Competitive and allelopathic effects of garlic mustard (*Alliaria petiolata*) on American ginseng (*Panax quinquefolius*)

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Abstract The purpose of this study was to test competitive and allelopathic effects of invasive garlic mustard on American ginseng (*Panax quinquefolius*) seedlings under natural conditions. For comparative purposes, we also examined the potential effects of the native striped violet (*Viola striata*). In order to partition effects of resource competition and chemical suppression via allelopathy, field soils were amended with activated carbon or left unamended. Activated carbon positively affected ginseng growth as well as biomass of competitors. Ginseng mortality tended to increase with garlic mustard presence, though activated carbon alleviated this response. Garlic mustard had no significant effect on ginseng seedling growth, while striped violet suppressed shoot length in the absence of activated carbon. Our results showed a surprising effect of activated carbon on plant growth, a potential allelopathic effect of the native striped violet and suggest that newly invaded ginseng populations with low densities of garlic mustard may be able to withstand its effects. However, recruitment within invaded populations may decline.

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Introduction

Invasive plant species alter natural ecosystems (Ehrenfeld and Scott 2001; Vitousek et al. 1987) and reduce native plant recruitment and diversity through competition (McCarthy 1997; Thomson 2005; Stinson et al. 2007). Some invasive plants are allelopathic, releasing chemical compounds into the environment that suppress performance of neighboring plants (Rice 1984). Plants that have co-evolved with an allelopathic species may be less susceptible to allelopathic compounds while newly exposed species (such as those in an invaded range) may exhibit less resistance (Callaway and Aschehoug 2000; Callaway et al. 2008). The noteworthy success of some of the most aggressive invasive plants is attributed to these secondary compounds (Bais et al. 2003; Callaway and Ridenour 2004; Prati and Bossdorf 2004; Stinson et al. 2006; Gomez-Aparicio and Canham 2008; Callaway et al. 2008).

Many previous experiments investigating allelopathy have consisted of bioassays using aqueous plant extracts in petri dishes (Roberts and Anderson 2001; Butcko and Jensen 2002) or sterilized media. While

results from bioassays indicate potential interference, their relevance to natural environments is uncertain, especially in the absence of soil (Inderjit and Weston 2000). In field soil environments, allelochemical effects may be either greater or less than observed in vitro. For example, allelochemicals in soils can inhibit fungal mutualists (Roberts and Anderson 2001; Stinson et al. 2006; Callaway et al. 2008) which would amplify negative effects. On the contrary, allelochemicals may be degraded by bacteria much more rapidly in soils, reducing their bioavailability and negating their potential effects (Schmidt 1988). Alteration of soils by allelopathy (Heisey 1990; Heisey 1996; Prati and Bossdorf 2004; Roberts and Anderson 2001; Stinson et al. 2006) may change nutrient dynamics and ultimately ecosystem properties (Wardle et al. 1998), effects which are excluded from traditional bioassays. The differences between the effects of allelochemicals in vitro versus soils illustrates the need for more field studies of allelochemical effects (Nilsson 1994; Nilsson et al. 2000; Rich 2004; Gomez-Aparicio and Canham 2008; Cipollini et al. 2008).

One way to test for allelopathy in the field is to amend soils with activated carbon (Nilsson and Zackrisson 1992; Nilsson 1994; Rich 2004). Activated carbon has an affinity for large organic compounds (Cheremisinoff and Morresi 1978; Inderjit and Callaway 2003). This factor has led to the use of activated carbon by several studies to remove or immobilize allelochemicals in the soil; however, activated carbon may also cause some nutrient interference (Lau et al. 2008). Several studies have used activated carbon to quantify allelopathic effects (Nilsson 1994; Ridenour and Callaway 2001; Wardle et al. 1998; Callaway and Ascheoug 2000; Prati and Bossdorf 2004; Cipollini et al. 2008). Activated carbon also has been suggested for use as a restoration tool after invasion (Cipollini 2002; Kulmatiski and Beard 2005; Cipollini et al. 2008).

Garlic mustard (*Alliaria petiolata*, (Bieb.) Cavara & Grande) is a Eurasian biennial that has spread rapidly in forests in southern Canada and 36 states in the U.S. (Cavers et al. 1979; Nuzzo 2000; Welk et al. 2002; USDA 2008). This species forms dense, monotypic stands within forest edges, floodplains, undisturbed understories, and even old-growth forests (Nuzzo 1993; Nuzzo 1999; Stinson et al. 2006; Weber and Gibson 2007), reducing growth,

abundance, and diversity of native species (McCarthy 1997; Stinson et al. 2006; Stinson et al. 2007).

Evidence for the role of allelopathic chemicals in garlic mustard's competitive ability is contradictory or inconclusive. One early study found that extracts from garlic mustard did not affect seeds or seedlings of radish, winter rye, hairy vetch, and lettuce (McCarthy and Hanson 1998). However, several phytotoxic chemicals such as glycosides, phenolic acids, cyanide, and glucosinolates were isolated from leaf, stem, and root extracts of garlic mustard (Vaughn and Berhow 1999; Cipollini 2002; Cipollini et al. 2005; Cipollini and Gruner 2007). A short-term competition study by Meekins and McCarthy (1999) showed that garlic mustard reduced growth of Chestnut oak (*Quercus prinus*) seedlings, yet was outcompeted by boxelder (*Acer negundo*) and spotted jewelweed (*Impatiens capensis*). Removal of garlic mustard increased native diversity in invaded areas (McCarthy 1997; Stinson et al. 2007), suggesting its ability to suppress native recruitment. Growth suppression may be due to indirect competition, direct competition (allelopathy), or both. Prati and Bossdorf (2004) found that garlic mustard-contaminated substrate had a greater allelopathic influence on a North American species than on a European congener, supporting the "novel weapons" hypothesis (Callaway and Aschehoug 2000). Garlic mustard was later shown to reduce growth of mycorrhizal tree seedlings such as red maple (*Acer rubrum*) and white ash (*Fraxinus americana*) (Stinson et al. 2006), as well as suppress North American arbuscular mycorrhizal fungi (AMF) more than European AMF (Callaway et al. 2008) and ectomycorrhizal fungi (Wolfe et al. 2008). Suppression of fungal mutualists by garlic mustard may have community level impacts in eastern deciduous forests.

American ginseng (*Panax quinquefolius* L.) is a culturally significant and economically important native medicinal plant found throughout the eastern deciduous forest (Bailey 1999; Robbins 2000; McGraw et al. 2003). Similar to many other understory species, American ginseng forms vesicular arbuscular mycorrhizal associations (VAM) (Seo and Anderson 1990). Concern over the sustainability of harvesting American ginseng spurred its listing under Appendix II of CITES (Convention on International Trade in Endangered Species of Wild Fauna and Flora). Poor harvest practices (Van der Voort and

McGraw 2006), deer browse (McGraw and Furedi 2005), inbreeding, and reduced genetic diversity within small populations (Cruse-Sanders and Hamrick 2004, Mooney and McGraw 2007) are all factors that may cause decline of ginseng populations. Recently, high levels of invasive plant species, including garlic mustard, have been documented within natural ginseng populations (Wixted and McGraw 2009). Garlic mustard was found in 5 out of 30 natural populations, with 7.1% of the 4,291 censused ginseng plants having garlic mustard within a 2-m proximity (Wixted and McGraw 2009). Therefore, the objectives of this research were to examine the following questions: (1) Is there evidence for a competitive and/or allelopathic effect of an invasive non-native competitor (garlic mustard) on ginseng seedling survival and growth? (2) Is there evidence for a competitive and/or allelopathic effect of a native competitor (striped violet) on ginseng seedling survival and growth? (3) Is there a differential allelopathic effect of a native and invasive competitor on ginseng seedling survival and growth? Our experiment was designed to answer these questions in a natural field setting to observe how ginseng seedlings, the most vulnerable stage of ginseng growth, could be affected by one generation of garlic mustard (i.e., over 2 years).

Methods

Site characteristics

The study site was located on a northeast-facing slope at ca. 580-m elevation in the western Allegheny plateau region of Monongalia County, West Virginia. Precise coordinates are withheld for conservation reasons. The soils were appropriate for ginseng since a natural population was found there in a 60–80-year-old mixed mesophytic, second growth forest dominated by tulip poplar (*Liriodendron tulipifera*) and black cherry (*Prunus serotina*) with spicebush (*Lindera benzoin*) in the understory. The main soil type in the study area was Dekalb stony loam characterized as moderately deep and excessively drained sandstone soils (Soil Survey Staff 2008). The site lacked garlic mustard invasion, although invasions were known from sites nearby (<2 km away).

Study species

Garlic mustard is a biennial that has seeds which germinate in early spring, and seedlings remain in an evergreen basal rosette during the first year of growth (Cavers et al. 1979). During the second year of growth, rosettes rapidly bolt, growing ca. 2 cm/day between mid-April and mid-May (Anderson et al. 1996). The flowers begin anthesis in early spring (Cavers et al. 1979). From July through October, seeds are passively dispersed and plants senesce. Garlic mustard is self-pollinating, exhibits high rates of seed production (Cavers et al. 1979; Anderson et al. 1996), and can thrive in a variety of conditions, all traits contributing to its success.

Striped violet (*Viola striata*, Aiton) was chosen as a potential native competitor for ginseng due to the similarity of its leaf shape and size to that of garlic mustard rosettes as well as its abundance within natural ginseng populations (Wixted, pers obs. 2006). Striped violet is a perennial understory species found throughout mesic, deciduous forests within the eastern United States (USDA 2008; Strausbaugh and Core 1978). It produces chasmogamous and cleistogamous flowers from April to June, and seeds germinate the following spring (Strausbaugh and Core 1978). This species was identified using Strausbaugh and Core (1978).

American ginseng is a long-lived perennial herb with a thick taproot attached to a rhizome (Charron and Gagnon 1991). Ginseng seeds germinate following a 19 month (or more) dormancy period and begin growth during late April to early May (Hackney and McGraw 2001). The one-leaf seedling phase lasts ca. 1–7 years (McGraw, unpublished data). After sufficient root growth, a second leaf will appear to produce a juvenile plant, which may or may not produce flowers (Lewis and Zenger 1982; Charron and Gagnon 1991). In General, ginseng plants produce a third or fourth leaf within 5–10 year or more; plants can be highly variable in maturation rate (Anderson et al. 1993). Most adult ginseng plants flower, but fruit and seed production are sporadic, increasing as plant size increases (Shahi 2007).

Experimental design

In late April 2006, 2 weeks prior to transplanting, stratified, wild-simulated New York ginseng seeds

were germinated in a greenhouse in native soil from the study area. Garlic mustard and striped violet seedlings were collected from a local site in Morgantown, WV and acclimated to the greenhouse.

A 50 m × 50 m plot was set up in the study area and competition "arenas" (10-cm depth × 15 cm diameter plastic pipe) were buried throughout the original grid at random locations, avoiding only dense fern patches and excessively rocky microsites. Arenas were used to limit root competition from species other than the treatment plant as well as to standardize the amount of soil used for the treatments. Six treatments were set up in a 3 × 2 factorial design with two factors (activated carbon and competitor) using 50 replicates of each: (1) ginseng alone, (2) ginseng with striped violet, or (3) ginseng with garlic mustard, planted in either (a) mixed native soil or (b) mixed native soil amended with GRO–SAFE activated carbon (NORIT Americas Inc.). Treatments with activated carbon (AC) were used to remove potential allelopathic influences thereby partitioning these effects from those of resource competition. Activated carbon was added in a ratio of 11.3 g/kg of dried soil, a method and amount similar to a field study in eastern deciduous forests adopted by Rich (2004). Rich's (2004) study examined a dose–response for three herbaceous perennial species to determine the amount of AC to add to field soils. The AC concentration, furthermore, was similar to amounts used in studies by Callaway and Aschehoug (2000) and Prati and Bossdorf (2004). Since addition of AC requires substantial disturbance to mix and sieve soil, unamended, native soil was also mixed and sieved. An earlier study by Prati and Bossdorf (2004) found that AC addition did not affect garlic mustard growth or reproduction.

Ginseng seedlings were placed in randomly assigned treatments at the beginning of May 2006, and each arena was given a unique ID. Seedlings that senesced within the first week were replaced because we assumed they died from transplanting stress. Every 2 weeks throughout the growing season, plants other than the transplants were removed. Deer and other large herbivores were excluded from the experiment with wire mesh cages to ensure adequate sample size, and leaf litter from the site was added to the arenas to prevent unnatural soil drying. In order to prevent garlic mustard invasion at the study site, siliques were removed prior to dispersal in late June 2007 and dried to allow calculation of total competitor biomass.

Garlic mustard plants were harvested at the start of senescence in mid-July 2007 to determine total biomass. At the conclusion of the experiment in late August 2007, striped violet was harvested with ginseng seedlings prior to violet senescence. Total biomass of competitors was examined to determine whether AC would alter the size of competitors, thus interacting in an unanticipated manner with the dependent variable—presence of a competitor.

Dependent variables

In June 2006 and 2007, after full leaf expansion, leaf area was determined by acquiring digital images of individual ginseng leaves silhouetted against a solid white background with a ruler in the field of view. Shadows were removed in Adobe Photoshop v.7.0 as needed. Leaf area was then quantified using NIH ImageJ v.1.37 (Rasband 2005). Relative growth rate on a leaf area basis (RGR_{LA}) for ginseng was then calculated using Eq. 1 (McGraw and Garbutt 1990).

$$RGR_{LA} = \ln(\text{leaf area } 2007) - \ln(\text{leaf area } 2006)$$
(1)

In addition, the number of leaves on each ginseng seedling was also counted in 2007 (the number was always 1 at the end of 2006).

Survival of ginseng seedlings and competitors was recorded at the end of August in 2006 and 2007. At the conclusion of the second growing season, in August 2007, all remaining ginseng and striped violet plants were harvested. If ginseng shoot tissue was not present, then the arena was searched for a viable root with the apical bud still present. After harvest, root tissue of all plants was thoroughly rinsed to remove soil. Ginseng seedlings were separated into root and shoot portions. The length of each portion was then measured before drying at 65°C for 48 h. The root to shoot ratio was also calculated to test for biomass allocation differences among treatments. Any ginseng seedlings which lacked shoot tissue at the conclusion of the experiment were excluded from root length, root mass, root:shoot ratio, and total biomass analyses.

Data analysis

The experimental design permitted several analyses. In the first two analyses, the data were analyzed as a

3×2 factorial experiment, with competition treatment (no competitor, violet, or garlic mustard) and AC treatment (−AC, +AC) as main effects. This analysis was used to determine whether there was a competitive (competition treatment main effect) or differential allelopathic effect (competitor × AC interaction). Log-likelihood tests were used to analyze seedling survival and leaf size fate (one vs. two leaves in 2007) with the same effects in the model. When the competition treatment was statistically significant for the log-likelihood tests, we additionally performed analyses of the 2×2 analyses as appropriate to distinguish the effect of each competitor (vs. no competitor) and comparing the effect of violet vs. garlic mustard (Fig. 1). Ginseng size and growth variables were analyzed using a two-way ANOVA.
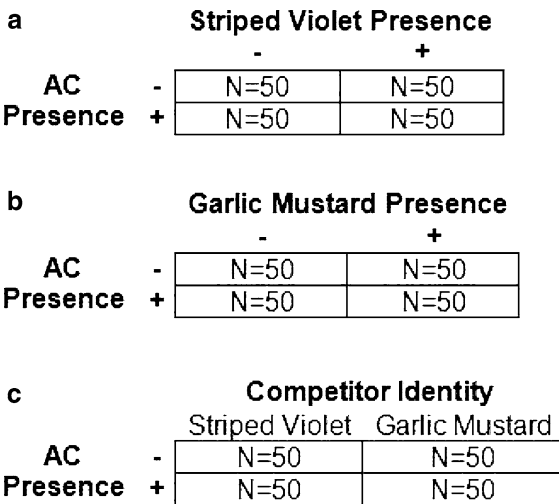
The response of competitor biomass was also analyzed using a two-way ANOVA with competitor identity and AC as the main effects. Finally, an ANCOVA was used for the competition treatments to determine whether the effect of competitor size (continuous) on ginseng biomass varied depending on AC treatment (nominal). Normality of the residuals was tested using a Shapiro-Wilk goodness of fit test. Homogeneity of variances was assessed using Bartlett's test. Data were log transformed if normality

and/or homogeneity of variance assumptions were not met. A Tukey-Kramer HSD post hoc test was used after ANOVAs to compare means among levels for significant model effects. All the data were analyzed using SAS JMP v. 6.0 (SAS Institute 2005).

Results

In the first growing season (2006), ginseng mortality was not significantly affected by the presence of AC or the addition of a competitor. However, by the end of the second season (2007), ginseng seedling mortality was higher in competition treatments with or without AC in the soil (3×2 analysis; competitor main effect; L-R $\chi^2 = 27.5268$, $P < 0.0001$). On examining the effect of one competitor species at a time (2×2 analysis), it is clear that in 2007, ginseng seedling mortality was higher in striped violet treatments with or without AC in the soil (competitor main effect; L-R $\chi^2 = 23.17$, $P < 0.0001$; Fig. 2a). On analyzing the effects of the invasive garlic mustard (Fig. 1b), we found that in 2007, seedling mortality was 2.5 times higher in garlic mustard treatments with or without AC in the soil (competitor main effect; $F = 23.13$, $P < 0.0001$), an effect comparable to that seen with native violet. In 2007, there was also a trend suggesting a greater effect of garlic mustard on ginseng mortality in the absence of AC (competitor × AC interaction; L-R $\chi^2 = 2.91$, $P = 0.0859$; Fig. 2b). When analyzing only the competition treatments (Fig. 1c), ginseng mortality was not significantly different when in competition with garlic mustard versus violet, nor did AC affect mortality ($P > 0.05$).

The frequency of ginseng seedlings progressing from the one leaf to the two leaf stage in 2007 tended to increase in AC treatments regardless of competitor presence or identity (AC main effect; L-R $\chi^2 = 3.17$, $P = 0.0749$). In terms of growth variables, ginseng appeared insensitive to the presence of a competitor, regardless of competitor identity (Table 1). AC positively affected ginseng root length, shoot mass, root mass, total biomass, and the relative growth rate of leaf area for ginseng seedlings, whether the native or invasive competitor was present or not (AC main effect; Tables 1, 2). The root:shoot ratio was also not significantly affected by AC or competitor treatment (Table 1). In the absence of AC, striped violet suppressed ginseng shoot length, but this effect was

a Striped Violet Presence

AC Presence		−	+
	−	N=50	N=50
	+	N=50	N=50

b Garlic Mustard Presence

AC Presence		−	+
	−	N=50	N=50
	+	N=50	N=50

c Competitor Identity

AC Presence		Striped Violet	Garlic Mustard
	−	N=50	N=50
	+	N=50	N=50

Fig. 1 Three two-way factorial permutations of the six treatments examining **a** differential effects of violet presence with and without activated carbon (AC), **b** differential effects of garlic mustard presence with and without AC, and **c** differential effects of the native striped violet and the invasive garlic mustard as a function of AC treatment
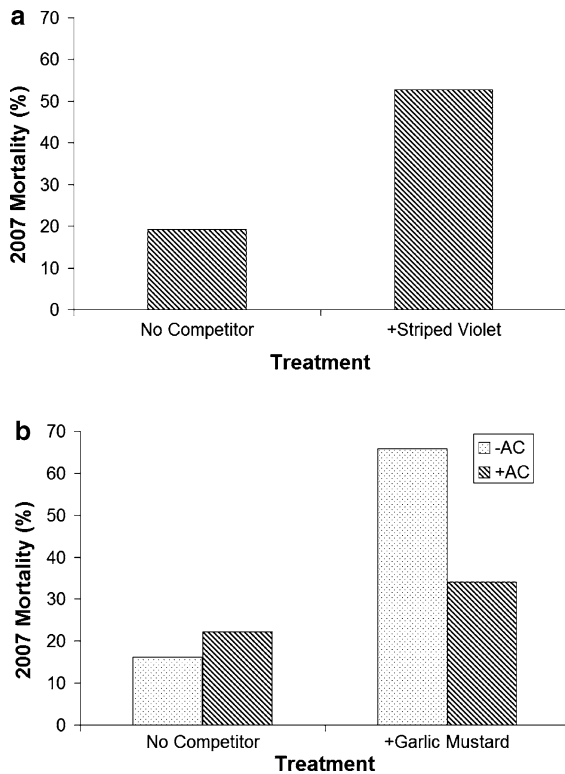
Fig. 2 **a** Effect of striped violet presence on ginseng seedling mortality in 2007 and **b** differential effect of garlic mustard on ginseng seedling mortality (2007) depending on AC treatment after two growing seasons

not observed in the presence of AC (competitor × AC interaction; $F = 8.70$, $P = 0.0100$; Fig. 3).

The biomass of garlic mustard was significantly higher than that of striped violet, regardless of AC presence or absence (Species effect; $F = 29.93$, $P = 0.0001$; Fig. 4a). There was also a trend suggesting AC treatment stimulated competitor biomass (AC main effect; $F = 2.80$, $P = 0.0985$; Fig. 4b).

When examining the competition treatments, ginseng biomass was found to increase as competitor biomass increased ($F = 8.7142$, $P = 0.0045$), yet

this effect did not depend on carbon treatment or competitor identity ($P > 0.05$).

Discussion

In the presence of a competitor, ginseng seedling mortality increased, and this effect was comparable in magnitude for the native and invasive competitor. However, within garlic mustard treatments, there was a tendency for this mortality effect of garlic mustard to be reduced in the presence of activated carbon. Ginseng survival was 21% higher in garlic mustard treatments that contained AC in the soils than in soil without AC. While reduced resource availability likely increased ginseng seedling mortality within competition treatments, the differential response of ginseng to carbon treatment in the presence of garlic mustard is consistent with an allelopathic effect as well. Given that ginseng has low seed production, an increase in mortality could have significant consequences for population growth. Moreover, given the relatively low level of exposure to competition ($n = 1$ competitor within a 15 cm diameter arena), even higher levels of competition might be expected to enhance this mortality effect.

A puzzling result was the positive correlation between competitor biomass and ginseng biomass in surviving seedlings. This result seems likely to be due to the combination of a relatively low level of competition created by our treatments and microsite variation in conditions for plant growth. In our experiment, competition arenas in "good" microsites supported greater growth of both competitor and ginseng, while arenas in poor microsites affected both negatively. Morris (1999) found a high degree of variation in fungal and bacterial biomass in soils at scales as small as 1–10 cm. In addition, close proximity to tree species such as sugar maple (*Acer saccharum*) may affect nitrogen mineralization rates

Table 1 ANOVA summary table with P values

Effect	Shoot length	Root length	Shoot mass	Root mass	Root: shoot ratio	Total biomass	RGR_{LA}
Competitor ID	0.6113	0.6804	0.2815	0.3935	0.3198	0.3728	0.8105
AC (±)	**0.0243**	**0.0065**	**0.0022**	**0.0124**	0.1998	**0.0049**	**0.0026**
Competitor ID × AC	**0.0100**	0.7029	0.1408	0.2811	0.5565	0.2053	0.2801

Significant values ($P < 0.05$) are bold faced. All dependent variables except the relative growth rate of leaf area were log transformed

Table 2 Summary table of backtransformed least-squared means and 95% confidence intervals for ginseng dependent variables with a significant AC effect
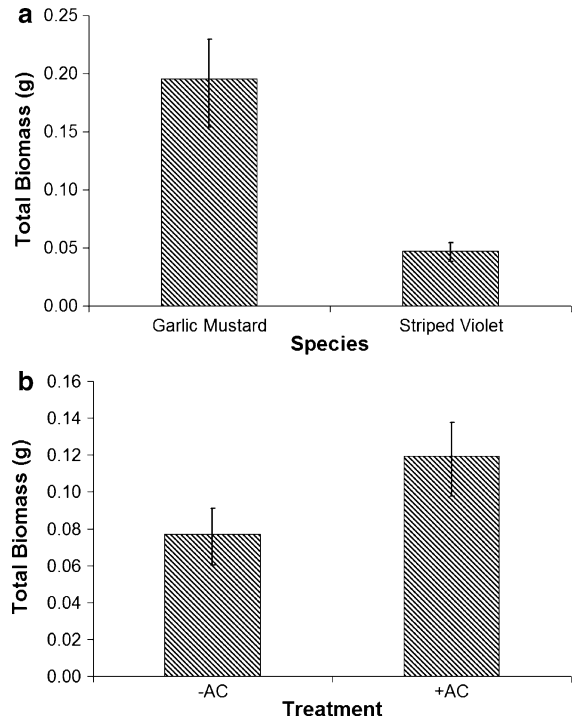
Dependent variable	−AC	+AC	Change (%)
Root length (cm)	9.3311 (8.4572, 10.2956)	11.1911 (10.2654, 12.2004)	20
Shoot mass (g)	0.0532 (0.0457, 0.06195)	0.0731 (0.0639, 0.0836)	37
Root mass (g)	0.1013 (0.0861, 0.1191)	0.1332 (0.1155, 0.1537)	32
Total biomass (g)	0.1559 (0.1338, 0.1816)	0.2087 (0.1824, 0.2087)	34
RGR$_{LA}$	0.4891 (0.3270, 0.6513)	0.8308 (0.6786, 0.9830)	70
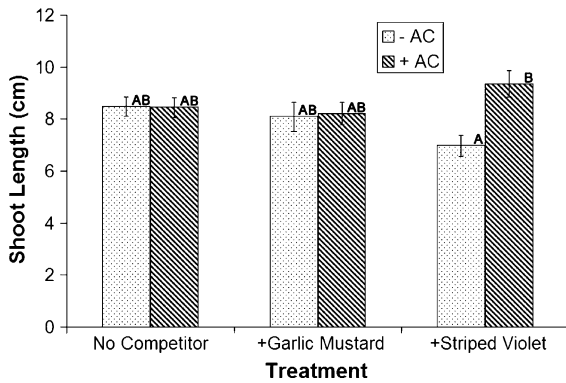
Fig. 3 Differential effect of competitor species on mean shoot length (cm) in ginseng seedlings after two growing seasons depending on AC treatment; means with the same letter are not significantly different using the Tukey-Kramer HSD a posteriori test

Fig. 4 a Competitor biomass after two growing seasons and **b** effect of AC treatment on biomass of the competitor after two growing seasons

in soils (Boerner and Koslowsky 1989). Decomposition of leaf litter (McClaugherty et al. 1985) and distribution of parent material (Vankat et al. 1977) may cause fine-scale patterning of soil nutrients. One or more of these factors may have led to the positive biomass correlation between competitors and ginseng in our experiment.

The effect of striped violet on ginseng shoot length depended on AC treatment with ginseng shoot length significantly increasing in striped violet treatments with AC. This differential response was a surprising result which may be explained by two possible causes: allelopathy or resource competition. Until the recent studies by Lau et al. (2008), Cipollini et al. (2008), and our results showing positive effects of activated carbon, we would have concluded that the native striped violet was allelopathic despite its tendency to co-occur with American ginseng. However, AC may have altered the degree of competition for nutrients. Competition with neighbors has been well documented to affect survival and growth

(Aarssen and Epp 1990), and plants with similar niches may be stronger competitors (Goldberg and Werner 1983). Striped violet and ginseng may have been strong competitors which may be the reason why stems of ginseng seedlings were longer, even as the competitor biomass increased in carbon treatments. This effect may also explain why ginseng was more negatively affected by striped violet presence than the presence of the larger garlic mustard. Furthermore, if AC altered limiting soil nutrients, then the competitive effect of violet through resource depletion may have been reduced as well. Future

research should further examine the potential for striped violet allelopathy.

The lack of allelopathic effects of garlic mustard on ginseng seedling growth may be due to a variety of factors. Our study simulated a newly invaded area with a low density of garlic mustard. However, earlier studies have found that garlic mustard grows better in previously invaded sites (Klironomos 2002). In addition, increased N and P availability, soil pH, and base cation availability were associated with field populations of garlic mustard (Rodgers et al. 2008). These nutrient effects could be cumulative over time, creating a beneficial feedback for garlic mustard but not for native species. In addition, Vaughn and Berhow (1999) found that glucosinolate concentrations in garlic mustard were the highest in the fall, potentially indicating that decomposition of garlic mustard would produce the most inhibitory effect. A recent study by Barto and Cipollini (2009) also suggested that allelopathic effects of garlic mustard may be due to degradates of glucosinolates and flavonoid glycosides or an unknown effect. In our study, we removed garlic mustard before it had a chance to decompose in order to measure its biomass. Furthermore, garlic mustard seeds have been found to contain high levels of sinigrin, a phytotoxic glycoside (Larsen et al. 1983). It is possible that the combined effects of compounds leaching from seeds and adult decomposition alter soils over time to benefit further invasion and prevent native growth. In addition, while garlic mustard has been found to allelopathically reduce growth in species such as *Geum laciniatum* (Prati and Bossdorf 2004), such effects may be species specific (Meekins and McCarthy 1999).

The lack of competitive effects of garlic mustard on ginseng seedling growth is also interesting. Overall, garlic mustard was a large competitor that many times grew taller than and shaded ginseng seedlings. Meekins and McCarthy (1999) found species' specific effects of garlic mustard rosettes on native plants such as chestnut oak (*Quercus prinus*), box elder (*Acer negundo*), and spotted jewelweed (*Impatiens capensis*). In that study, garlic mustard was outcompeted by the latter two species; however, garlic mustard was able to reduce the growth of chestnut oak (Meekins and McCarthy 1999). The researchers attributed chestnut oak's reduced biomass in the presence of garlic mustard to light competition. However, ginseng is a shade-obligate species and growth was not significantly affected by resource competition from garlic mustard in our study.

The presence of AC in the soil-affected several ginseng growth variables, regardless of competitor presence or identity. The recent study by Lau et al. (2008) found that addition of AC in unfertilized soils significantly increased pH, phosphorus, potassium, and iron concentrations in soil leachates, whereas calcium and magnesium decreased. While the study by Lau et al. (2008) was conducted over a single week, it showed immediate effects of AC that could persist over a longer time frame. Lau et al. (2008) also conducted three growth experiments in which they added AC to potting media and found that in most herbaceous species, AC presence increased biomass. The increase in biomass was attributed to changes in nutrient availability. In addition, Cipollini et al. (2008) found that in the presence of invasive plants such as garlic mustard and Amur honeysuckle (*Lonicera mackii*), AC addition aided the native spotted jewelweed (*Impatiens capensis*) by either increasing native survival, fruit number and/or plant height. However, in the absence of aboveground biomass of the invasive species, AC had a negative effect which may have been due to AC being adsorbed to inorganic nutrients (Cipollini et al. 2008). A similar phenomenon may have been responsible for increased ginseng growth in AC treatments in our study.

In addition to AC's effects on soil nutrients, two other factors may explain our results. First, inhibitory organic compounds already found in the soil at the study area and our treatment design may have resulted in enhanced growth as AC removed the compounds from the rhizosphere. Interrupted fern (*Osmunda claytoniana*) existed in parts of the study area, and caution was taken to avoid planting treatments directly in fern patches since previous studies have demonstrated allelopathy in ferns (Stewart 2005; Hanson and Dixon 1986). However, by the end of the second year, some ferns had encroached near treatments. Second, our treatments also required mixing of the soils as well as removal of other herbaceous species which created more control over our experimental design but deviated from natural conditions. For example, Booth et al. (2006) found that mixed soil cores had higher mineralization and NH_4^+ assimilation, while NO_3^- consumption declined.

Overall, our results do show a surprising effect of AC on both ginseng and biomass of the competitor species. These results reaffirm the need for a better understanding of how AC alters soil nutrients, especially over time. Our results from this field study parallel the short-term greenhouse experiments conducted by Lau et al. (2008) and fieldwork by Cipollini et al. (2008) which suggest that AC treatments may confound test species' growth and bias allelopathy assumptions. Ginseng mortality increased with the addition of a competitor, and trends in the data suggest that ginseng survival in the presence of garlic mustard depended on carbon treatment, suggestive of allelopathy. However, ginseng seedlings which survived tended to grow larger with the addition of a competitor, regardless of competitor identity. The data also suggest that striped violet allelopathically decreased ginseng shoot length. The lack of evidence for strong competitive effects of either species on ginseng growth may indicate ginseng seedling tolerance of low levels of competition; however, reduced recruitment of ginseng seedlings via increased mortality may be sufficient to significantly lower population growth rates in invaded populations.

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