effects of host-plant intraspecific diversity by manipulating the genotypic diversity within a plant population or by manipulating the genotypes present in a population (Crutsinger et al. 2006; Johnson and Stinchcombe 2007; Hughes et al. 2008).

One approach to community genetics studies is to manipulate plant population genotypic diversity (Hughes et al. 2008; Bailey et al. 2009) in an effort to elucidate these effects on community-level processes. Ecosystem stability and productivity have been shown to increase with host-plant population genotypic diversity (Hughes and Stachowicz 2004; Tews et al. 2004), resulting in a host-plant population capable of supporting an abundant and diverse insect community (Crutsinger et al. 2006). For example, Crutsinger et al. (2006) found that plots containing 12 genotypes had 36% higher aboveground net primary productivity (ANPP) compared to single genotype plots. Furthermore, the plots with the highest genotypic diversity also had the highest herbivore and predator species richness, suggesting that intraspecific plant genotypic diversity can affect multiple trophic levels. The authors attributed the positive relationship between genotypic diversity and ANPP to niche complementarity, or beneficial interactions between genotypes, in polyculture plots. Furthermore, in support of the more individuals hypothesis (Srivastava and Lawton 1998), polyculture plots were able to support a diverse arthropod community through increases in ANPP by providing insects with greater food and microhabitat availability (Tilman et al. 2006; Haddad et al. 2009). Niche complementarity between genotypes resulting in increased ANPP provides a potential mechanism for the positive relationship between intraspecific genotypic diversity of a host-plant population and interspecific diversity of the insect community.

An alternate approach to community genetics studies considers how genetic variation between individuals (i.e., genotype identity) can affect associated communities (Johnson and Stinchcombe 2007). For example, Wimp et al. (2004) found that genetic diversity among individual *Populus* (cottonwood) hybrids accounted for 60% of arthropod community diversity. This result highlights the importance of genotype identity to the structure of associated communities. Community genetics studies rely on the assumption that individuals in a plant population vary genetically in traits that will influence the fitness of associated communities (Hughes et al. 2008). Variation in plant constituents, such as defensive compounds, is an ecologically important trait that can affect arthropod community structure (Wimp et al. 2007; Bidart-Bouzat and Kleibenstein 2008; Gols et al. 2008). A community genetics approach may be especially important when considering how plant defenses affect community structure, as there is generally a high level of genetic variation in expression of these traits (Wimp et al. 2007; Bidart-Bouzat and Kleibenstein 2008; Gols et al. 2008).

### **Effects of plant quality on arthropod community structure**

Host plant selection by phytophagous insects is partially driven by differences in plant nutritional quality (Basset 1991; Barber and Marquis 2011). Variables that affect plant quality and may influence host choice include foliar carbon and nitrogen concentrations and plant resistance and defense traits. In addition, life-history traits of an arthropod species will affect herbivore distribution and response to changes in plant quality (Huberty and Denno 2006; Zehnder et al. 2009).

Nitrogen, which is essential for the synthesis of amino acids and proteins, is considered the most limiting nutrient for phytophagous insects (Mattson 1980). The nitrogen content of plants (1-5%) is significantly lower than that of insects (~10%), creating a

disparity in the nitrogen available to herbivorous insects (Mattson 1980; Huberty and Denno 2006). Many studies have demonstrated the positive effects of nitrogen addition on herbivore densities and fitness related variables (Cisneros and Godfrey 2001; Stiling and Moon 2005; Huberty and Denno 2006; Zehnder et al. 2009). For example, nitrogen fertilization resulted in increased body size, greater survival, reduced development time, and higher densities of two planthopper species, *Prokelisia dolus* and *P. marginatna* (Huberty and Denno 2006). Plants with higher foliar nitrogen content would be expected to support a more robust and abundant insect community, but individual responses to changes in nutritional quality may be species-specific and not reflective of the arthropod community as a whole. For example, Zehnder et al. (2009) found that leaf chewer abundance was more responsive to changes in foliar nitrogen concentration, while phloem feeder abundance was more responsive to increases in carbon based structural compounds. Furthermore, the extent to which changes in plant quality will affect an individual may be mediated by the diet breadth of a species. When faced with a nutritionally poor food source, a polyphagous, generalist insect is able disperse and feed on nutritionally higher quality plants, while the limited host-plant range of a specialist insect may force an individual to remain and feed on a nutritionally poor plant.

As sedentary organisms, plants have evolved resistance and defense mechanisms to deter or reduce the inevitable damage from herbivores. Plant resistance and defense traits have either a morphological or chemical basis and will vary both within and between plant species and across spatial and temporal scales (Maddox and Root 1987; Hengxiao et al. 1999; Agrawal 2010; Hakes and Cronin 2011a).

One way plants lessen the negative fitness effects of herbivore pressure is through morphological resistance traits. Tougher leaves make penetrating leaf tissue difficult for herbivores, and plants with tough leaves generally experience reduced herbivory compared to plants with more tender leaves (Sagers and Coley 1995). Variation in feeding pressure from herbivores may act as a selective force driving differential allocation to resistance traits between genotypes. For example, by assessing the genetic variability and broad-sense heritability of common resistance and tolerance traits in *Solidago altissima*, Hakes and Cronin (2011b) found that the herbivore community acted as strong selective force for increased tolerance and reduced resistance in host-plants. Furthermore, the authors predicted that selective pressure from the herbivore community would ultimately lead to a decrease in the frequency of resistant genotypes in the *S. altissima* population. Additionally, leaf toughness is expected to have disproportionate effects across insect feeding guilds, as leaf chewers and miners should be more negatively influenced by leaf toughness than phloem feeders (Zehnder et al. 2009). Consequently, arthropod community composition, and especially the occurrence of certain feeding guilds within the community, can drive selection for resistance traits in the plant community.

Plants have also evolved a suite of chemical defenses in response to herbivory. There are two main classes of plant chemical defense: (1) constitutive defenses, which are always expressed regardless of herbivore pressure, and (2) induced defenses, which are expressed only as a response to herbivore damage (Howe and Jander 2008). The type and magnitude of chemical defenses utilized by an individual plant is species-specific and will vary as a result of host-plant genotype identity and interactions between the neighboring plant community, herbivore community, and abiotic factors (Gershenson and Engelberth 2010).

Carbon-based phenolic compounds are one group of constitutive secondary metabolites that play a role in plant chemical defense against herbivores and pathogens (Dudt and Shure 1994; Gershenzon and Engelberth 2010). Tannins are a defensive compound that deter insect feeding and act as toxins to reduce herbivore fitness upon ingestion (Forkner et al. 2004; Gershenzon and Engelberth 2010). For example, Kopper et al. (2002) found that developmental time of the tussock moth larvae (*Orgyia leucostigma*) was 44% longer when fed a moderate tannin diet compared to a control-no tannin diet. Additionally, growth rates for moths fed a moderate tannin diet were 42% lower compared to the control (Kopper et al. 2002). However, insect responses to carbon-based defenses vary and specialist insects that have long been associated with a specific host-plant species may have adaptive mechanisms to cope with the presence of these otherwise toxic compounds (Appel 1993; Barbenhenn et al. 2003). Thus, in plant communities with high tannin concentrations, one may predict that through adaptation, specialist herbivores will have a competitive advantage over generalist herbivores and may come to dominate the insect community.

Lignin is another carbon-based phenolic compound that deters herbivory. Within the plant, lignin has a primary structural function as well as a secondary defensive function (Gershenzon and Engelberth 2010). The chemical structure of lignin that makes the compound ideal for providing physical support to the plant also contributes to the secondary defensive properties of the compound (Gershenzon and Engelberth 2010). Lignin increases leaf toughness, making it difficult for phytophagous insects to pierce plant tissue and access nutritional resources. In addition, the lignin is difficult for herbivores to digest, further enhancing the repellent properties of the compound.

Induced resistance strategies allow a plant to alter its defenses based on the severity of herbivore pressure (Karban 2011). Thus, induced resistances provide plants a means of conserving resources: allowing an individual to favor investment towards growth or reproduction when herbivore pressure is low or shifting investment to produce defensive compounds as herbivory increases. Induced responses are generally categorized as either direct or indirect. A direct response affects the interaction between an herbivore and its host-plant. An indirect response signals higher trophic levels, affecting herbivore populations by attracting their natural enemies (Turlings et al. 1990).

One class of induced resistances, referred to as volatile organic compounds (volatiles), are emitted from the plant to the atmosphere, where the signals are then encountered by herbivores, predators, and parasites. Volatiles may also remain attached to plant tissue surfaces and can deter herbivores from feeding because of their foul taste (Gershenson and Engelberth 2010). A relevant group are the terpenoids, or terpenes (see Langenheim 1994 for review). Volatiles can directly affect herbivores, acting as a repellent to the plant-feeding insects, or indirectly, acting as an attractant to their predators (Maffei 2010). For instance, O'Reilly-Wapstra et al. (2007) showed that slugs, a significant plant herbivore, consumed less pine (*Pinus sylvestris*) needles and seedlings when they contained high levels of monoterpenes. This result indicated that (1) slugs were able to distinguish between differential levels of monoterpenes, preferentially feeding on tissues or whole plants with lower terpene concentrations, and (2) that terpenes directly deterred and reduced herbivore damage.

Evidence of increased plant fitness due to indirect induction of volatiles has been demonstrated in maize plants (*Zea mays*) fed on by cotton leaf worm (*Spodoptera littoralis*)

larvae (Fritzsche-Hoballah and Turlings 2001). Upon herbivory, maize plants released volatiles, attracting two parasitoids, *Cotesia rubecula* and *Camptocletis sonorensis*, to the damaged plant. The plants fed on by parasitized *S. littoralis* larvae produced 30% more seeds compared to plants fed on by unparasitized *S. littoralis* larvae. This example demonstrates how higher trophic levels can act as a selective force for plants to increase concentrations of volatile defensive compounds in an effort to reduce herbivore fitness and densities. If defensive compounds vary by genotype between conspecifics, possible explanations for effects of intraspecific diversity on arthropod communities may emerge.

Though the insect community can act as a selective force for plants to invest in high concentrations of defensive compounds, the abiotic environment can also play an important role in a plant's ability to produce these secondary metabolites. Phenolic compounds require a considerable carbon input, and, consequently, light and nutrient availability play a crucial role in a plant's ability to invest in these defensive compounds (Langenheim 1994; Hakes and Cronin 2011a). For example, Dudit and Shure (1994) found a positive relationship between light availability and total phenolics for both tulip poplar (*Liriodendron tulipifera*) and dogwood (*Cornus florida*) trees. To fully understand the factors driving insect community dynamics it is important to consider the complicated network of bottom-up and top-down forces acting simultaneously on both the plant- and insect-communities.

### **Effects of environmental variation on plant and insect communities**

An individual's phenotype is a function of the interaction between genotype and environment, and, therefore, environmental heterogeneity can affect the extended phenotype at the population level (Whitham et al. 2003). While many studies have established positive effects of host-plant genetic diversity on arthropod species richness (Dungey et al. 2000;

Wimp et al. 2004; Bangert et al. 2006; Crutsinger et al. 2006, 2008a, 2008b; Johnson et al. 2006), the relative contributions of host-plant genotype, environment, and a genotype by environment interaction on arthropod community dynamics are still largely unknown (Johnson and Stinchcombe 2007). Genotypes may respond differently in discrete environments, and, therefore, environmental variation should be regarded as a potential factor influencing the extended phenotype. For example, Maddox and Cappuccino (1986) found that *S. altissima* individuals grown in a low water treatment supported lower aphid abundances compared to individuals grown in a high water treatment, however, variation in aphid density between genotypes was only significant in the high water treatment. This result suggests that the susceptibility of *S. altissima* genotypes to aphid attack was regulated by the environment. Furthermore, varying edaphic conditions have been found to affect the rate of herbivory and oviposition among *Solidago* genets (Wise et al. 2006).

However, a study using aspen trees (*Populus tremuloides*) testing for the relative importance of genotype, environment, and a genotype by environment interaction on gypsy moth (*Lymantria dispar*) growth rate and developmental time found that genotype accounted for more than 76% of the variation in gypsy moth performance (Osier and Lindroth 2001). Additionally, Bangert et al. (2006) found a positive correlation between arthropod community diversity and genetic diversity of *Populus* trees that persisted across spatial scales from the individual (tree), stand, river, and regional level. Especially noteworthy was that this finding was scale dependent, with the relationship weakening as spatial scale increased (Bangert et al. 2006). This allows one to speculate that plant genetic diversity has a greater effect on arthropod community structure at smaller, local scales, while environmental variation may be more important to community structure across large spatial scales (Johnson

and Agrawal 2005; Bailey et al. 2009). Determining the relative importance of host-plant genetic diversity, environmental variation, and a genotype by environment interaction is essential to disentangling the factors structuring arthropod communities.

Generally, environmental variation increases with spatial scale, and communities become less similar as the distance between patches increases (Bangert et al. 2006; Hakes and Cronin 2011a). This dissimilarity may be especially pronounced between communities distributed along an elevational gradient (Ohsawa and Ide 2008). For example, changing environmental conditions along an altitudinal gradient often results in predictable differences in foliar quality between plant populations, which are reflected in arthropod community structure. Compared to low and mid elevation populations, plants growing at high altitudes must cope with suboptimal environmental conditions, particularly a cooler and shorter growing season (Ohsawa and Ide 2008). A shortened growing season is reflected in plant population differentiation of life history traits, including growth rate and reproductive effort (Olsson and Ågren 2002). Plants restricted by the length of the growing season are subject to strong selection for rapid growth, and, therefore, often allocate resources towards growth over defense. Alternatively, plant populations at lower elevations experience longer growing seasons and have more freedom to allocate resources to secondary metabolic processes (Olsson and Ågren 2002). For example, the foliar tannin content of *Betula papyrifera* trees was nearly two times higher in low elevations compared to high elevations (Erelli et al. 1998). This result, in addition to the finding that *B. papyrifera* trees from mountain habitats had higher growth rates than those from valley habitats, supports the prediction that plants under selection for fast growth do not invest heavily in constitutive defenses to deter herbivory, but rather rely on replacing damaged tissue through new growth.

Variation in abiotic conditions along an altitudinal gradient may also affect arthropod community structure. Insect species richness and abundance generally peak at low to mid elevations (Hodkinson 2005). For example, herbivore abundance in deciduous *Nothofagus pumilio* forests was 14-fold higher in low elevation compared to high elevation sites (Garibaldi et al. 2011). Furthermore, the difference in insect abundance resulted in a 2.5-fold increase in leaf area damaged by herbivores at low elevation compared to high elevation sites. This differential pattern of insect community structure along an altitudinal gradient often results in a selection pressure gradient for increased allocation to plant defense as elevation decreases (Salmore and Hunter 2001).

Temperature also directly affects insect growth and development, with lower temperatures generally resulting in a longer developmental time and reduced growth rates (Garibaldi et al. 2011). Therefore, even though plants at high elevations may be more susceptible to herbivory due to decreased allocation to defenses, potential defoliation by the herbivore community is limited by temperature dependent restrictions on insect activity such as consumption. For example, the leaf beetle, *Galerucella grisescens*, had decreased oviposition and consumption rates at higher elevations, despite the fact that foliar quality was higher at this site (Suzuki 1998). Decreased insect activity due to low temperatures allows plants at higher elevations to have higher nutritional quality (i.e., nitrogen content) for herbivores (Kröner 1989; Hodkinson 2005) while maintaining low levels of defensive compounds.

### **Study system**

The consequences of an extended phenotype may be particularly influential when an ecosystem is dominated by a few host-plant species (Wimp et al. 2004). *Solidago altissima*

(tall goldenrod) is a dominant herbaceous species common to old field ecosystems and roadsides across eastern North America (Pilson 1992). As a perennial plant, *S. altissima* produces an underground rhizome which sprouts multiple ramets to produce clones (Maddox et al. 1989). Genetic diversity of natural *S. altissima* populations is variable, ranging from 1-12 genotypes in less than a square meter (Maddox et al. 1989). This variation in genotype density provides a natural system for comparing the results of studies that experimentally manipulate *Solidago* genetic diversity. *Solidago* species also rely on a diverse pollinator community for sexual reproduction, allowing for investigations into how genetic diversity can affect pollination rates, which can then indirectly affect a number of ecosystem processes (Genung et al. 2010). *Solidago altissima* also supports a diverse herbivore community, with more than 100 species from several functional groups depending on the plant for food and habitat (Maddox and Root 1990). These characteristics make this species ideal for studying the effects of genetic diversity on associated communities and ecosystem function.

My study set out to answer the following 3 questions:

- 1) Does genetic diversity within a *S. altissima* population have an effect on arthropod species richness and abundance?
- 2) Does arthropod community structure vary between *S. altissima* genotypes collected from different sites?
- 3) How does genetically or environmentally mediated variation in leaf chemistry influence associated arthropod communities?

## METHODS AND MATERIALS

### **Field site**

The study site was located at the Appalachian State University (ASU) Gilley Research Station ( $36^{\circ} 17' 10.22''$  N,  $81^{\circ} 35' 11.69''$  W; elevation = 1055 meters) in Todd, NC. The system is characterized as an early-successional old-field ecosystem, composed primarily of *Solidago* species surrounded by a 120 + hectare forest.

### **Rhizome collection and propagation**

In summer 2009 *S. altissima* (tall goldenrod) ramets were collected by Jennifer Schweitzer and Joseph Bailey, researchers at the University of Tennessee-Knoxville. Ramets were collected from locations of varying elevation in east Tennessee: 227 meters (m), 260 m, 585 m, 885 m, 1126 m. Multiple spatially separated patches were sampled within each elevation. Because *S. altissima* is a clonal species with a compact rhizome structure (Maddox et al. 1989), each plant was considered to be a unique genotype. We recognize the need to verify genetic identity and are currently working with a microsatellite protocol.

Plants were potted and allowed to cold harden until December 2009 when they were transferred into the ASU greenhouse. In May 2010, rhizomes from 1-2 plants per genotype were cut into 3 centimeter (cm) sections and planted in 8.89 cm flats in a common greenhouse environment. Rhizome sections were placed horizontally in flats approximately

2.54 cm below the soil (FaFard 4M mix soil) surface. Plants were transferred to larger pots as needed. After three weeks, plants were moved outside until December 2010 when they were moved back into the greenhouse due to harsh weather.

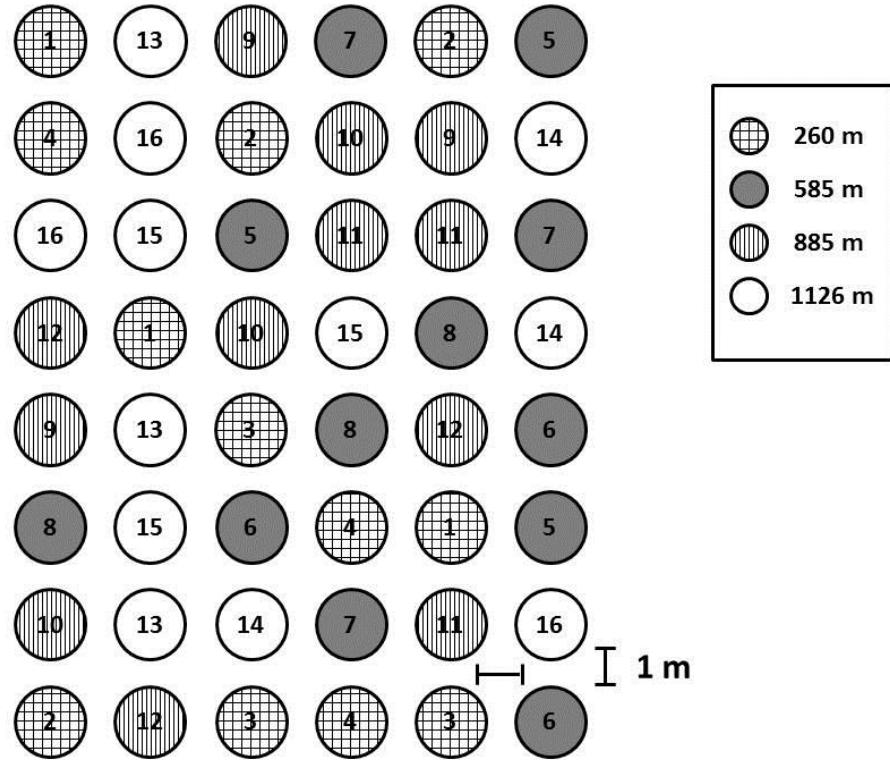
In early March 2011, 5 genotypes from selected elevations were chosen from the rhizome stock for inclusion in the common garden. Elevations and genotypes to be included in the common garden were selected based on available rhizome length. Fifty individuals per genotype were propagated in soil flats following the procedure described above. Upon initial planting, each rhizome flat was supplied with 50 milliliters (mL) of a 100:1 (water:concentrate) mix of root stimulator (Roots, Hummert International). Flats were stored in the greenhouse on vertical shelving units and rotated among shelves to reduce variation during the indoor growth phase (March 9, 2011 – May 19, 2011). Due to lack of sufficient rhizome growth, the common garden was scaled down to include 4 genotypes from 4 elevations (260 m, 585 m, 885 m and 1126 m), for a total of 16 genotypes. Each genotype was assigned a number 1-16. Genotypes from 260 m were numbered 1 – 4; genotypes from 585 m were numbered 5 - 8; genotypes from 885 m were numbered 9 - 12; and genotypes from 1126 m were numbered 13 - 16.

### **Common garden**

To eliminate natural *S. altissima* and other herbaceous vegetation in our planting area, glyphosate (Roundup; Monsanto) was twice applied to the field site (April 7, 2011 and April 30, 2011). Any remaining vegetation was removed by hand and the garden site was tilled (May 3–May 5, 2011). In addition, trees bordering the site were cleared to ensure all plots in the garden were exposed to similar sun conditions.

The common garden was established May 19, 2011. Using string, a horizontal grid was set up to delineate meter wide rows within the garden. Within alternating rows, a 1 x 1 m polyvinyl chloride (PVC) frame served as a plot border. For all 16 genotypes, eight clones from a single genotype were planted in a plot, and each plot was replicated 3 times ( $N = 384$ ). The 13 x 17 m garden was composed of 6 rows containing 8 plots each. Plots were spaced 1 m apart (Fig. 1). Plot location was randomly assigned prior to planting by drawing numbers out of a hat.

Plants of varying size were used to discourage bias based on differences in aboveground biomass between plots during insect host-plant selection. To ensure standardization of plant distribution between plots, a large (0.97 m diameter) and small (0.71 m diameter) hula hoop were placed within the 1 x 1 m PVC frame. Four individuals were then planted in each corner of the square frame, and four individuals were planted in the inner hula hoop, one at each cardinal direction. This method of plant distribution resulted in more circular shaped plots, which served to reduce edge effects. Each plot was watered for the first two weeks, as needed, to promote successful establishment of individuals in the field. Any plants that appeared overly small or unhealthy were replaced (less than 10 overall) with heartier individuals from the plant stock at the ASU greenhouse. Plants were allowed to grow undisturbed through June and July 2011. Plots were weeded by hand on a biweekly schedule and vegetation in the open areas surrounding each plot was mowed on a weekly basis.



**Fig. 1** Schematic of the 13 x 17 m common garden. Each circle represents a plot containing 8 clones from a single genotype ( $N = 384$ ). Numbers correspond to genotype where: genotypes from 260 m were numbered 1 – 4; genotypes from 585 m were numbered 5 – 8; genotypes from 885 m were numbered 9 – 12; and genotypes from 1126 m were numbered 13 – 16.

### Insect community

On August 9-10, 2011 a visual survey was conducted to quantify aphid abundance. Aphids that associate with *Solidago* generally aggregate on the upper portion of the plant stem (personal observation), and vacuum sampling methods are not sufficient to generate accurate estimates of aphid abundance. The aphid survey was conducted by visually counting aphids on each individual within a plot. Though aphid morphotypes were noted, the vast majority of encountered aphids were the Asteraceae specialists, *Uroleucon nigrotuberculatum*.

On August 10, 2011 plots were vacuum sampled to assess the entire insect community using a TORO Ultra Electric Blower Vac (Model 51599). A 1 x 1 x 1 m chamber constructed from PVC pipe and window screen was placed over the plots to prevent insects from fleeing once vacuum sampling began. Each plot was sampled for 90 seconds by vacuuming all plants. Insects were kept cool in zip lock bags in the field and transferred to a freezer in the lab.

Insects were separated from soil and plant particles with the aid of a Leica zoom 2000 dissecting microscope and then stored in 70% ethanol. All specimens were initially identified to family level (following Borror and White 1970), then assigned to a morphospecies. Identification to morphospecies level relies on morphological characteristics to differentiate between individuals, and, though it is less discerning than a classic species level identification, it is commonly used when qualifying extremely specious systems, such as an arthropod community (Derraik et al. 2002). A digital library of Gilley site insects was developed using a camera to examine morphological characters. These images were used as a reference when quantifying the entire community.

When characterizing the insect community, insect abundance was defined as the number of individuals counted, richness was defined as the number of morphospecies counted (Boulinier et al. 1998), and evenness was calculated using the Shannon-Weinnaer diversity index ( $H'$ ; Rieske and Buss 2001):

Evenness =  $H'/\log_e S$ , where  $S$  is the number of morphospecies in the sample.

$H' = \sum p_i \log p_i$ , where  $p_i$  is the number of morphospecies divided by the total number of insects.

Analysis of the *Uroleucon nigrotuberculatum* population was conducted separately from the rest of the insect community because this species dominated the insect community. In order to characterize the insect community separate from the preponderance of this dominant herbivore, aphids collected by vacuum sampling were analyzed along with the visual data.

Absolute and corrected community measures were analyzed. Corrected measures accounted for differences in aboveground biomass production between plots and were defined as plot level totals for abundance or richness per gram of biomass (abundance/g biomass and richness/g biomass).

### **Aboveground biomass**

Plant height and stem diameter were measured and used as predictor variables to establish an allometric equation for estimating aboveground biomass non-destructively. At least one plant was measured from each plot and 50% of the plots were sampled twice. Stem height (cm) was measured from the base of the stem to the tip of the apical meristem. A caliper was used to measure stem diameter (millimeters; mm) approximately 7.62 cm above the soil surface. Values were summed for individuals with multiple sprouts. After removing outliers, measurements from 40 plants were used to develop an allometric equation ( $y = 0.0022x + 6.367, p = 0.001, r^2 = 0.70$ ). This calculation was used to estimate total aboveground biomass in all plots non-destructively immediately after the insect sampling.

### **Foliar measures**

Leaves were collected the same day the insect community was sampled (August 10, 2011) to ensure that chemical constituents reflected the sampled insect community. For

terpene analysis, 4-10 leaves (enough to generate a 1.5 g sample for gas chromatography protocol) were collected from two randomly selected plants in each plot. Only fully expanded, mature leaves were collected. Samples were kept in a cooler until they were transported to the lab. Fresh leaves were weighed (grams; g) using a Mettler Toldeo AG245 balance before being stored in a freezer for later analysis.

For all other leaf measures including fresh weight, dry weight, leaf area, Nitrogen (N), and Carbon:Nitrogen (CN) three leaves from four randomly selected plants were collected in each plot (12 leaves per plot). Leaves were collected from the bottom, middle, and top of sampled plants. In the field, leaves were stored in a cooler, in zip lock bags containing a damp paper towel, and were later transferred to a refrigerator in the lab.

Leaves were randomly paired off (6 leaf pairs per plot) and fresh weight (g) was measured using a Mettler Toldeo AG245 balance. To determine leaf area ( $\text{cm}^2$ ), leaf pairs were run through a LiCor 3100 Area Meter. Leaves were then stored in a 60°C drying oven for at least 48 hours. Specific leaf weight (SLW;  $\text{mg}/\text{cm}^2$ ) was calculated from the dry weight and leaf area data. Dried leaf pairs were weighed again to determine foliar dry weight (g), and foliar water content (%) was calculated using the formula:

$$[(\text{fresh weight} - \text{dry weight})/(\text{fresh weight})] \times 100\%.$$

Three leaf pairs per plot were randomly selected for foliar N and CN analysis. Dried leaf material was ground to a fine powder using a Super-dent amalgamator on medium speed for 20 seconds. Ground foliar material was stored in a 60°C drying oven. Samples (5-8 milligrams; mg) were weighed on a Mettler Toldeo AG245 balance and analyzed for C and N concentration (mg/g) using a ThermoFinnigan Flash EA1112. CN ratio was calculated from foliar C and N levels.

### **Gas chromatography - terpenes**

Frozen leaves were cut into approximately 3 mm pieces and placed into a 50 mL culture tube. Fifteen mL of high performance liquid chromatography grade pentane was added atop the leaves and this mixture was ground for 60 seconds using a Polytron tissue homogenizer (Brinkmann Instruments). The pentane was then poured into a culture tube through a funnel lined with filter paper. Samples were evaporated to 0.5 mL using nitrogen gas. All equipment was cleaned with acetone to avoid contamination between samples.

Terpenes were quantified using a Shimadzo GC-14A Gas Chromatograph (GC) with a flame ionization detector and a Stabilwax column (30 m x 0.25 mm). A 1 microliter sample was injected into the GC using a syringe (Hamilton Co., MICROLITER 7000 series). The GC program had a total run time of 24 minutes: an initial oven temperature of 80°C was maintained for 2 minutes, then the oven temperature increased at 10°C/minute to a final temperature of 280°C; the final temperature was held for 2 minutes (modified from Johnson et al. 2007).

### **Statistical analysis**

A nested ANOVA with genotype nested within elevation (Proc Nested, SAS version 9.3) was used to analyze the main effects of genotype and elevation on insect community measures, aphid abundance, leaf chemistry, and plant biomass. A one-way ANOVA (Proc GLM, SAS version 9.3) was used to analyze the effect of genotype within each elevation. A *post hoc* Tukey test was used for pairwise comparisons (SAS version 9.3). Linear regression (Proc Reg, SAS version 9.3) was used to examine relationships between plant measures (independent variable) and insect responses (dependent variable). For this analysis, plot

means ( $n = 3$ ) were used at the level of genotype, while for elevation all genotypes ( $n = 4$ ) within an elevation were averaged. Significance levels were set at  $p \leq 0.05$ , and  $0.05 \leq p \leq 0.1$  are presented as marginally significant for all analyses. Data were log transformed (base 10) as appropriate to increase normality.

## RESULTS

### **Insect community**

Vacuum sampling collected a total of 996 individuals (excluding aphids), representing 10 orders and 6 feeding guilds (Tables 1, 2). Most individuals were from order Homoptera, which made up 45.1% of the community (Table 1). Herbivores were the dominant feeding guild, making up more than 75% of the captured insect community (Table 2).

**Table 1** Insect community abundance.

<b>Order</b>	<b>Abundance</b>	<b>% Total</b>
Homoptera	449	45.1
Hemiptera	188	18.9
Coleoptera	133	13.4
Diptera	94	9.4
Hymenoptera	45	4.5
Psocoptera	30	3.0
Lepidoptera	10	1.0
Orthoptera	6	0.006
Neuroptera	2	0.002
Mantodea	1	0.001
<b>Total</b>	<b>996</b>	

**Table 2** Insect abundance in guilds.

<b>Guild</b>	<b>Abundance</b>	<b>% Total</b>
Herbivore	763	76.6
Predator	39	3.91
Parasitoid	53	5.32
Detritivore	30	3.01
Fungivore	7	0.702
Pollinator	4	0.402
Unknown	100	10.0
<b>Total</b>	<b>996</b>	

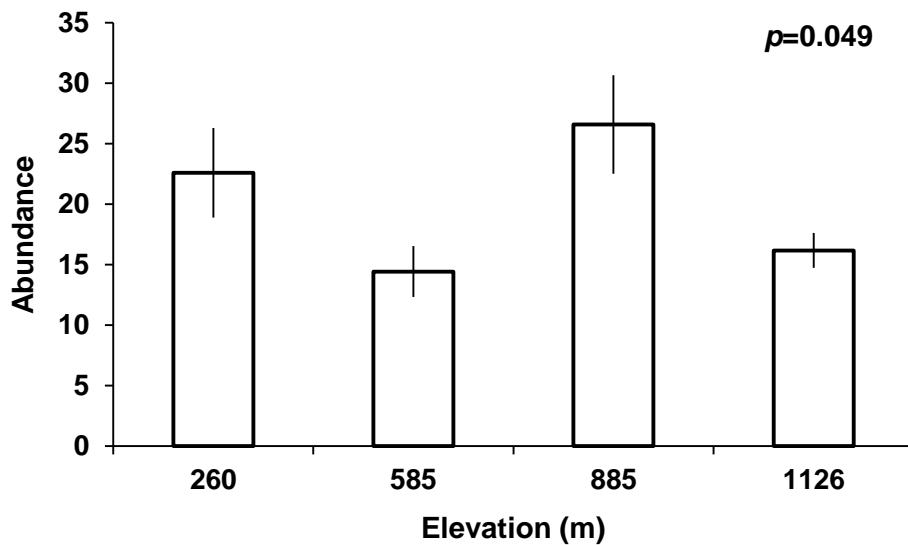
Total abundance was the only community measure significantly affected by elevation (Table 3), with plants from 585 m and 885 m supporting the lowest and highest insect abundances, respectively (Fig. 2). Abundance/g biomass was not significantly affected by elevation (Fig. 3). There was no genotype effect on abundance, or abundance/g biomass (Fig. 4, Table 3). There was a significant effect of genotype on abundance/g biomass in the lowest (260 m) and highest (1126 m) elevations (Fig. 5, Table 4).

**Table 3** *F* ratio, *p* value, and degrees of freedom (*df*)<sup>a</sup> for the effects of host-plant elevation and genotype on insect community measures. *n* = 48 (Proc Nested).

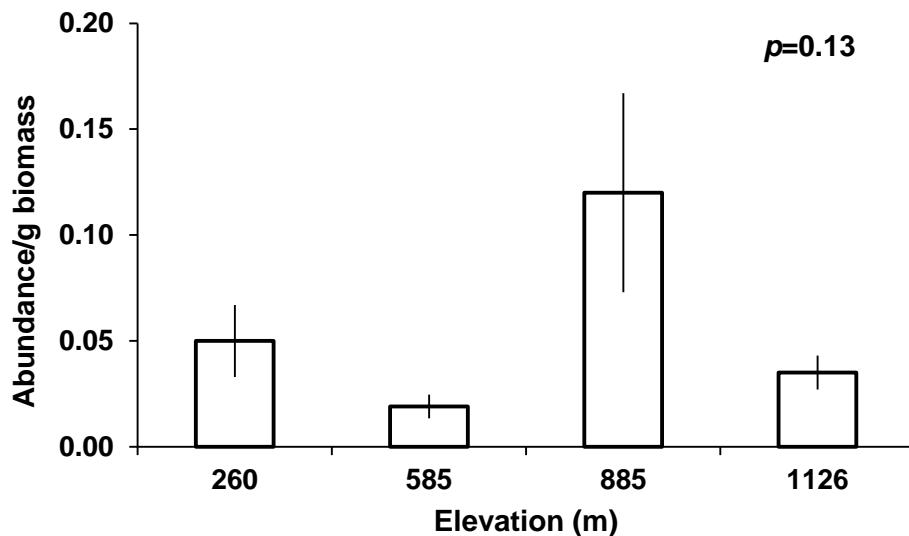
Community measure	Elevation		Genotype	
	<i>F</i>	<i>p</i>	<i>F</i>	<i>p</i>
Abundance	3.49	<b>0.0499</b>	0.99	0.4823
Abundance/g biomass	2.25	0.1344	1.54	0.1602
Richness	1.14	0.3717	0.47	0.9168
Richness/g biomass	1.12	0.3795	3.77	<b>0.0013</b>
Evenness	1.01	0.4237	0.99	0.4751

**Note:**  $p \leq 0.05$  and  $0.05 \leq p \leq 0.1$  presented in bold text.

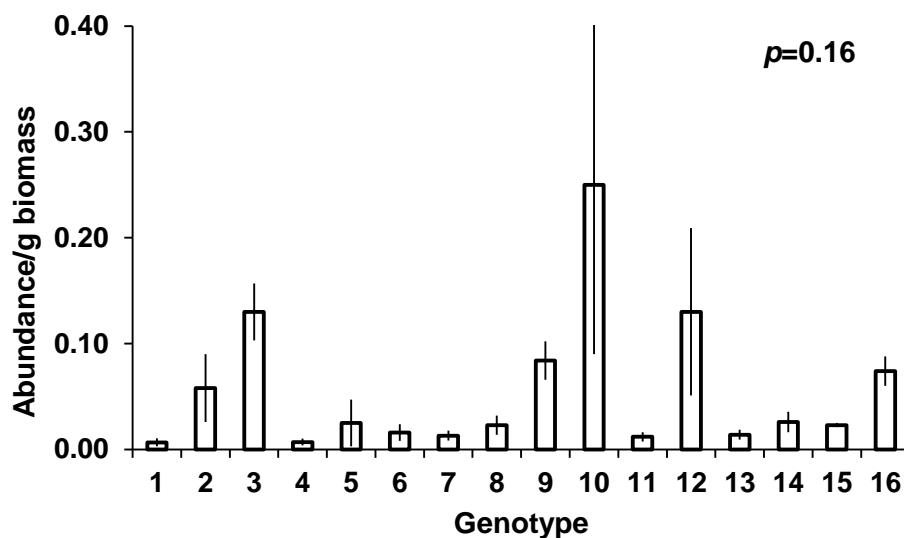
<sup>a</sup> Elevation *df* = 3, 32 Genotype *df* = 12, 32.



**Fig. 2** Effect of elevation on insect abundance (mean±SE;  $p \leq 0.1$  considered significant).



**Fig. 3** Effect of elevation on insect abundance/g biomass (mean $\pm$ SE;  $p \leq 0.1$  considered significant).

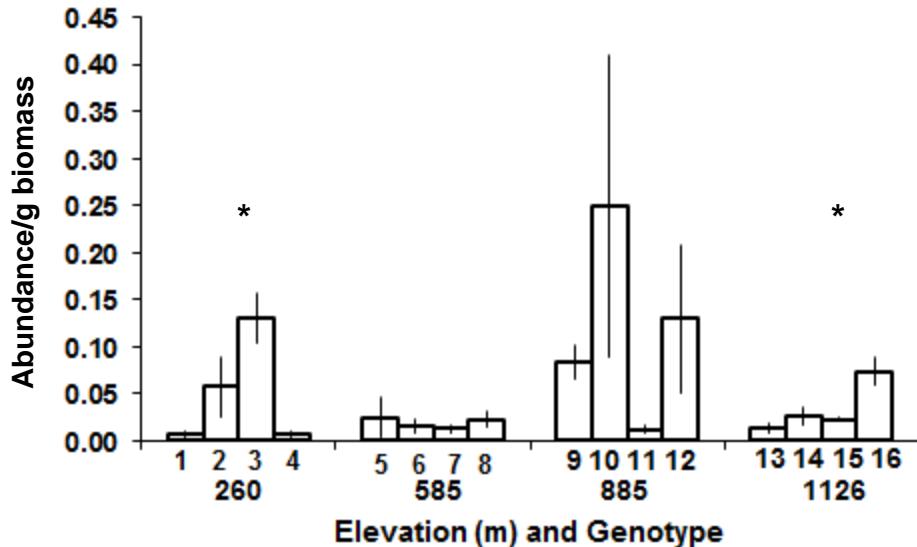


**Fig. 4** Effect of genotype on insect abundance/g biomass (mean $\pm$ SE;  $p \leq 0.1$  considered significant).

Community measure	260 m		585 m		885 m		1126 m	
	F	p	F	p	F	p	F	p
Abundance	1.22	0.3643	0.28	0.8352	1.01	0.4377	1.22	0.3647
Abundance/g biomass	7.47	<b>0.0105</b>	0.18	0.9071	1.18	0.3754	8.97	<b>0.0061</b>
Richness	0.96	0.4581	0.08	0.9627	0.28	0.8360	0.72	0.5651
Richness/g biomass	7.14	<b>0.0119</b>	0.18	0.9071	3.20	<b>0.0839</b>	9.47	<b>0.0052</b>
Evenness	0.99	0.4431	3.56	<b>0.0671</b>	0.61	0.6264	0.35	0.7889

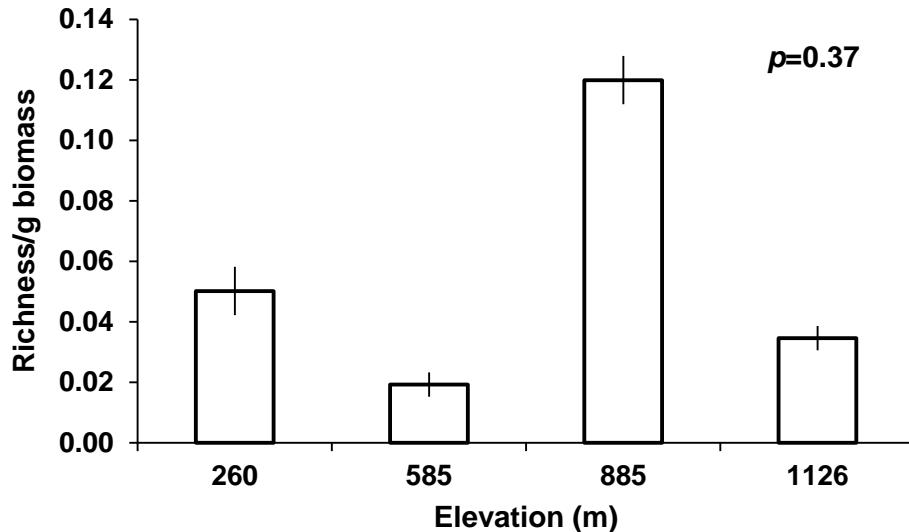
Note:  $p \leq 0.05$  and  $0.05 \leq p \leq 0.1$  presented in bold text.  
<sup>a</sup>  $df = 3, 8$ .

Table 4 F ratio, p value, and degrees of freedom ( $df^a$ ) for the effects of host-plant genotype on insect community measures within elevations.  $n = 48$  (Proc GLM).

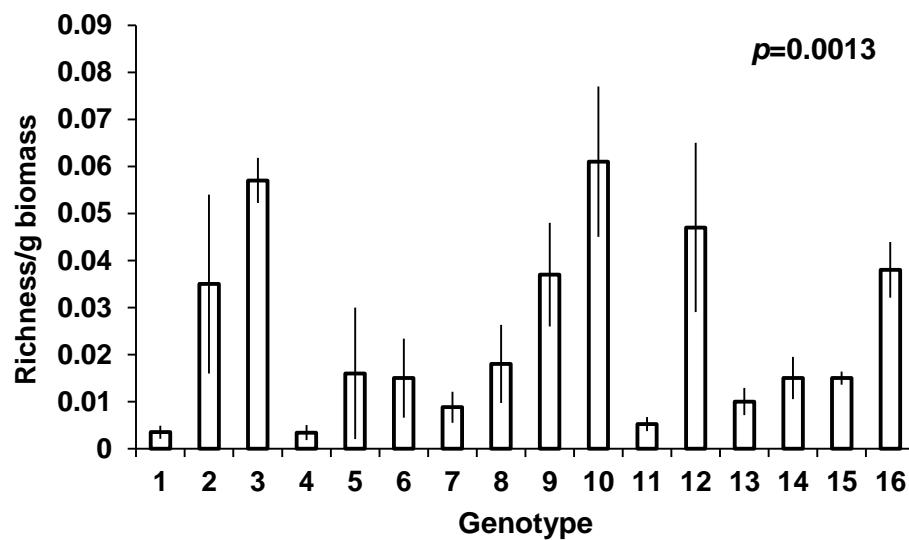


**Fig. 5.** Effect of genotype (1-16) on insect abundance/g biomass (mean $\pm$ SE) within elevations (260 m, 585 m, 885 m, 1126 m). \* = significant ( $p \leq 0.1$ ) genotype effect within elevation.

There was no elevation effect on richness/g biomass (Fig. 6, Table 3), though there was a pattern where the elevation with the highest abundance/g biomass also had the highest richness/g biomass (885 m), and the elevation with the lowest abundance/g biomass had the lowest richness/g biomass (585 m; Figs. 3, 6). Richness/g biomass was the only community measure significantly affected by genotype (Fig. 7, Table 3).

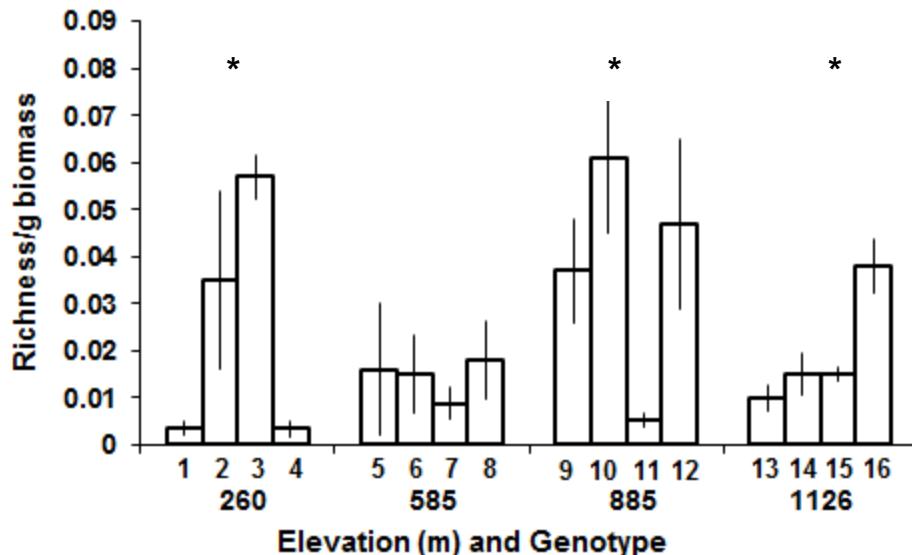


**Fig. 6** Effect of elevation on richness/g biomass (mean $\pm$ SE;  $p \leq 0.1$  considered significant).

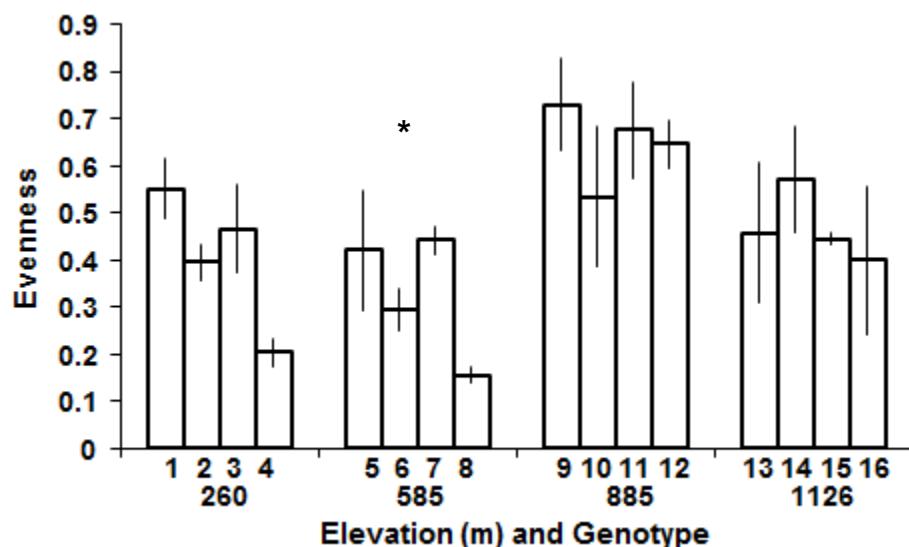


**Fig. 7** Effect of genotype on richness/g biomass (mean $\pm$ SE;  $p \leq 0.1$  considered significant).

Within elevations 260 m, 885 m, and 1126 m there was a genotype effect on richness/g biomass (Table 4), with genotypes 3, 10, and 16 supporting the greatest richness/g biomass compared to the other genotypes within these elevations, respectively (Fig. 8). There was a marginally significant genotype effect on community evenness but only on plants from 585 m (Fig. 9, Table 4).

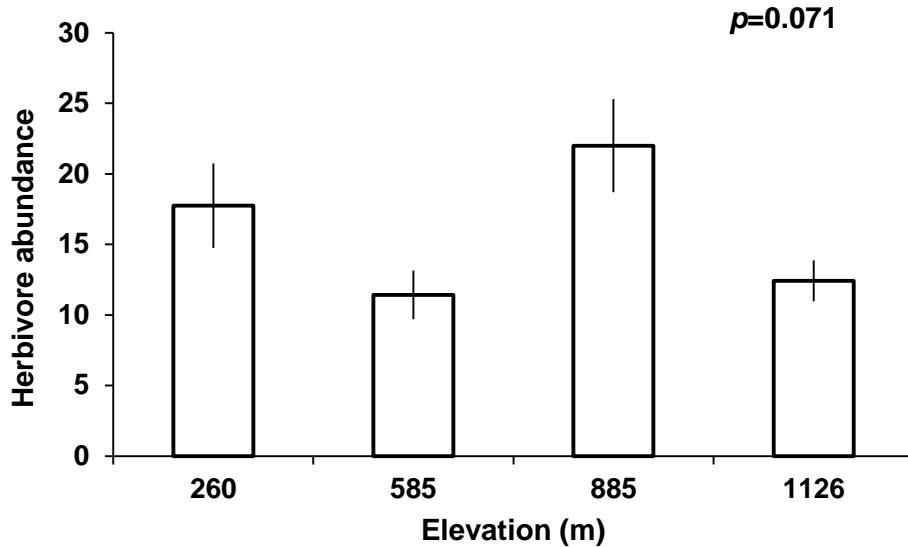


**Fig. 8** Effect of genotype (1-16) on richness/g biomass (mean $\pm$ SE) within elevations (260 m, 585 m, 885 m, 1126 m). \* = significant ( $p \leq 0.1$ ) genotype effect within elevation.

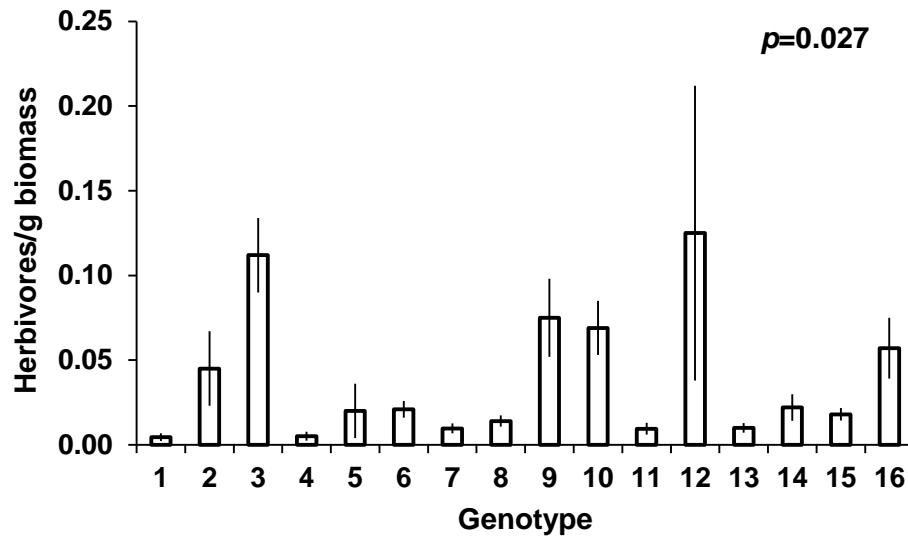


**Fig. 9** Effect of genotype (1-16) on evenness (mean $\pm$ SE) within elevations (260 m, 585 m, 885 m, 1126 m). \* = significant ( $p \leq 0.1$ ) genotype effect within elevation.

Herbivores were the only feeding guild affected by host-plant elevation or genotype (Figs. 10, 11, Table 5). Within elevations 260 m and 1126 m there was a genotype effect on herbivores/g biomass (Table 6). Within elevation 585 m there was a genotype effect on predators/g biomass (Table 6).



**Fig. 10** Effect of elevation on herbivore abundance (mean $\pm$ SE;  $p \leq 0.1$  considered significant).



**Fig. 11** Effect of genotype on herbivore abundance (mean $\pm$ SE;  $p \leq 0.1$  considered significant).

**Table 5** *F* ratio, *p* value, and degrees of freedom (*df*)<sup>a</sup> for the effects of host-plant elevation and genotype on feeding guild distribution. *n* = 48 (Proc Nested).

<b>Feeding guild</b>	<b>Elevation</b>		<b>Genotype</b>	
	<b><i>F</i></b>	<b><i>p</i></b>	<b><i>F</i></b>	<b><i>p</i></b>
Herbivore	3.03	<b>0.0711</b>	1.44	0.1994
Herbivore/g biomass	1.62	0.2360	2.34	<b>0.0274</b>
Predator	1.14	0.3721	1.16	0.3485
Predator/g biomass	0.34	0.7991	0.96	0.5021
Parasitoid	2.06	0.1593	0.40	0.9510
Parasitoid/g biomass	1.01	0.4206	0.91	0.5507

**Note:**  $p \leq 0.05$  and  $0.05 \leq p \leq 0.1$  presented in bold text.

<sup>a</sup> Elevation *df* = 3, 32 Genotype *df* = 12, 32.

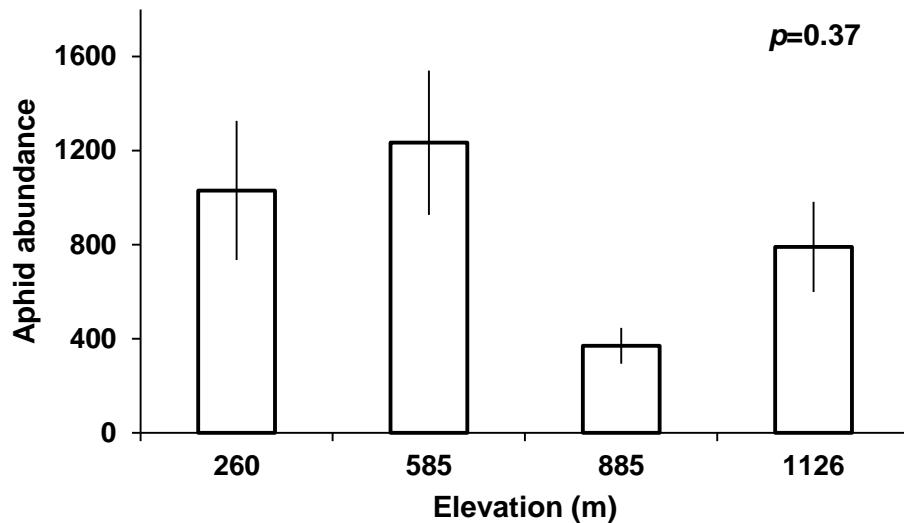
Feeding guild	260 m		585 m		885 m		1126 m	
	F	p	F	p	F	p	F	p
Herbivore	2.12	0.1760	0.64	0.6130	1.28	0.3450	1.24	0.3588
Herbivore/g biomass	10.38	<b>0.0039</b>	0.36	0.7869	1.22	0.3627	4.21	<b>0.0461</b>
Predator	1.28	0.3442	1.67	0.2503	0.83	0.5122	0.83	0.5122
Predator/g biomass	0.49	0.7018	3.27	<b>0.0799</b>	2.11	0.1766	0.60	0.6316
Parasitoid	1.29	0.3437	0.11	0.9512	0.14	0.9359	0.69	0.5820
Parasitoid/g biomass	0.57	0.6482	0.59	0.6396	0.90	0.4829	1.16	0.3816

Note:  $p \leq 0.05$  and  $0.05 \leq p \leq 0.1$  presented in bold text.  
<sup>a</sup>  $df = 3, 8$ .

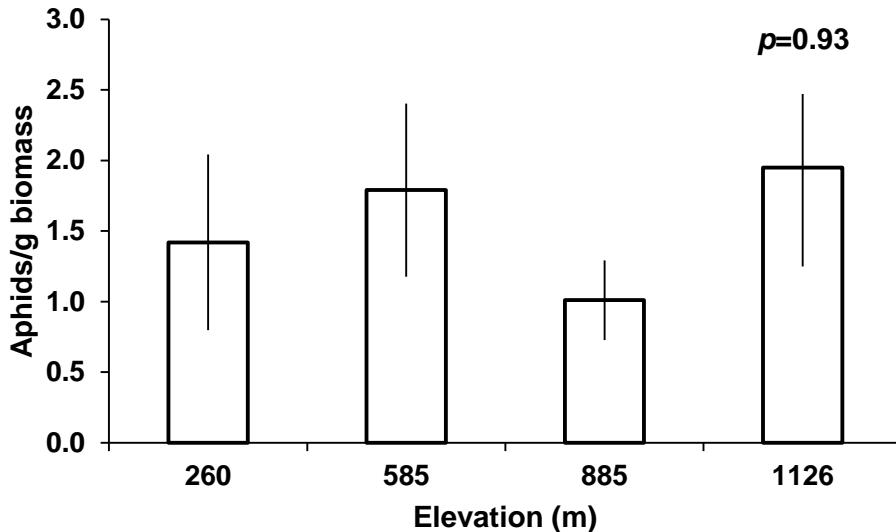
Table 6 F ratio, p value, and degrees of freedom ( $df$ )<sup>a</sup> for the effects of host-plant genotype on feeding guild distribution within elevations.  $n = 48$  (Proc GLM).

### *Uroleucon nigrotuberculatum*

Aphid abundance far exceeded that of any other morphospecies with 3,711 aphids quantified from vacuum and visual methods. Though two morphotypes were collected, the overwhelming abundance was that of the specialist, *Uroleucon nigrotuberculatum*. There was no elevation effect on aphid abundance or aphid abundance/g biomass (Figs. 12, 13, Table 7), while genotype had a significant effect on these abundance measures (Figs. 14, 15). Genotype 8 supported the highest absolute aphid abundance (Fig. 14), and genotype 16 supported the highest aphid abundance/g biomass (Fig. 15).



**Fig. 12** Effect of elevation on absolute aphid abundance (mean $\pm$ SE;  $p \leq 0.1$  considered significant).



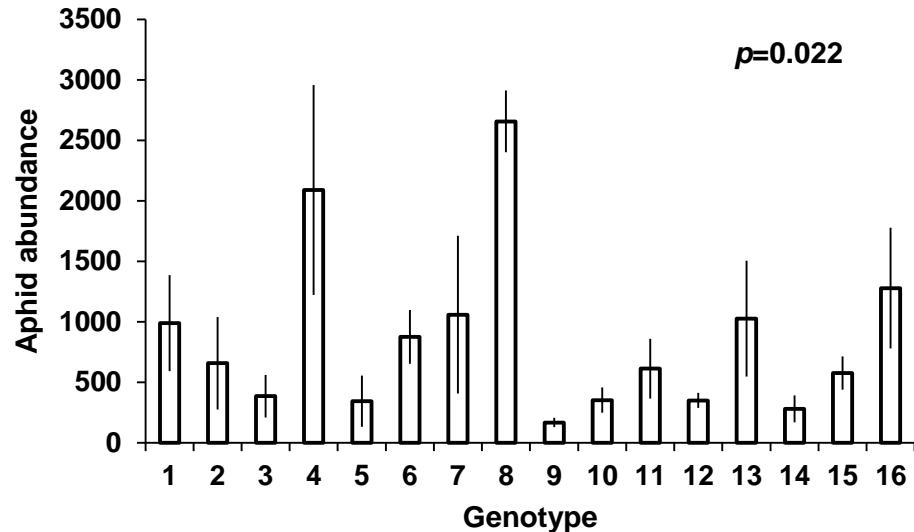
**Fig. 13** Effect of elevation on aphid abundance/g biomass (mean±SE;  $p \leq 0.1$  considered significant).

**Table 7**  $F$  ratio,  $p$  value, and degrees of freedom ( $df$ )<sup>a</sup> for the effects of host-plant elevation and genotype on aphid abundance.  $n = 48$  (Proc Nested).

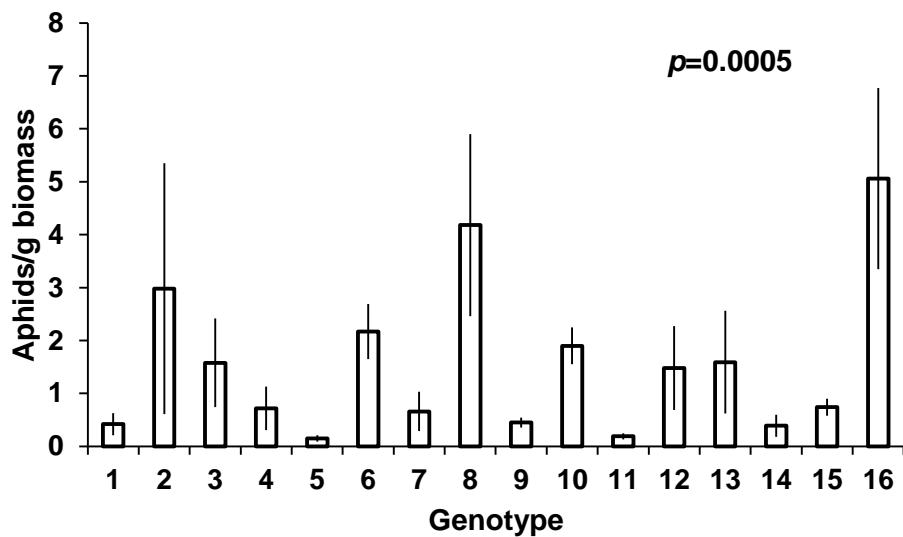
	Elevation		Genotype	
	<i>F</i>	<i>p</i>	<i>F</i>	<i>p</i>
Total aphids	1.14	0.3714	2.43	<b>0.0224</b>
Aphids/g biomass	0.014	0.9337	4.29	<b>0.0005</b>

**Note:**  $p \leq 0.05$  and  $0.05 \leq p \leq 0.1$  presented in bold text.

<sup>a</sup> Elevation  $df = 3, 32$  Genotype  $df = 12, 32$ .

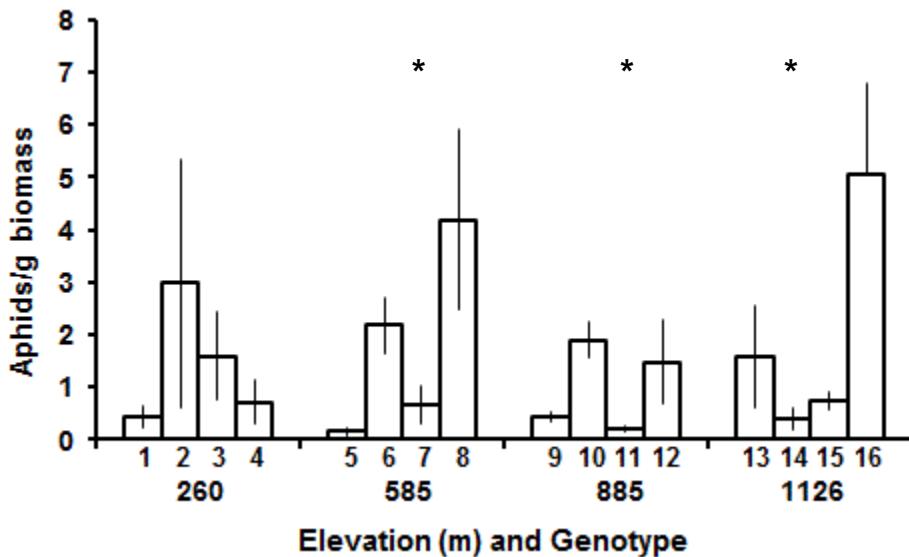


**Fig. 14** Effect of genotype on absolute aphid abundance (mean $\pm$ SE;  $p \leq 0.1$  considered significant).



**Fig. 15** Effect of genotype on aphid abundance/g biomass (mean $\pm$ SE;  $p \leq 0.1$  considered significant).

Genotype affected aphid abundance/g biomass in all but the lowest elevation (Fig. 16, Table 8). Within elevation 585 m, genotype 8 supported the highest aphid abundance/g biomass. Among plants from 885 m, genotypes 10 and 12 supported a significantly higher number of aphids compared to genotype 11. Among plants from 1126 m, genotypes 14 and 16 supported the lowest and highest aphid abundance/g biomass respectively.



**Fig. 16** Effect of genotype (1-16) on aphid abundance/g biomass (mean $\pm$ SE) within elevations (260 m, 585 m, 885 m, 1126 m). \* = significant ( $p \leq 0.1$ ) genotype effect within elevation.

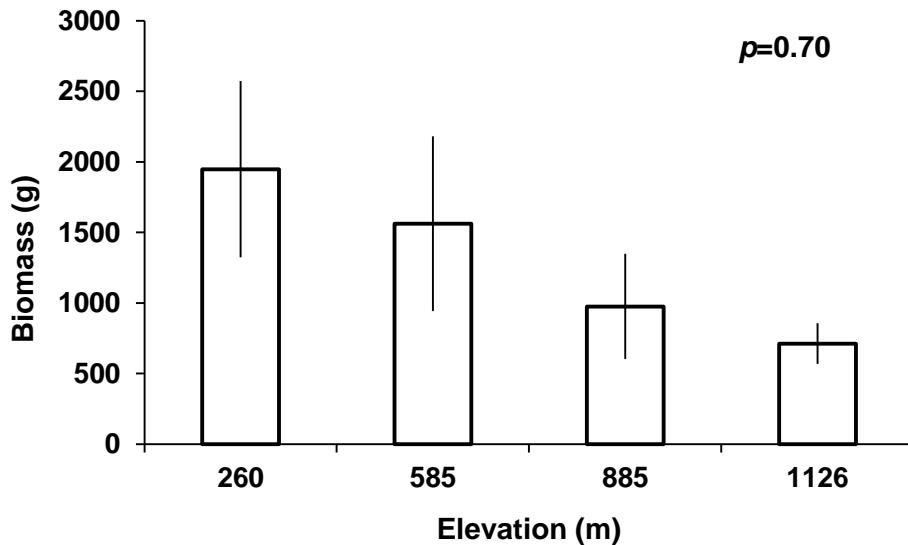
	260 m		585 m		885 m		1126 m	
	F	p	F	p	F	p	F	p
Total aphids	1.85	0.2162	3.80	<b>0.0581</b>	1.75	0.2336	2.06	0.1846
Aphids/g biomass	0.72	0.5691	11.55	<b>0.0028</b>	9.40	<b>0.0053</b>	5.26	<b>0.0269</b>

<sup>a</sup> Note:  $p \leq 0.05$  and  $0.05 \leq p \leq 0.1$  presented in bold text.  
<sup>a</sup>  $df = 3, 8$ .

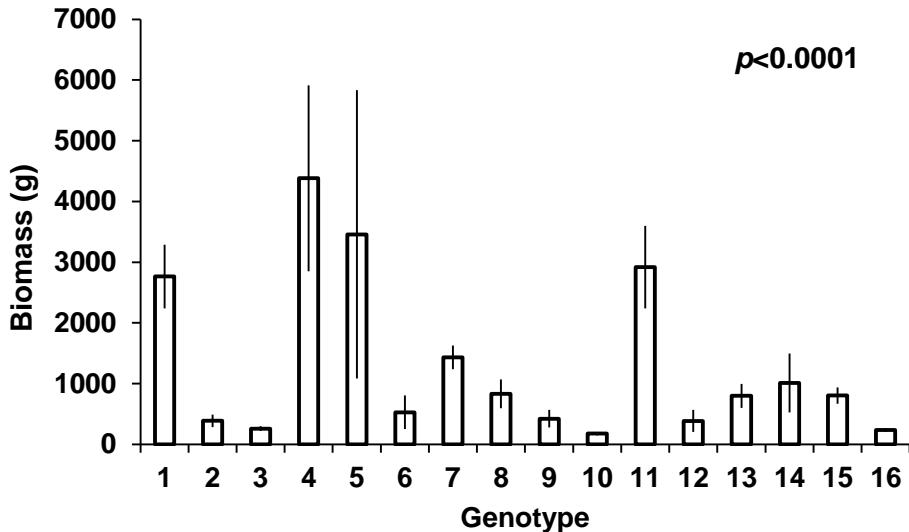
Table 8 F ratio, p value, and degrees of freedom ( $df^a$ ) for the effect of host-plant genotype on aphid abundance within elevations.  $n = 48$  (Proc GLM).

## Biomass

There was no significant difference in aboveground biomass production between plants from different elevations; however, there was a trend where plants from lower elevations had higher aboveground biomass production (Fig. 17). There was a highly significant genotype ( $p < 0.0001$ ) effect on total aboveground biomass production (Fig. 18), and a significant genotype effect on total aboveground biomass production within all elevations except 585 m (data not shown).



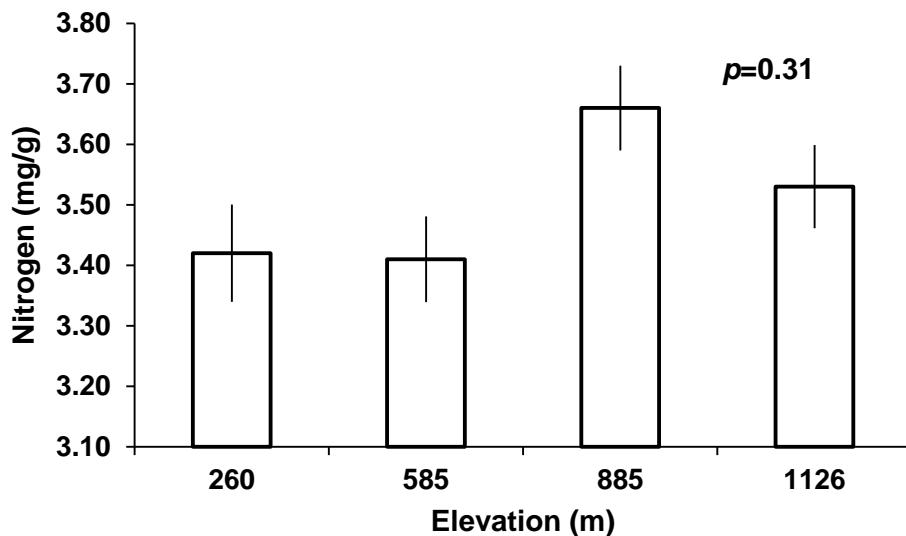
**Fig. 17** Effect of elevation on total aboveground biomass production (mean $\pm$ SE;  $p \leq 0.1$  considered significant).



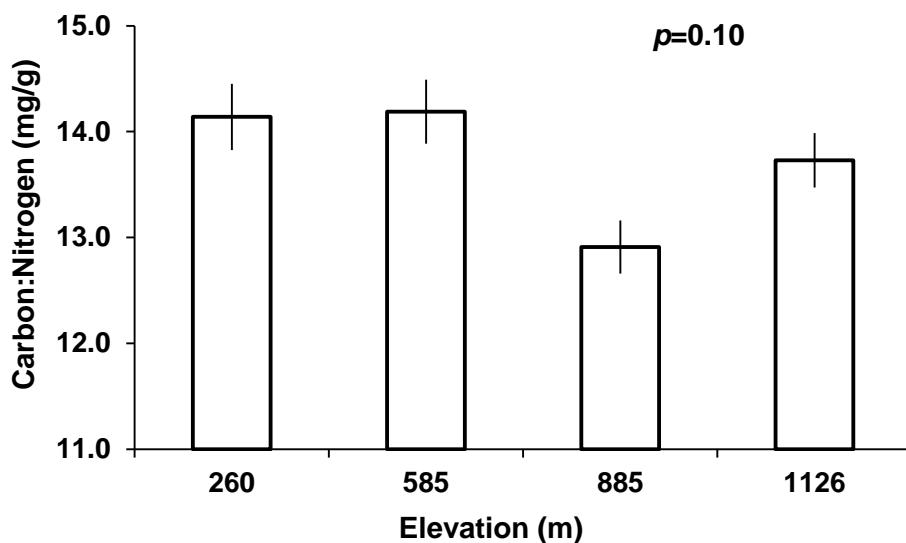
**Fig. 18** Effect of genotype on total aboveground biomass production (mean $\pm$ SE;  $p \leq 0.1$  considered significant).

### Foliar chemistry

Though not significant, there was a trend for higher foliar N at higher elevations (Fig. 19). There was a marginally significant effect of elevation on foliar CN concentration (Fig. 20, Table 9). Foliar N and CN were both significantly affected by genotype (Table 9). Genotype 9 had the highest foliar N and lowest CN concentrations, while genotypes 3 and 6 had the lowest foliar N and highest CN concentrations (Figs. 21, 22). There was a genotype effect on foliar N within elevations 260 m and 885 m, and on CN concentrations among plants from 885 m (Table 10).



**Fig. 19** Effect of elevation on foliar Nitrogen concentrations (mean $\pm$ SE;  $p \leq 0.1$  considered significant).



**Fig. 20** Effect of elevation on foliar Carbon:Nitrogen (mean $\pm$ SE;  $p \leq 0.1$  considered significant).

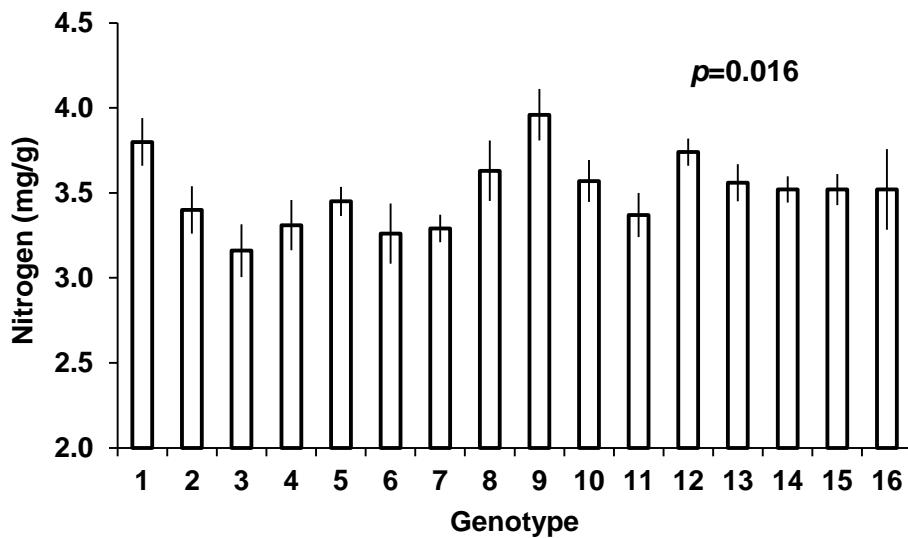
**Table 9** *F* ratio, *p* value, degrees of freedom (*df*)<sup>a</sup> and *n* for the effects of host-plant elevation and genotype on foliar variables (Proc Nested).

	Elevation		Genotype	
	<i>F</i>	<i>p</i>	<i>F</i>	<i>p</i>
Water (%)	4.87	<b>0.0193</b>	1.43	0.1511
Specific leaf weight	0.30	0.8223	1.59	<b>0.0939</b>
Nitrogen	1.33	0.3112	2.19	<b>0.0159</b>
Carbon:Nitrogen	2.59	<b>0.1010</b>	1.81	<b>0.0526</b>

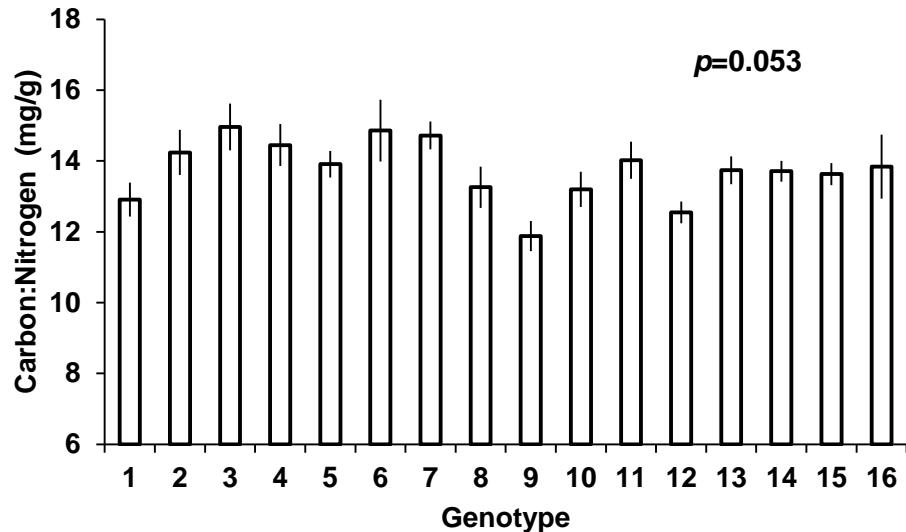
**Note:**  $p \leq 0.05$  and  $0.05 \leq p \leq 0.1$  presented in bold text.

<sup>a</sup> Foliar characteristics: Elevation  $df=3, 272$  Genotype  $df=12, 272$ .  $n = 288$ .

Foliar chemistry: Elevation  $df=3, 128$  Genotype  $df=12, 128$ .  $n = 144$ .



**Fig. 21** Effect of genotype on foliar Nitrogen concentrations (mean±SE;  $p \leq 0.1$  considered significant).

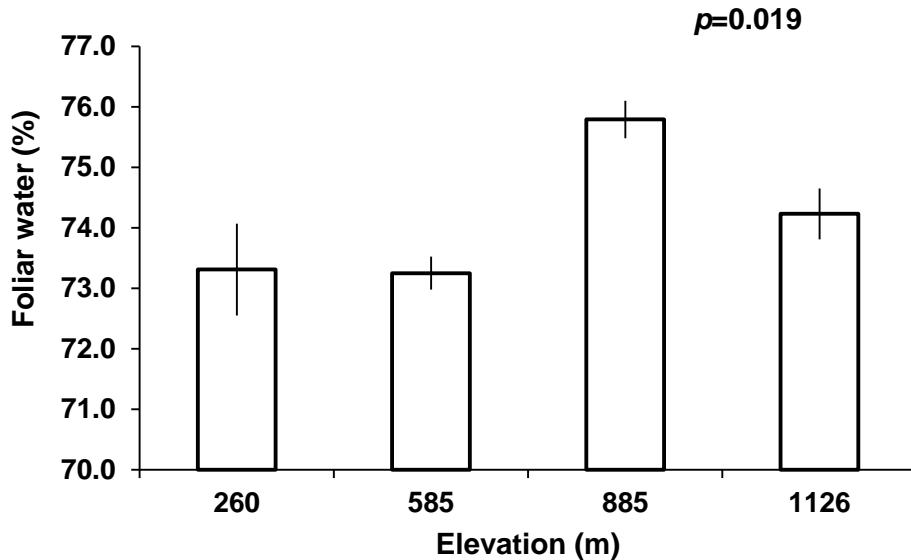


**Fig 22.** Effect of genotype on foliar Carbon:Nitrogen (mean $\pm$ SE;  $p \leq 0.1$  considered significant).

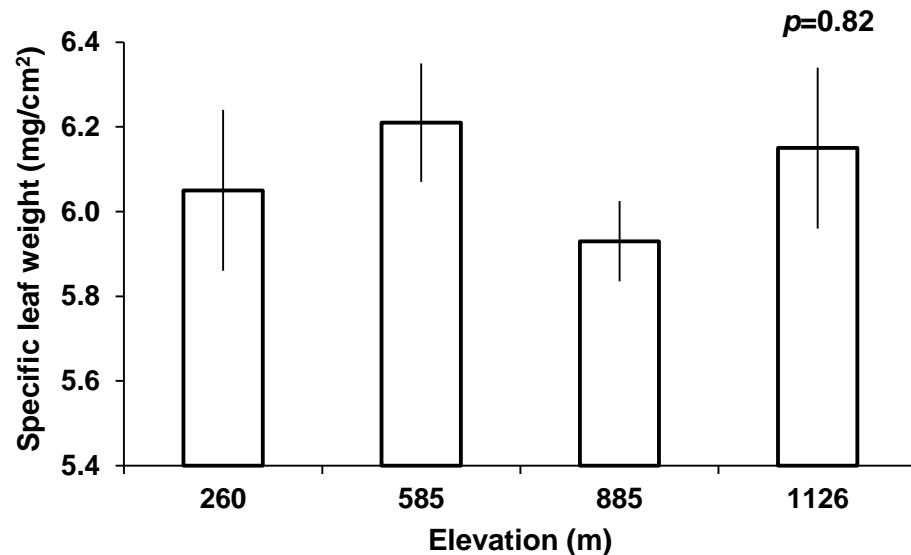
	<i>F</i>	<i>p</i>	<i>F</i>	<i>p</i>	<i>F</i>	<i>p</i>	<i>F</i>	<i>p</i>
260 m			585 m		885 m		1126 m	
Water (%)	0.89	0.4531	3.43	<b>0.0219</b>	2.54	<b>0.0636</b>	2.37	<b>0.0780</b>
Specific leaf weight	0.08	0.9722	6.23	<b>0.0008</b>	5.38	<b>0.0022</b>	1.01	0.3916
Nitrogen	3.48	<b>0.0270</b>	1.56	0.2190	4.07	<b>0.0147</b>	0.02	0.9967
Carbon:Nitrogen	2.15	0.1132	1.59	0.2120	4.19	<b>0.0131</b>	6.03	0.9938
<b>Note:</b> $p \leq 0.05$ and $0.05 \leq p \leq 0.1$ presented in bold text.								
Foliar characteristics: $df = 3, 32, n = 144$ .								
<sup>a</sup> Foliar chemistry: $df = 3, 68, n = 288$ .								

**Table 10** *F* ratio, *p* value, degrees of freedom ( $df^a$ ) and *n* for the effect of host-plant genotype on foliar characteristics within elevations (Proc GLM).

There was a significant elevation effect on foliar water content but not SLW (Figs. 23, 24, Table 9). Plants from 885 m had the highest foliar water content while plants from the two lowest elevations had the lowest foliar water content (Fig. 23). Genotype significantly affected SLW but not foliar water content (Table 9).

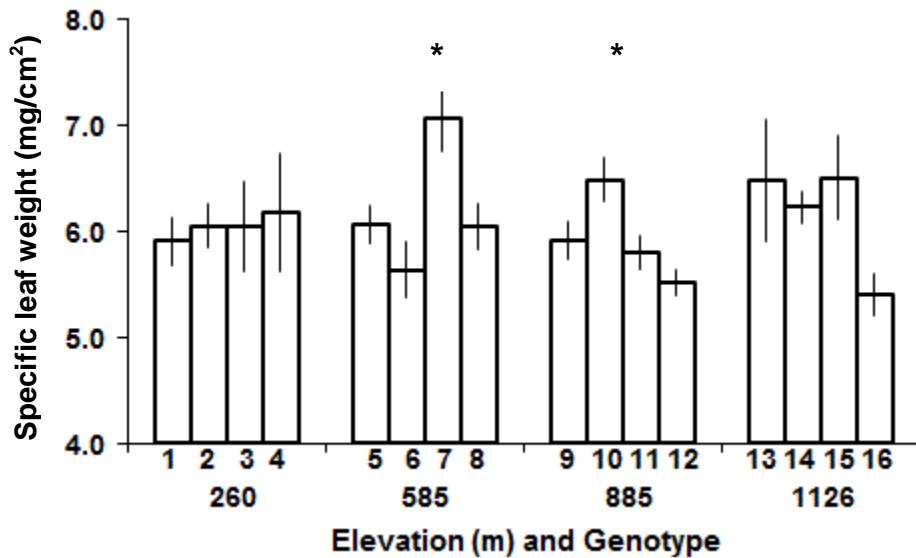


**Fig. 23** Effect of elevation on foliar water content (mean $\pm$ SE;  $p \leq 0.1$  considered significant).



**Fig. 24** Effect of elevation on specific leaf weight (mean $\pm$ SE;  $p \leq 0.1$  considered significant).

Genotype affected foliar water content in all but the lowest elevation, 260 m (Table 10). There was a significant genotype effect on SLW within the two middle elevations, 585 m and 885 m (Fig. 25, Table 10).



**Fig. 25.** Effect (mean $\pm$ SE) of host-plant genotype (1-16) on specific leaf weight (SLW) within elevations (260 m, 585 m, 885 m, 1126 m). \* = significant ( $p \leq 0.1$ ) genotype effect within elevations.

## Terpenes

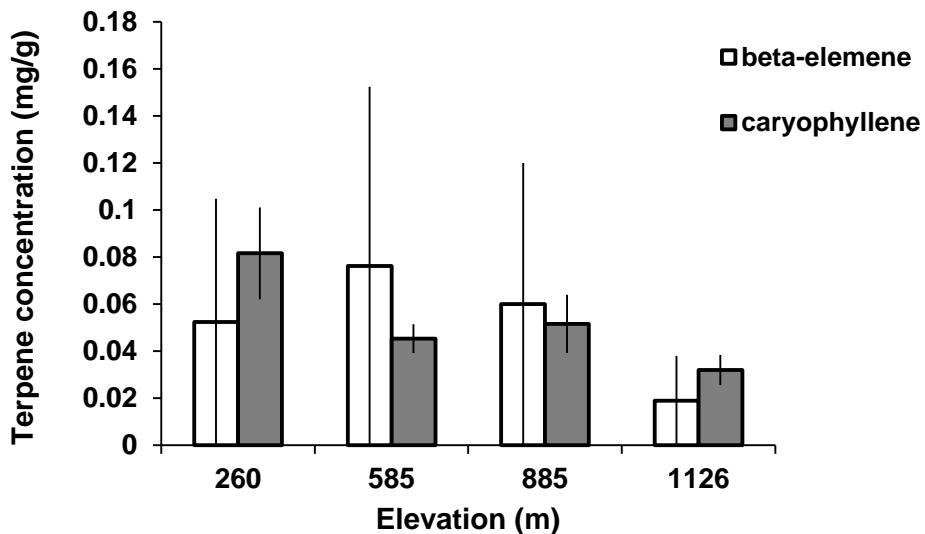
Elevation significantly affected the production of two terpenes:  $\beta$ -elemene and caryophyllene (Table 11). Plants from 260 m and 585 m produced the lowest and highest amount of  $\beta$ -elemene respectively, while the reverse was true for caryophyllene (Fig. 26). There was a significant genotype effect on the production of  $\alpha$ -pinene,  $\beta$ -pinene, limonene, and  $\gamma$ -elemene (Table 11). Total terpene production did not vary between elevations or genotypes; however, there was a trend for higher foliar terpene concentrations at lower elevations (Fig. 27).

**Table 11** *F* ratio, *p* value, and degrees of freedom (*df*)<sup>a</sup> for the effect of host-plant elevation and genotype on terpene production. *n* = 96 (Proc Nested).

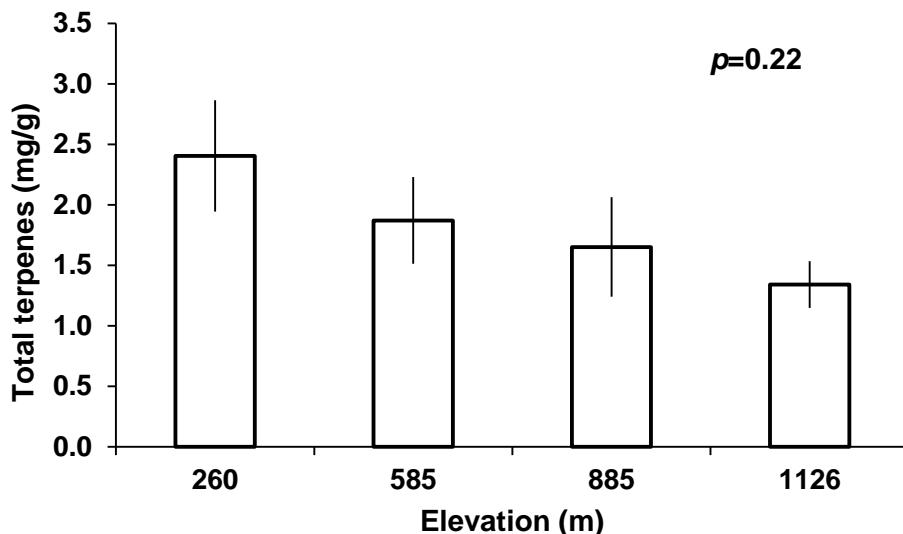
Compound	Elevation		Genotype	
	<i>F</i>	<i>p</i>	<i>F</i>	<i>p</i>
$\alpha$ -pinene	0.03	0.9928	2.00	<b>0.0344</b>
$\beta$ -pinene	0.67	0.5891	1.87	<b>0.0509</b>
Limonene	0.76	0.5385	3.37	<b>0.0005</b>
Bornyl acetate	0.05	0.9861	1.51	0.1373
$\beta$ -elemene	5.03	<b>0.0174</b>	0.78	0.6657
Caryophyllene	2.70	<b>0.0928</b>	1.08	0.3911
Germacrene D	2.03	0.1640	0.92	0.5316
$\gamma$ -elmemene	0.80	0.5189	2.14	<b>0.0232</b>
Total Terpenes	1.69	0.2227	0.88	0.5718

**Note:**  $p \leq 0.05$  and  $0.05 \leq p \leq 0.1$  presented in bold text.

<sup>a</sup> Elevation *df* = 3, 79 Genotype *df* = 12, 79.



**Fig. 26** Effect of elevation on beta-elemene (*p* = 0.018) and caryophyllene (*p* = 0.093) production (mean±SE; *p* ≤ 0.1 considered significant).



**Fig. 27** Effect of elevation on total terpene production (mean $\pm$ SE;  $p \leq 0.1$  considered significant).

Depending on the compound, there was a significant genotype effect on terpene production within all elevations, but no compound differed among genotypes at all elevations (Table 12). Four compounds,  $\alpha$ -pinene, limonene, bornyl acetate, and  $\gamma$ -elemene, were affected by genotype among plants from 260 m (Table 12). Among plants from 585 m  $\beta$ -pinene, limonene, and bornyl acetate were affected by genotype. Only  $\gamma$ -elemene was affected by genotype among plants from 885 m. Genotype affected  $\beta$ -elemene, caryophyllene, and Germacrene D among plants from 1126 m. There was no genotype effect on total terpene production within any elevation.

**Table 12** *F* ratio, *p* value, and degrees of freedom ( $df$ )<sup>a</sup> for the effect of host-plant genotype on terpene production within elevations.  $n = 96$  (Proc GLM)

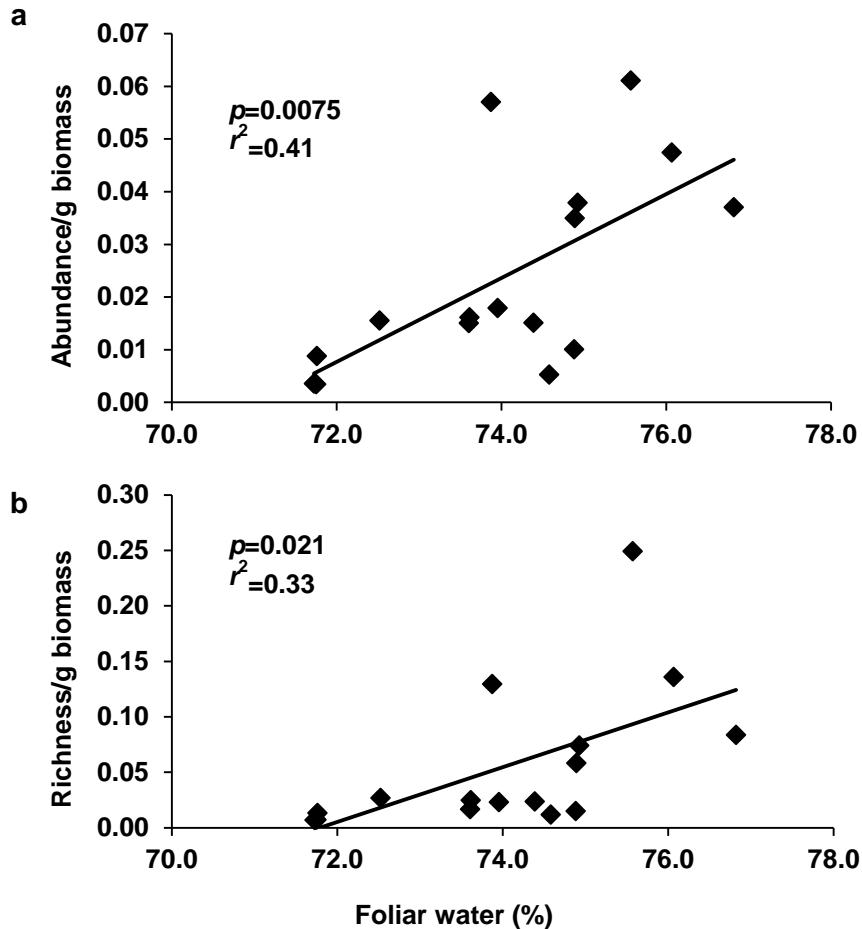
Compound	260 m		585 m		885 m		1126 m	
	<i>F</i>	<i>p</i>	<i>F</i>	<i>p</i>	<i>F</i>	<i>p</i>	<i>F</i>	<i>p</i>
$\alpha$ -pinene	3.91	<b>0.0239</b>	0.81	0.5017	1.55	0.2321	1.78	0.1840
$\beta$ -pinene	0.78	0.5192	8.07	<b>0.0010</b>	1.00	0.4129	2.28	0.1108
Limonene	6.89	<b>0.0023</b>	5.51	<b>0.0064</b>	0.91	0.4535	1.34	0.2889
Bornyl acetate	2.52	<b>0.0873</b>	2.80	<b>0.0666</b>	1.06	0.3894	2.15	0.1260
$\beta$ -elemene	1.01	0.4105	0.38	0.7691	0.57	0.6394	3.62	<b>0.0309</b>
Caryophyllene	1.24	0.3217	0.26	0.8552	0.35	0.7896	4.39	<b>0.0158</b>
Germacrene D	1.41	0.2683	0.51	0.6788	0.34	0.7997	3.21	<b>0.0450</b>
$\gamma$ -elemene	8.41	<b>0.0008</b>	1.75	0.1883	7.42	<b>0.0016</b>	0.90	0.4591
Total Terpenes	1.47	0.2540	0.48	0.7001	0.40	0.7520	1.61	0.2193

Note:  $p \leq 0.05$  and  $0.05 \leq p \leq 0.1$  presented in bold text.

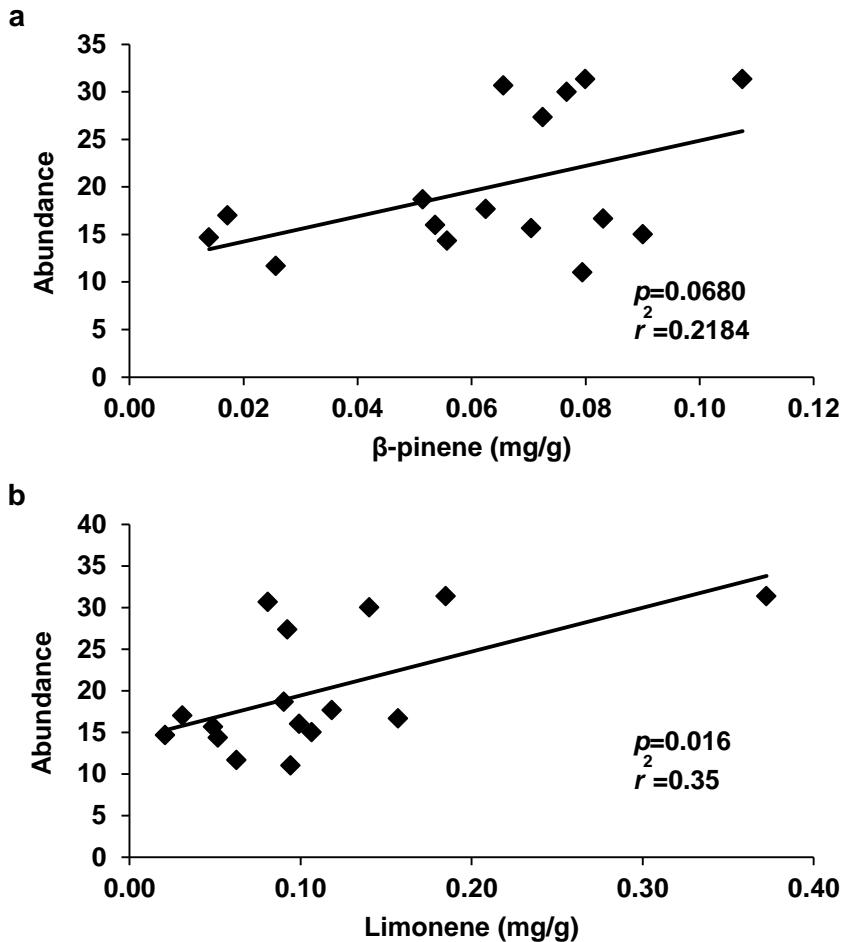
<sup>a</sup>  $df = 3, 19$ .

### Relationship between leaf and insect community measures

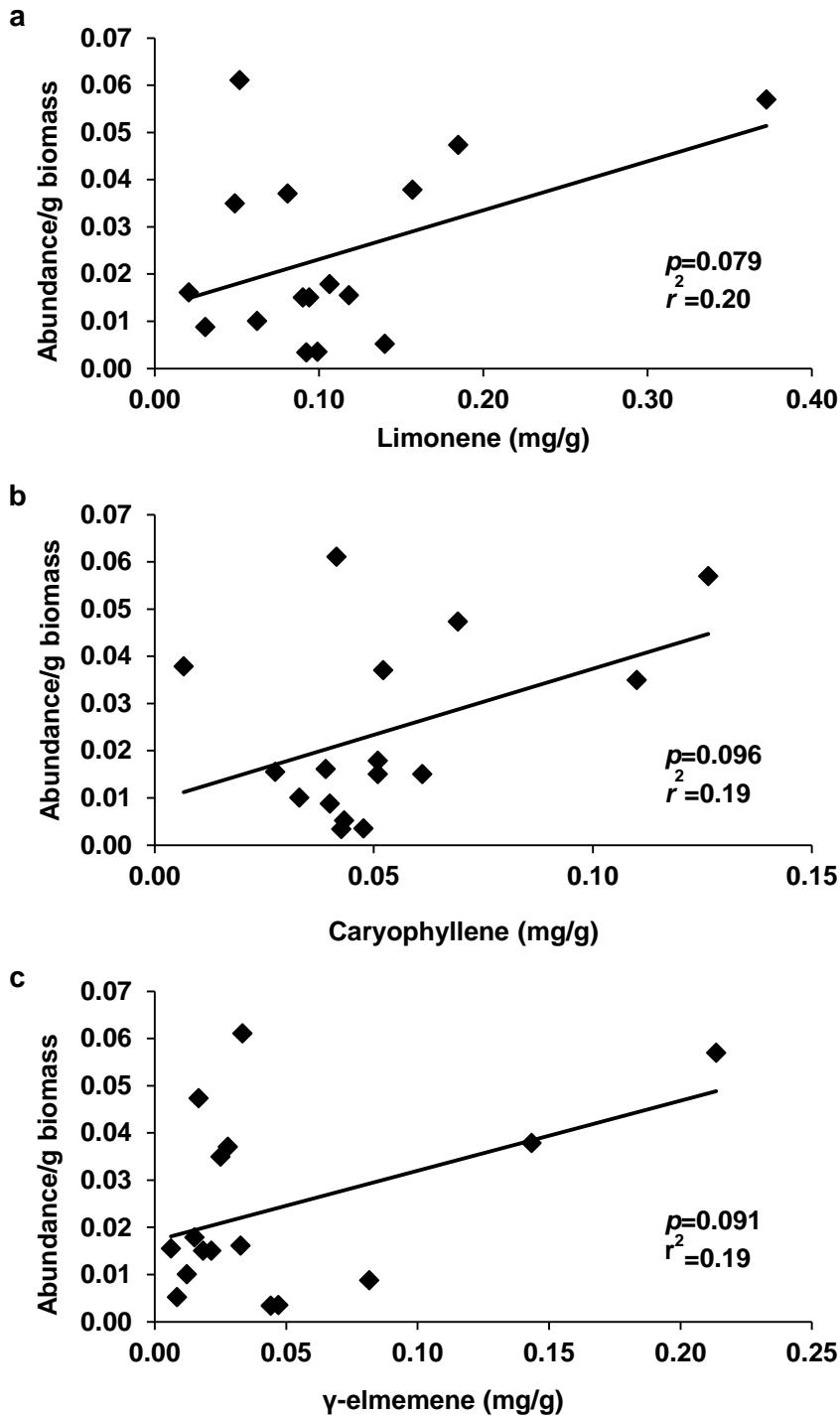
Among genotypes, abundance/g biomass and richness/g biomass were positively related to foliar water content (Fig. 28a, b). Among genotypes, foliar terpene concentrations were positively related to several community measures. Abundance was related to  $\beta$ -pinene and limonene (Fig. 29a, b), abundance/g biomass was related to limonene, carophyllene, and  $\gamma$ -elemene (Fig. 30a, b, c), and richness was related to foliar limonene concentration (Fig. 31).



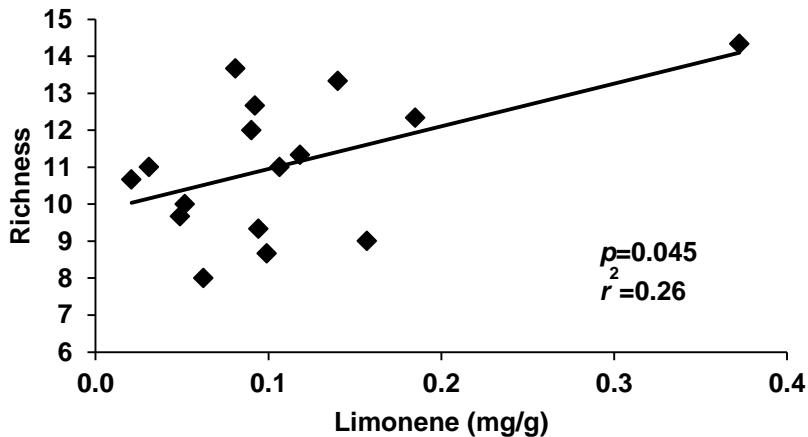
**Fig. 28** Relationship between genotype and foliar water content for the dependent variables (a) abundance/g biomass, and (b) richness/g biomass. Points represent genotype means ( $n=16$ ;  $p \leq 0.1$  considered significant).



**Fig. 29** Relationship between genotype and (a) beta-pinene, and (b) limonene concentration for the dependent variable abundance. Points represent genotype means ( $n=16$ ;  $p \leq 0.1$  considered significant).

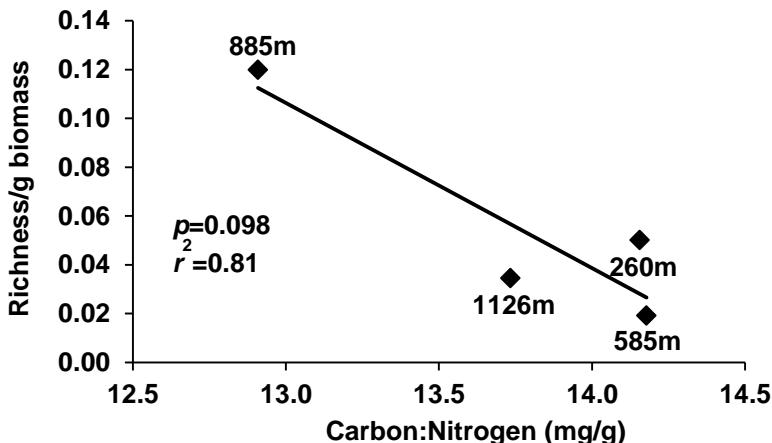


**Fig. 30** Relationship between genotype and (a) limonene, (b) caryophyllene and (c)  $\gamma$ -elemene concentration for the dependent variable abundance/g biomass. Points represent genotype means ( $n=16$ ;  $p \leq 0.1$  considered significant).

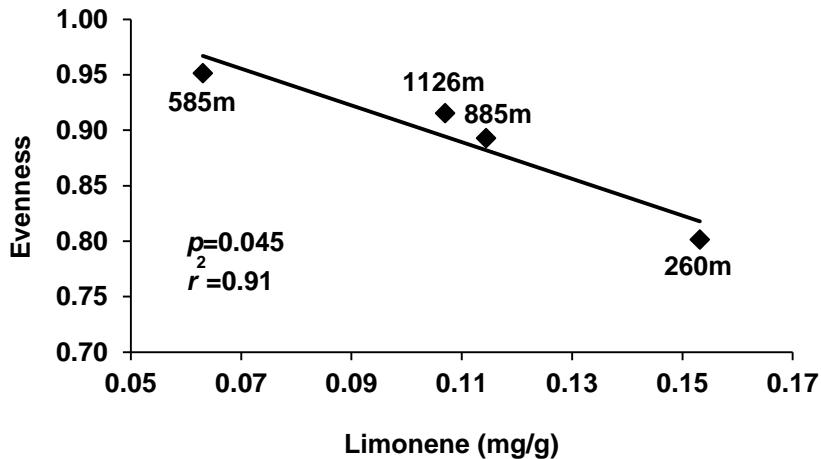


**Fig. 31** Relationship between genotype, foliar limonene concentration, and the dependent variable, richness. Points represent genotype means ( $n=16$ ;  $p \leq 0.1$  considered significant).

Among elevations, richness/g biomass was negatively related to foliar CN, where plants from the two lowest elevations had the highest CN ratios and lowest richness/g biomass (Fig. 32). Evenness was negatively related to limonene, where the lowest elevation had the most dissimilar composition of morphospecies (Fig. 33).



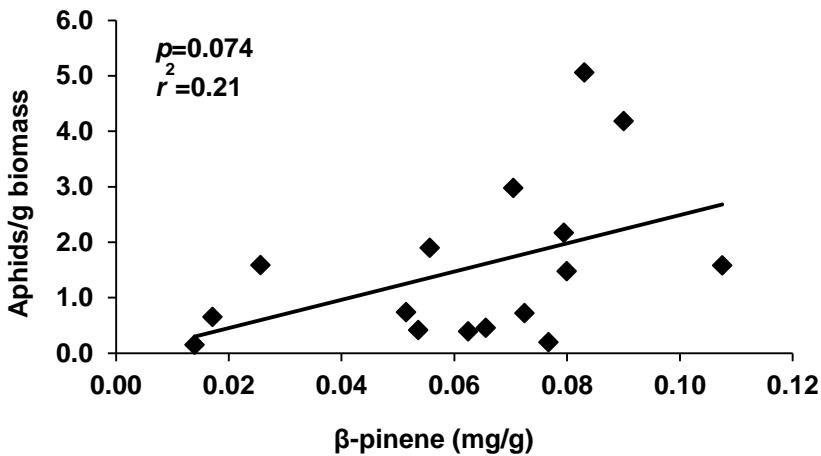
**Fig. 32** Relationship between elevation, foliar Carbon:Nitrogen, and the dependent variable, richness/g biomass. Points represent elevation means ( $n = 4$ ;  $p \leq 0.1$  considered significant).



**Fig. 33** Relationship between elevation, limonene concentration, and the dependent variable, evenness. Points represent elevation means ( $n = 4$ ;  $p \leq 0.1$  considered significant).

#### Relationship between leaf measures and *U. nigrotuberculatum* abundance

For genotype, only a single variable was significantly related to aphid measures, where abundance/g biomass increased with  $\beta$ -pinene concentration (Fig. 34).

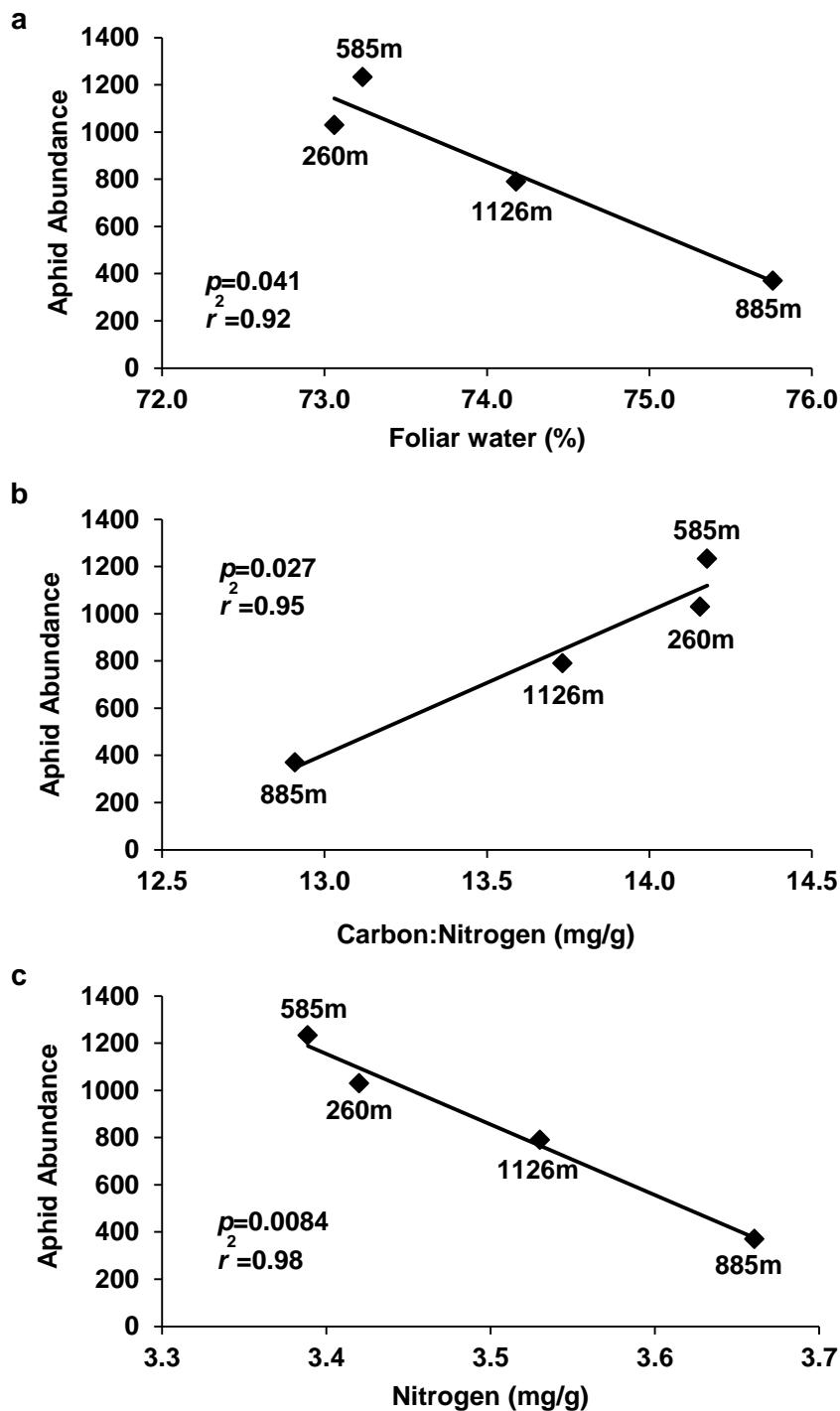


**Fig. 34** Relationship between genotype, foliar beta-pinene concentration, and the dependent variable, aphids/g biomass. Points represent genotype means ( $n = 16$ ;  $p \leq 0.1$  considered significant).

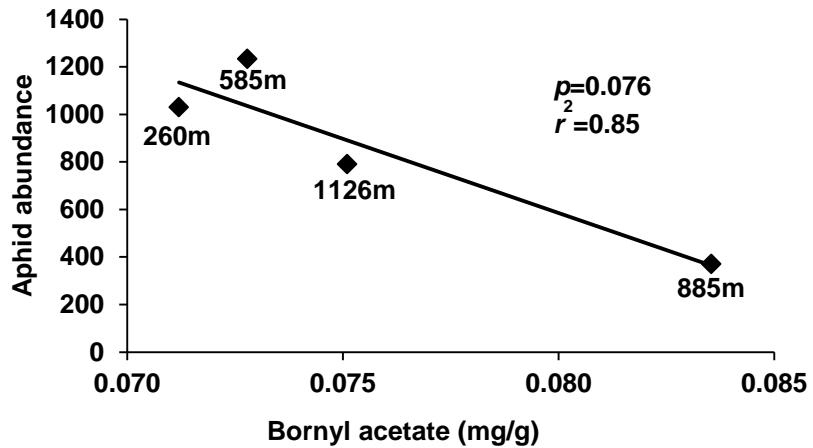
For elevation, there was a strong negative relationship between foliar water content and aphid abundance, where plants from the two lowest elevations had the lowest foliar water content and the highest aphid abundances (Fig. 35a). There was a strong positive

relationship between aphid abundance and foliar CN by elevation, with plants from the two lowest elevations having the highest CN ratios and aphid abundances (Fig. 35b). There was a negative relationship between aphid abundance and foliar N content, where plants from the two lowest elevations had the lowest foliar N concentrations and highest aphid abundances (Fig. 35c).

Among elevations, bornyl acetate was the only terpene related to aphid abundance, where plants from the lowest elevations produced the least amount of bornyl acetate and supported the highest number of aphids (Fig. 36).



**Fig. 35** Relationship between elevation and foliar (a) water content, (b) Carbon:Nitrogen, and (c) Nitrogen, for the dependent variable aphid abundance. Points represent elevation means ( $n = 4$ ;  $p \leq 0.1$  considered significant).



**Fig. 36** Relationship between elevation, bornyl acetate concentration, and the dependent variable aphid abundance. Points represent elevation means ( $n = 4$ ;  $p \leq 0.1$  considered significant).

## DISCUSSION

I employed a common garden approach to assess the potential for host-plant genetic and environmental variation to structure an associated insect community and influence the colonization of a dominant herbivore species. Because plant and insect communities are tightly linked, genetic and environmental variation between host-plants in traits important to insects, such as foliar quality or defensive compounds, are expected to influence the associated insect community. My study contributes to a broader understanding of the factors structuring insect communities by focusing on how genetic and, potentially, environmental variation in a plant population effects communities and colonization. Understanding the factors that drive plant-insect interactions is especially important because this association can affect a number of ecosystem processes. Additionally, examining the effects of population diversity on associated communities may have conservation implications where maintaining a high level of population genetic diversity may be just as important to the associated community as the level of interspecific diversity in a system. The widespread distribution and dominance of *S. altissima* and its interaction with a diverse insect community makes this foundation species particularly relevant to addressing questions of the factors structuring insect communities.

My results show that both host-plant genotype and native environment affected important plant and insect measures. The strong genotype effect on the aphid, *U. nigrotuberculatum*, suggests that genetic variation between host-plants may be particularly influential in colonization by specialist insects. Several plant characteristics such as foliar

water, CN, and terpene concentration were related to insect community and aphid measures, but drawing clear conclusions about mechanisms driving insect community dynamics remains difficult due to the limited strength of these relationships. Observed genotype effects on plant and insect measures within certain elevations suggest that (1) environment may be more important than variation across genotypes, and (2) plants express plasticity in physiological responses when grown outside of their native environment.

A key question in my study asked if host-plant genotype had an effect on insect community abundance, richness, or evenness. Because a diverse insect community relies on the dominant host-plant *S. altissima* and the potential for a high level of genetic variation in natural *S. altissima* populations exists, I expected that insects would preferentially choose certain host-plants based on genotype. In addition, because the native environment in which a plant develops may impose conditions affecting ecologically important traits like foliar quality, I expected some influence of environment on host-plant choice. This preferential host-plant selection would result in certain genotypes in the common garden supporting a higher abundance and diversity of insects, as well as affecting species distribution (i.e., evenness). In addition to the entire insect community, I expected similar responses by the specialist aphid that dominated my insect samples.

The result that richness/g biomass differed significantly between genotypes provides evidence that insect community composition may be affected by host-plant genotype as reported by others (Whitham et al. 1994; Dungey et al. 2000; Crutsinger et al. 2008b). Crutsinger et al. (2008b) observed a similar community level response where insect community richness varied nearly two-fold between distinct *S. altissima* genotypes. Herbivore/g biomass was affected by genotype, an expected result due to the direct

relationship between this feeding guild and their host-plants. This result also suggests that host plant genotype does not affect higher trophic levels (but see Schädler et al. 2010).

Genotype strongly affected the aphid (*U. nigrotuberculatum*) population. For example, there was over a 30-fold difference between the genotypes with the highest and lowest aphids/g biomass. This result suggests that aphids selectively chose host-plants based on genetic variation between plants. As a *Solidago* specialist, it is not uncommon for *U. nigrotuberculatum* to carry out an entire life cycle on a single plant (Pilson and Rausher 1995), making host-plant selection particularly crucial for this species. Johnson (2008) demonstrated the importance of host-plant genotype identity in an evening primrose population to the specialist aphid, *Aphis oestlundii*, with aphid densities ranging 75-fold among plant genotypes. Considering the observed effects of genotype on the insect community and aphid population suggest that host-plant genotype does structure associated insect communities, but that the magnitude of this effect may be mediated by community composition, where certain species in a community are more reliant on host-plant genotype than others.

A number of plant constituents important to insects were affected by genotype, providing potential mechanisms for observed differences in insect community structure between host-plant genotypes. Aboveground biomass, SLW, foliar N, and CN were all significantly affected by genotype. Foliar chemistry has been shown to vary among genotypes in an oak tree (*Quercus laevis*) population (Madritch and Hunter 2002, 2005), providing support for the genotype effect on foliar N and CN that I observed. For volatile terpenes, foliar concentrations of  $\alpha$ -pinene,  $\beta$ -pinene, limonene, and  $\gamma$ -elemene also varied between genotypes. Semiz et al. (2007) examined variation in the terpene profiles between

nine Scots pine (*Pinus sylvestris*) populations and found evidence that both the presence and concentration of terpenoids was regulated by a genetic component. However, the authors did emphasize the role of environment in driving genetic adaptation in terpene profiles.

Differences between genotypes in these foliar variables provide evidence that a genetic component influences the expression of certain plant characteristics that are important to the insect community. The observed genotype effect on insect and plant measures supports the idea that variation at a small spatial scale, such as between plant clusters in the same field, influences insect community structure.

When investigating the potential for host-plant genotype to influence insect community structure, one must also recognize the role environment plays in regulating phenotypic expression. For example, Maddox and Cappuccino (1986) found that the susceptibility of *S. altissima* genotypes to aphid population growth was dependent upon water availability, where aphid abundance differed among genotypes only in the high water treatment. Additionally, Rossi and Stilling (1998) found a significant difference in the number of galls initiated on distinct genotypes of the sea daisy (*Borrichia furtescens*), suggesting a genetic component in sea daisy susceptibility to the gall fly (*Asphondylia borrichiae*). However, when sea daisy populations were exposed to variable abiotic conditions, the authors found that certain genotypes became more susceptible to the gall fly in shaded environments. These studies demonstrate how environment can regulate host-plant phenotype to affect the structure of dependent communities.

A second question of my study asked if insect community structure varied between host-plants collected from different native environments. A statistically significant elevation effect suggests that the abiotic forces in a plant's native environment impose inherent

developmental or physiological responses to a new environment, causing a plant to react differently than genotypes collected from other sites.

Insect community abundance was significantly affected by host-plant elevation. Herbivores were the only feeding guild affected by elevation, but herbivore/g biomass was not suggesting that herbivores responded to differences in biomass between plants from different elevations. Aphid abundance did not vary between plants from different elevations, suggesting that environment does not play a key role in host-plant selection by *U. nigrotuberulatum*. This further supports the proposed idea that aphids may rely more heavily on host-plant genotype during colonization. Interestingly, plants from elevation 885 m supported the highest insect community abundance and the lowest aphid abundance, while plants from 585 m supported the lowest insect community abundance and the highest aphid abundance (Figs. 3, 11), which may further support that the *U. nigrotuberulatum* population and rest of the insect community rely on different cues when selecting a host-plant.

While there was no significant elevation effect on aboveground biomass production, there was a trend for biomass production to decrease as elevation increased. This is in contrast with the prediction that plants at higher elevations allocate more resources towards growth to cope with shorter growing seasons (Olsson and Ågren 2002). Confounding such a response is that in my common garden experiment all plants experienced the same abiotic conditions, which are not necessarily reflective of their native habitat. Thus, my experiment provides evidence of plasticity in plant physiological responses where plants from higher elevations may allocate resources to underground growth, such as fine roots, enabling more efficient nutrient uptake in environments that often have slower rates of nutrient turnover (Oleksyn et al. 1998).

There was a marginally significant elevation effect on foliar CN concentrations, where foliar quality increased with elevation. This result is consistent with the established trend that plants from higher elevations also have higher foliar quality (Kröner 1989). Additionally, this evidence further supports the hypothesis that plants collected from the highest elevations allocated resources to underground growth, effectively increasing foliar quality through higher rates of nutrient uptake by underground structures. Despite being grown outside of their native environment, my results suggest that plant characteristics important to insects, like foliar quality, are, in part, inherent in the environment, providing evidence for an environmental contribution to insect community structure.

Water content was significantly affected by elevation. Plants collected from 885 m had the highest foliar water content and also supported the highest insect abundances (Figs. 3, 21). This suggests that insect community abundance may be a consequence of environmental variation in foliar characteristics between individuals within a plant population.

Terpenoids are carbon-based defensive compounds and are potentially greatly influenced by both abiotic (Langenheim 1994; Glynn et al. 2003) and genetic variation (Dungey et al. 2000).  $\beta$ -elemene and caryophyllene concentrations were terpenes significantly affected by elevation. There was a trend for plants from lower elevations to have higher total terpene concentrations. This is consistent with the idea that plants at lower elevations are under increased selective pressure from the herbivore community (Hodkinson 2005; Garibaldi et al. 2011) and allocate more resources to defense (Karban 2011). Additionally, this is supported by the finding that plants from lower elevations had higher foliar CN, and, thus, were of lower nutritional quality. Several of these foliar measures were

also significantly different between genotypes. Though my study design did not explicitly allow me to separate variation among genotypes from those of environment, the nested ANOVA clearly demonstrated a combination of these factors effecting important plant constituents.

The ANOVA utilized in this study allowed me to account somewhat for the effect of genotype on the insect community and plant measures separate from elevation. This approach allowed me to draw conclusions about the relative importance of genotype and environment, where variables affected by genotype only at certain elevations would support a genotype by environment interaction, where environment regulates the expression of certain traits. Abundance/g biomass and richness/g biomass were affected by genotype within certain elevations. Interestingly, abundance/g biomass varied by genotype in the lowest and highest elevations only. These elevations represent the two “extremes” in my experiment, and it may be that plants from these sites are under greater selective pressure to adapt their environments, or are expressing the highest amount of phenotypic plasticity. Plants from lower elevations are generally exposed to intense pressure from the herbivore community (Hodkinson 2005; Garibaldi et al. 2011), while plants from higher elevations must cope with suboptimal growing conditions, such as a shorter growing season and decreased temperatures (Olsson and Ågren 2002). These spatially determined selective pressures may stimulate adaptation among genotypes, resulting in certain individuals in a plant population becoming more or less appealing to the insect community.

Aphids/g biomass varied among genotypes within elevations 585 m, 885 m, and 1126 m, but not among plants from the lowest elevation (Fig. 15, Table 6). Because genotype may be particularly important to host-plant selection by this specialist insect, it is not surprising

that individuals in a plant population may adapt to cope with varying selective pressures from the aphid population within and between sites. This again supports the role of environment in regulating the phenotypic expression of traits important to the insect community. It would be interesting to compare aphid population abundances at each of these sites, particularly focusing on differences between the lowest elevation, in which there was no genotype effect, and the remaining three elevations. This may help determine if local pressure from the aphid population drives genetic differentiation between host-plants.

The third question of my study addressed how genetic or environmental variation in leaf chemistry influenced associated arthropod communities. I observed differences in insect and plant parameters due to elevation and genotype, and the relationships between them provide insight into potential mechanisms for my observations. Results from regression analyses show a significant positive relationship between abundance/g biomass, richness/g biomass, and foliar water content by genotype. This suggests that foliar water content is important to insect community structure and that insects may seek out host-plants based on genetic differences in this characteristic.

For defensive chemicals positive, yet relatively weak, relationships between abundance/g biomass and foliar concentrations of limonene, caryophyllene, and  $\gamma$ -elemene suggest that variation among some allelochemicals due to genotype may be a mechanism influencing host-plant colonization. Additionally, there was a positive relationship between insect community richness and foliar limonene concentrations at the level of genotype. In combination with a weak positive relationship between aphids/g biomass and  $\beta$ -pinene, these results were somewhat unexpected, as one would predict plants with higher concentrations of defensive compounds to support a less abundant and diverse insect community. Because

these variables were only weakly related, it is difficult to draw strong conclusions about the role terpenes play in structuring the insect community across genotypes. Nonetheless, my data do provide impetus for a deeper investigation into the role of terpenoids in a genetically diverse plant species on the associated insect community.

There was a strong negative relationship between aphid abundance and foliar water content by elevation, where plants from the lowest elevations had the lowest water content but highest aphid abundance. This result suggests that factors other than foliar water content may be important to host-plant selection by *U. nigrotuberculatum* because one would expect aphid abundance to be positively related to foliar water content. The strong positive relationship between aphid abundance and foliar CN, and the negative relationship between aphid abundance and foliar N by elevation were unexpected because higher quality plants are predicted to have higher insect abundances. Because vascular and leaf tissue constituents can vary, determining plant quality based on leaf measures might not reflect plant quality as a whole. Because aphids are phloem feeders, foliar water or CN concentrations may not be the best parameters to use when examining a population that utilizes vascular, rather than foliar tissue (Johnson 2008). It has been shown that high aphid densities on a single plant can create a nutrient sink, where nutrients are diverted from leaves to the phloem (Denno and Kaplan 2006). The relationship between richness/g biomass and foliar CN by elevation was more intuitive, where higher quality plants supported the highest number of morphospecies.

## Conclusion

In conclusion, I found evidence for both host-plant genetic and environmental variation in *S. altissima* populations to play a role in structuring both the associated insect community and influencing colonization by a dominant herbivore. Genotype was more

important than native environment in host-plant choice by the specialist, *U. nigrotuberculatum*. This result suggests that the magnitude of a genotype effect depends on community composition, where communities dominated by specialist insects are more affected by host-plant genotype than by environment. This was further supported by the finding that among the insect community only richness was significantly affected by genotype. The finding that plant measures important to insects also varied by genotype further implicates a genetic component to insect community structure. The finding that host-plant native environment affected community level but not aphid abundance, additionally supports the idea that specialist insects may be more sensitive to small-scale genetic differences in host plants, while the rest of the community may respond to large-scale differences stimulated by environmental variation in abiotic conditions.

The observed genotype effect within elevations suggests that a genotype by environment interaction may affect insect community structure. Plants are faced with unfamiliar abiotic conditions when grown outside of their native environment, and genetic variation between individuals may result in differential performance in a new environment. This variable plant performance under different environmental conditions may then be reflected in the associated community where certain genotypes become more or less appealing to insects. My results warrant the need for future studies to test for a true genotype by environment interaction by replicating host-plant genotype at multiple sites.

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## **BIOGRAPHICAL SKETCH**

Megan Ann Avakian was born on February 14, 1985 in Anderson, South Carolina. Megan attended Appalachian State University and in 2008 received a Bachelor of Science degree in Biology with a concentration in Ecology and Environmental Biology. Megan entered the Biology graduate program at Appalachian State University in August 2009 and received her Master of Science degree in August 2012. Megan began working at an environmental consulting firm in May 2012.