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ECOSYSTEMS

Hemlock Declines Rapidly with Hemlock Woolly Adelgid Infestation: Impacts on the Carbon Cycle of Southern Appalachian Forests

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ABSTRACT

The recent infestation of southern Appalachian eastern hemlock stands by hemlock woolly adelgid (HWA) is expected to have dramatic and lasting effects on forest structure and function. We studied the short-term changes to the carbon cycle in a mixed stand of hemlock and hardwoods, where hemlock was declining due to either girdling or HWA infestation. We expected that hemlock would decline more rapidly from girdling than from HWA infestation. Unexpectedly, in response to both girdling and HWA infestation, hemlock basal area increment (BAI) reduced substantially compared to reference hardwoods in 3 years. This decline was concurrent with moderate increases in the BAI of co-occurring hardwoods. Although the girdling treatment resulted in an initial pulse of hemlock needle inputs, cumulative litter inputs and O horizon mass did not differ between treatments over the study period. Following girdling and HWA infestation, very fine root biomass declined by 20–40% in 2 years, which suggests hemlock root mortality in the girdling treatment, and a reduction in hemlock root production in the HWA treatment. Soil CO_2 efflux (E_{soil}) declined by approximately 20% in 1 year after both girdling and HWA infestation, even after accounting for the intra-annual variability of soil temperature and moisture. The reduction in E_{soil} and the concurrent declines in BAI and standing very fine root biomass suggest rapid declines in hemlock productivity from HWA infestation. The accelerated inputs of detritus resulting from hemlock mortality are likely to influence carbon and nutrient fluxes, and dictate future patterns of species regeneration in these forest ecosystems.

Key words: carbon cycling; eastern hemlock; hemlock woolly adelgid; litter fall; O horizon; root biomass; soil respiration; southern Appalachians.

INTRODUCTION

Large-scale outbreaks of exotic pests and pathogens substantially alter forest structure, composition,

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AEN performed research and analyzed data; NW performed research, analyzed data, and wrote the article; CRF contributed new methods, analyzed data, and wrote the article; RLH designed the study; JMV conceived of and designed the study; and BDK performed research.

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In the last century, eastern North American forests have experienced the widespread decline and mortality of dominant tree species, such as American chestnut (*Castanea dentata* (Marsh.) Borkh) and American elm (*Ulmus americana* L.), due to exotic pathogens (Liebhold and others 1995; Orwig and Foster 1998; Ellison and others 2005). Currently, hemlock woolly adelgid (*Adelges tsugae* Annand), an invasive exotic insect, is responsible for the decline of eastern hemlock (*Tsuga canadensis* (L.) Carr.). Hemlock woolly adelgid (HWA) is a small aphid-like insect that feeds on stored sugars in xylem ray parenchyma at the base of needles (Young and others 1995). HWA attacks hemlock trees of all ages and sizes, and infested trees seldom recover (Orwig and Foster 1998; Kizlinski and others 2002).

The mortality of eastern hemlock is likely to influence stand and ecosystem processes of eastern forests (Ellison and others 2005; Ford and Vose 2007). Eastern hemlock is a foundation species (Ellison and others 2005) due to its slow-growing, shade-tolerant, and late-successional characteristics that help create important structural diversity at the stand and landscape levels (Orwig and Foster 1998; Tingley and others 2002). Although hemlock is individually less productive than co-occurring hardwood species (Catovsky and others 2002), stands of hemlock and hardwoods are more productive than stands of hardwoods alone (Kelty 1989; Busing and others 1993). This may be due to the evergreen nature of hemlock and canopy stratification among species in these stands. Hemlock also contributes to a significant accumulation of organic matter in comparison with other species (Campbell and Gower 2000), because its litter decomposes slowly (Elliott and others 1993).

The species expected to replace hemlock, and their projected influence on forest community composition and function, differ across the distribution of hemlock in eastern North America (Ellison and others 2005). Following the loss of hemlock in southern Appalachian forests, the change in forest composition is likely to follow one of two trajectories: replacement by *Rhododendron maximum*, an evergreen ericaceous shrub or replacement by a mixture of advance regeneration tree species. If hemlock is replaced by *R. maximum*, a species which restricts the recruitment of other, more productive species into the canopy, then stand level primary productivity may be dramatically reduced from current levels (Day and Monk 1977; Nilsen and others 2001). Replacement by *R. maximum* is also likely to enhance soil carbon due to the high tannin content of leaf and root litter, and reduce soil N availability to hardwood species (Wurzburger and Hendrick 2007). In contrast, replacement by a mix of advance regeneration and early successional tree species (for example, *Acer rubrum, Betula lenta*, and *Liriodendron tulipifera*) will promote soils with relatively high N availability (Boettcher and Kalisz 1990), that have the potential to sustain high productivity (Day and Monk 1977).

Despite widespread HWA infestation, and the importance of eastern hemlock to eastern forest structure and function, we know little about how the process of hemlock mortality affects the forest carbon cycle. In the northeast, eastern hemlock stands infested with HWA display progressive needle loss from decreased needle and bud production, and branch dieback (McClure 1991; Orwig and Foster 1998; Jenkins and others 1999; McClure and Cheah 1999; Stadler and others 2005; Cobb and others 2006; Eschtruth and others 2006). Past research has simulated HWA-induced mortality by girdling trees, resulting in a rapid termination of photosynthate supply (Yorks and others 2003). However, the available evidence suggests that HWA infestation results in a more gradual decline toward mortality than the girdling process simulates (Orwig and Foster 1998; Stadler and others 2005, 2006). The belowground responses to HWA infestation are likely to be significant, and mirror those aboveground; root biomass production accounts for a significant proportion of forest productivity (Joslin and others 2001), and root respiration can account for up to one-half of total E_{soil} (Bond-Lamberty and others 2004) in similar forest ecosystems. No studies have reported hemlock fine root production or E_{soil} patterns in infested stands, nor have they tracked the short-term progression of hemlock decline to the forest carbon cycle.

The objectives of this research were to compare the rates of hemlock decline induced by either girdling or HWA infestation, and to quantify concurrent changes in the carbon cycle in southern Appalachian hemlock stands. We conducted a girdling treatment alongside neighboring hemlock stands that, within months, became infested with HWA. We hypothesized that girdling would result in rapid and significant changes in the carbon cycle by terminating photosynthate allocation belowground (Figure 1). In contrast, we hypothesized that HWA infestation would result in a more gradual decline of hemlock due to the slower progression toward mortality and reduced productivity. More specifically, we hypothesized that: (1) aboveground indicators of decline (reduced basal area growth and enhanced leaf litter fall) would show little progression in HWA-infested stands, compared to the rapid progression in the girdled stands; (2) litter horizon (Oi) mass would reflect litter fall patterns of declining trees and be greater in girdled than in HWA stands at the end of the study period (Figure 1B, C); and (3) belowground

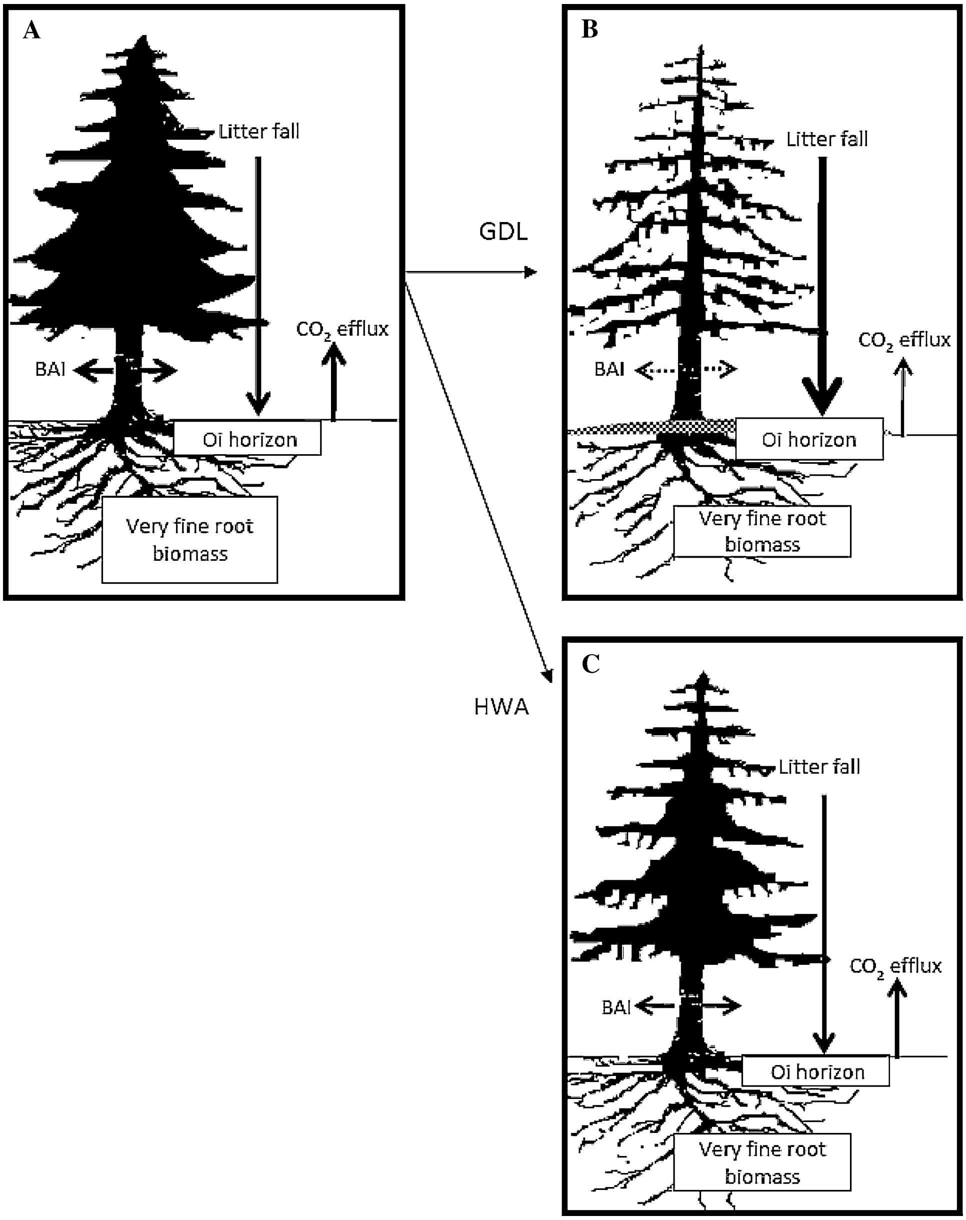

Figure 1. Hypothesized changes in the carbon cycle during the study period in girdled (GDL) plots, (**A**–**B**), and in hemlock plots infested with hemlock woolly adelgid (HWA) (**A**–**C**). BAI denotes basal area increment. Sizes of boxes and arrows among panels indicate magnitude of hypothesized changes.

indicators of decline (reduced very fine root standing biomass and altered E_{soil}) would show little progression in the HWA-infested stands compared to the rapid progression in the girdled stands.

Methods

Site Description and Experimental Design

The study site is located in the Coweeta basin in the Nantahala Mountain Range of western North Carolina, USA. Climate in the basin is classified as marine humid temperate with cool summers and mild winters (Swift and others 1988). Average annual temperature in the Coweeta basin is 13°C and average annual rainfall is 178 cm. Study plots were established in low-elevation (730–1,040 m) cove hardwood forests (Elliott and others 1999). Plots were located in the riparian zones along Shope Fork and Ball Creek in Inceptisols of the Cullasaja soil series. The dominant species were eastern hemlock, blackgum (*Nyssa sylvatica* Marsh.), white oak (*Quercus alba* L.), tulip poplar (*Liriodendron tulipifera* L.), red maple (*Acer rubrum* L.), black birch (*Betula lenta* L.), and rhododendron (*Rhododendron maximum* L.).

In our experimental treatment plots, hemlock comprised more than half of the total basal area. The experimental design had two treatments with four replicate 20 m × 20 m plots ($n = 8$). Treatments were randomly assigned to the plots. Within each plot, we stratified five subplots (0.1104 m^2) on a grid. Subplots were established to account for within-plot spatial heterogeneity for litter inputs and E_{soil} measurements (described below). All hemlock trees in the girdled plots (and within 5 m of the plot boundaries) were girdled by handsaw or chainsaw at breast height (1.37 m) in July 2004. Girdling depth was intended to sever the cambium, phloem, and sapwood. We re-girdled trees during annual surveys in 2005 and 2006 if they still appeared alive. Hereafter, we refer to plots as girdled (GDL) treatments and HWA-infested (HWA) treatments, respectively.

HWA was first noticed on a few trees along the main roads in the Coweeta basin in the fall of 2003 and in Macon county, NC as early as 2002 (USDA Forest Service 2002). An extensive survey of permanent vegetation plots in the Coweeta basin found little or no infestation in the rest of the basin in 2003, but significant infestation throughout the basin by 2005 (K. Elliott, unpublished data). We first observed HWA infestation in the HWA plots in December 2004; however, infestation levels appeared low and the crowns were full and healthy. By 2005, study plots were heavily infested with HWA and reduced crown vigor was obvious. Because our first observations of HWA infestation were in the fall of 2004 and not in the spring, infestation of the study trees was likely due to dispersal of the second generation of crawlers from nearby areas in early summer. This second generation settles on new growth, but instead of feeding on hemlock, it enters aestivation until October when they begin to feed (McClure 1989).

Basal Area Increment

In 2004, we tagged all trees with stem diameters greater than 2.54 cm at 1.37 m height. Stems with diameters larger than 15 cm were fitted with dendrometer bands (Cattelino and others 1986) at 1.37 m in 2004 and smaller diameter stems were measured annually manually with a tape. Because we installed dendrometer bands in 2004, the first year we could calculate stem growth was 2005, which was post-girdling and HWA infestation. To provide three successive years of data, we included 2007 stem measurements. We determined annual basal area increment (BAI) from the difference of successive annual growth measured at the end of each year. To isolate differences in BAI due to treatments rather than climate, we calculated a ratio of hemlock BAI to the BAI of hardwood stems in the neighboring hardwood plots. The hardwood plots ($n = 4$) had the same dimensions and hardwood composition as the experimental plots, but received no treatment, and contained less than 3% of basal area as hemlock. To assess whether co-occurring hardwoods in the treatment plots were responding to the treatments, we calculated a ratio of hardwood BAI to the BAI of hardwood stems in the hardwood plots. These ratios accounted for inter-annual variability in growth due to climate because tree diameter growth is highly reflective of climatic conditions (Fritts 1976).

Litter Fall

We collected leaf and fine branch (ca. ≤ 1 cm in diameter) litter fall monthly from five circular 0.110 m^2 area litter traps per plot (one per subplot) during April–November in 2004, 2005, and 2006. Litter was separated into hemlock needles, hemlock fine branch material, and non-hemlock components (other leaves, other fine branch material, and seeds, and so on), dried at 60°C to a constant mass, and weighed. Several months of litter samples from 2004 were lost and because our measures did not represent a complete growing season, we restricted our analysis of litter data to trends over 2005 and 2006.

O Horizon

We measured O horizon mass (g m^{-2}) three times during 2006 (March, July, and December). In each plot, we cut six O horizon samples with a knife using a 0.09 m^2 template from random locations along three transects. The determination of the O and A horizon boundary was based on a visual assessment of organic content and color differences between the organic layer and mineral soil. Roots were not removed from the samples. Samples were separated into fresh litter (Oi horizon) and fragmented litter plus humus (Oe + Oa horizons), dried at 60°C to a constant mass, and then pooled by plot for each horizon. All samples were then ground, and analyzed for percent carbon by dry combustion (Harmon and Lajtha 1999). The Oe + Oa horizon samples were ashed to correct for mineral soil in the samples.

Root Biomass

We determined total standing root biomass from soil cores collected in July 2004 and July 2006. In each plot, cylindrical soil samples 5 cm in diameter and 20 cm in depth (including the O horizon) were collected at random locations along three transects. Four to nine subsamples per plot were collected in 2004, and four were collected in 2006. Roots were hand-washed using a 2 mm sieve and sorted into three diameter size classes, fine (0.5–1 mm), medium (1–2 mm), and coarse (>2 mm), then dried to a constant mass at 60°C and weighed. Very fine root biomass (<0.5 mm) was determined from the remainder of the sample using the line-intercept method (Hendrick and Pregitzer 1993), where root biomass was estimated from measures of root length and of specific root length (cm g^{-1}).

The distribution of root biomass between the O and A horizons was determined for three of the cores collected in 2004 using the same method as described above. We further sorted the very fine root fraction into hemlock and non-hemlock, and we grouped fragmented pieces of roots that could not be positively identified into an unknown root category. All roots were dried to a constant mass and weighed. Hemlock roots were identified under a dissecting microscope based upon a pinnate branching pattern, color, and the presence of an ectomycorrhizal mantle. Ectomycorrhizal roots of hemlock were distinguished from those of another pinnately branched ectomycorrhizal host (*Betula spp.*) based on the diameter of pinnate root orders (main axis of hemlock roots have a larger diameter than root tip), after an examination of seedling root systems from the field.

Soil Abiotic Variables and CO_2 Efflux

Soil CO_2 efflux (E_{soil}, μmol m^{-2} s^{-1}) was measured monthly from May through October in 2004–2006 (LI-COR 6400-09, LI-COR Inc., Lincoln, NE) for a total of 125, 168, and 222 individual measurements for 2004, 2005, and 2006, respectively. PVC soil collars were permanently installed at each subplot in May 2004. During each sampling, soil temperature (T_{soil}, °C, LI-COR 6400-09, LI-COR Inc., Lincoln, NE) and volumetric soil moisture (θ, % v/v, Field Scout TDR 100, Spectrum Technologies, Inc.) were measured at 0–15 cm depth.

To make inferences in our plots during the study period while controlling for the effects of T_{soil} and θ, we constructed an empirical model predicting E_{soil} ($\hat{E}_{soil}$) as a function of T_{soil} and θ (Proc NLIN, SAS). The model was parameterized using data collected during 2004 (before infestation was observed, 62 measurements) in the HWA plots. Because the relationship between E_{soil} versus T_{soil} increased exponentially, and the relationship between E_{soil} versus θ decreased exponentially, our model had the following form:

$$\hat{E}_{soil} = \beta_0 \cdot e^{\beta_1 T_{soil}} \cdot e^{\beta_2 \theta}, \quad (1)$$

where β_0, β_1, and β_2 were fitted coefficients. Approximate R^2 was calculated as 1-(sum of squares residual/uncorrected total sum of squares). Coefficients were interpreted as significantly different from zero if their approximate 95% confidence intervals did not overlap zero. Normalized coefficients were estimated as the product of the coefficient and the ratio of the approximate standard deviations of the coefficients (for example, $\beta^*(SD\beta_1/SD\beta_2)$). This model represents E_{soil} in our hemlock stands before observation of HWA infestation or perturbation from girdling, and incorporated the influences of temperature and moisture on E_{soil}.

To test for significant departures in E_{soil} over time due to the treatments, we used the above model parameterized with 2004 data, along with measured T_{soil} and θ during other times (for example, 2005 and 2006) to predict ''expected'' E_{soil}, that is, $\hat{E}_{soil}$. We excluded values of T_{soil} and θ that were outside the range of values used to parameterize the model. The percentage of measurements excluded in the analysis was 13% in 2004 (in the GDL treatment) and 28% in 2005 and 2006. Departures from expected, calculated as observed (E_{soil}) minus expected ($\hat{E}_{\text{soil}}$) divided by expected, were then compared.

Statistical Analysis

We tested for treatment and time effects in the response variables above using a split-plot design (PROC GLM, SAS) to account for non-independent, repeated measurements over time. Treatment was the whole-plot factor and time was the split-plot factor for the following response variables: BAI, leaf and branch litter, root biomass, soil abiotic variables and E_{soil}. For O horizon mass, we only had one pooled measurement for 2006, and therefore we tested for treatment and horizon effects using a split-plot design to account for non-independent, repeated measurements within the O horizon. Treatment was the whole-plot factor and O horizon fraction (that is, Oi or Oe + Oa) was the split-plot factor. We used a post-hoc means separation technique to determine differences among all factor combinations. All percentage or fractional data were arcsin or arcsin-square root transformed due to lack of independence between the mean and the variance in these types of data (Dowdy and Wearden 1991). We transformed other variables to meet normality assumptions if necessary. We interpreted differences as significant at the $\alpha = 0.10$ level.

RESULTS

Aboveground Indicators of Decline

Our aboveground measurements generally supported our first hypothesis; hemlock in the GDL plots declined more rapidly than those in HWA plots as indicated by BAI and leaf litter inputs. Compared to nearby hardwoods, hemlock trees had significantly lower BAI over time ($F_{2,12} = 14.82$, $P < 0.01$), regardless of whether they were girdled or infested with HWA (Figure 2A). However, as expected, hemlock BAI in the GDL plots declined more rapidly over time compared to those in the HWA plots (treatment by time interaction $F_{2,12} = 6.86$, $P = 0.01$). By the third year of infestation or after girdling, hemlock trees in those respective plots were not different from one another and only grew 0–13% of that of the nearby hardwood trees. Co-occurring hardwoods in both the GDL and HWA treatment plots showed indications of increased growth in response to the treatments 2 years after girdling and infestation (no treatment effect, but a significant year effect, $F_{6,12} = 4.53$, $P = 0.03$; Figure 2B).

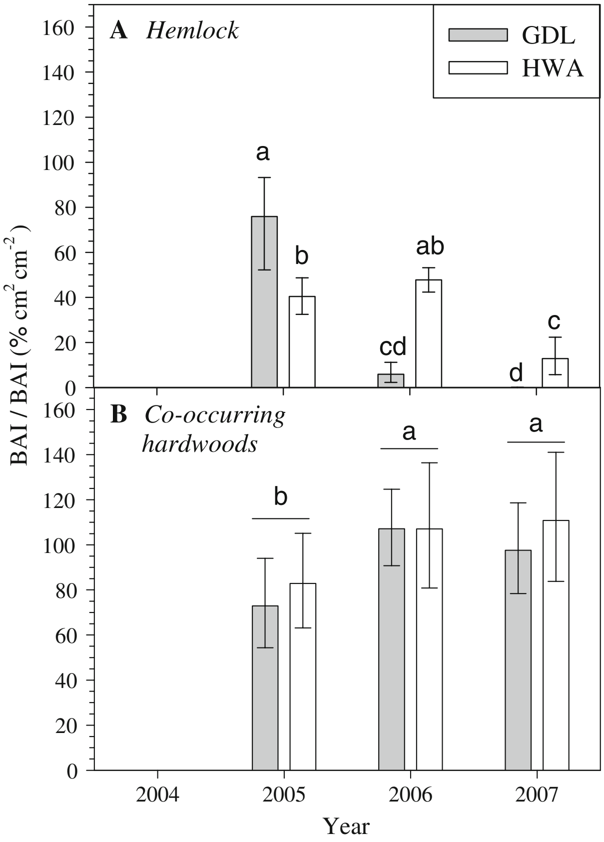

Figure 2. Mean annual basal area increment (BAI) of hemlock trees (**A**) and co-occurring hardwood trees (**B**) in girdled (GDL) and HWA treatments relative to the BAI of nearby hardwood trees in a stand with less than 3% hemlock basal area. Bars denote standard error. Different letters denote statistically significant differences among treatments and years in (**A**) and among years in (**B**) ($\alpha = 0.10$). Girdling occurred in July 2004. HWA infestation was observed in December 2004.

Needles and leaves were the major components of canopy litter fall, constituting 75–80% of the litter mass. In HWA plots, hemlock needles represented 25% of the total leaf litter, which was consistent over the 3 years of the study. In the GDL plots, however, hemlock leaf litter accounted for 35% of total litter in 2005 and declined to only 7% of the total litter in 2006.

Although we lack litter fall data from 2004, data from 2005 and 2006 support our hypotheses of hemlock decline in response to HWA and girdling. Hemlock leaf litter in the GDL plots was 1.5 times greater than that in the HWA plots in 2005 ($P = 0.06$). By 2006, this trend had reversed; hemlock leaf litter in the GDL plots was only 35% of that collected in the HWA plots ($P = 0.04$, Figure 3A). In the HWA plots, hemlock leaf litter was relatively consistent between years, and did not

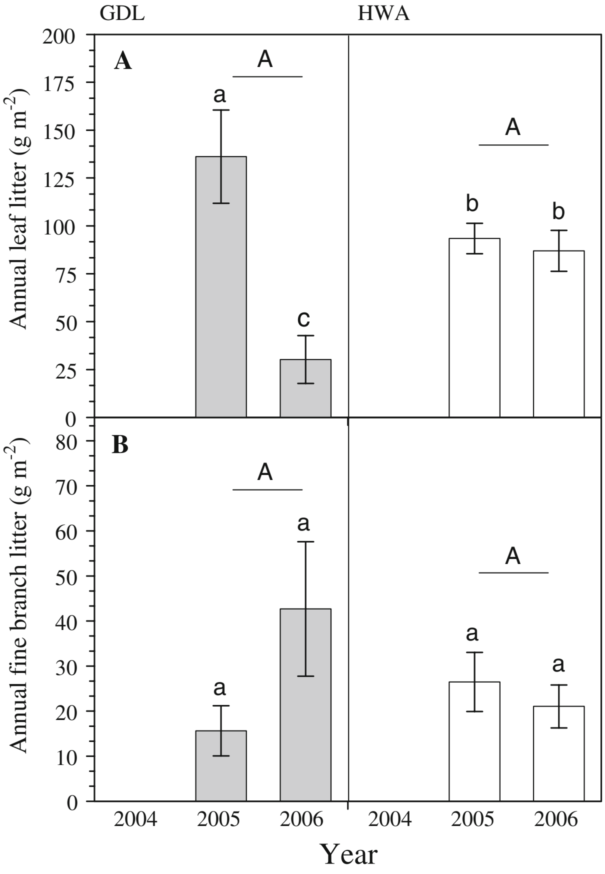

Figure 3. Mean annual hemlock leaf (**A**) and fine branch (**B**) litter fall in girdled (GDL) and HWA treatments during the study period. Bars denote standard error. Different uppercase letters denote statistically significant differences between treatments and different lowercase letters denote differences between treatments and years ($\alpha = 0.10$). Girdling occurred in July 2004. HWA infestation was observed in December 2004.

decrease significantly over time, as it did in the GDL plots (treatment by time interaction, $F_{1,6} = 14.13$, $P < 0.01$). Fine branches were a smaller component of the total litter fall, constituting roughly 10% of the total litter mass. In the GDL plots, fine branch litter fall was 2.7 times greater in 2006 than 2005, but remained relatively constant in the HWA plots. Hemlock fine branch litter did not differ significantly over time ($F_{1,6} = 1.04$, $P = 0.35$) or between treatments ($F_{1,6} = 0.62$, $P = 0.46$, Figure 3B).

O Horizon

Mass of the Oi and Oe + Oa horizons did not differ significantly between treatments in 2006 (Table 1), and did not support our second hypothesis that O horizon mass would increase in the GDL plots relative to the HWA plots. For both treatments, the Oi horizon had less mass than did the Oe + Oa horizon ($F_{1,6} = 133.15$, $P < 0.01$). Neither percent carbon nor g C m^{-2} of the O horizon differed between treatments (Table 1).

Belowground Indicators of Decline

We found partial support for our third hypothesis that in GDL plots belowground indicators of mortality would progress more rapidly than those in the HWA plots. Although very fine root biomass and E_{soil} both declined over time for both the GDL and HWA treatments, unexpectedly, the two treatments were not significantly different from each other.

Total root biomass did not differ between GDL and HWA treatments. In general, most root biomass was in the coarse and very fine categories (Table 2, Figure 4), accounting for 32% and 45% of the total belowground biomass, whereas the medium and fine root categories accounted for 14% and 10% of the total, respectively. Coarse, medium and fine root biomass did not differ between treatments (Table 2). Very fine root biomass decreased significantly over time by 38% and 22% in the GDL and HWA plots ($F_{1,6} = 3.91$, $P = 0.09$, Figure 4) and there was no significant interaction

Table 1. Mean Plot Oi and Oe + Oa Horizon Mass and Percent Carbon (% C) Sampled in 2006[1] in Girdled (GDL) and HWA Plots

	GDL		HWA	
	g m^{-2}	%C	g m^{-2}	%C
Oi	581.60 (55.98) a	46.76 (1.63) a	529.10 (74.86) a	47.73 (2.23) a
Oe + Oa	1737.32 (167.74) b	46.51 (1.10) a	1551.32 (135.78) b	45.14 (1.60) a
Oe + Oa (AFDM)[2]	1476.79 (208.95)		1284.89 (53.87)	

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[1] *Girdling occurred in July 2004. HWA infestation was observed in December 2004.*

[2] *AFDM denotes ash-free dry mass.*

Plot values were the average of six samples collected from three seasons from four experimental plots per treatment (n = 4) in 2006. Standard error shown in parentheses. No significant treatment effect was detected. Different letters within a column denote significant differences between Oi and Oe + Oa horizons.

Table 2. Mean Standing Biomass (g m^{-2}) of Coarse (>2 mm), Medium (1–2 mm), and Fine (0.5–1 mm) Root Categories in Girdled (GDL) and HWA Plots

	GDL[1]		HWA[2]	
	2004	2006	2004	2006
Coarse	392.91 (115.49)	454.24 (110.89)	612.56 (114.76)	839.51 (104.55)
Medium	224.95 (56.33)	209.66 (29.64)	208.40 (18.76)	235.76 (10.50)
Fine	147.71 (17.08)	143.27 (17.64)	187.17 (27.99)	145.33 (9.57)

[1] *Girdling occurred in July 2004.*
[2] *HWA infestation was observed in December 2004.*
Standard errors are shown in parentheses. No significant main effects (treatment or time) or interactions were detected for any root category.

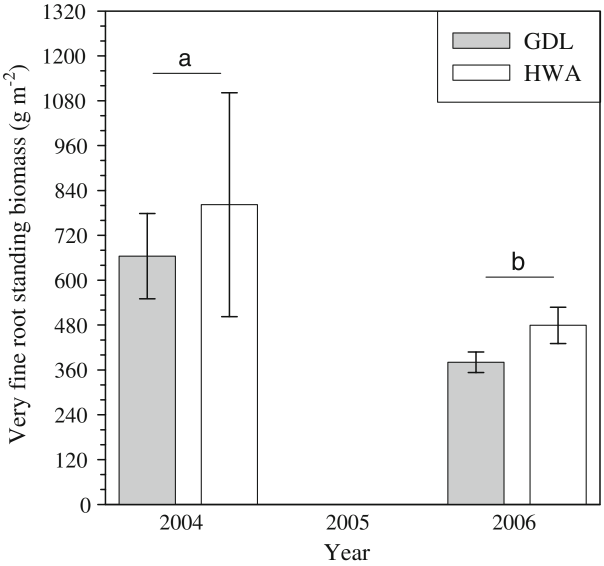

Figure 4. Mean very fine root (<0.5 mm diameter) standing biomass for girdled (GDL) and HWA treatments during 2004 and 2006. Bars denote standard error. The treatment effect was not significant. Different lowercase letters denote a significant year effect across treatments ($\alpha = 0.10$). The treatment by year interaction was not significant. Girdling occurred in July 2004. HWA infestation was observed in December 2004.

between treatment factors. Biomass did not decline more rapidly in the GDL plots compared to the HWA plots as we had predicted, however. In a subsample of three cores collected in 2004, hemlock roots represented at least 32%, and as much as 66% (including unknown root fragments), of very fine root biomass in each soil core (data not shown). Just over one-half of hemlock very fine root biomass occurred in the O horizon (54% versus 44% in the A horizon). In contrast, the very fine root biomass of non-hemlock species in the stand was preferentially distributed in the A horizon (63% versus 37% in the O horizon) (data not shown).

Soil CO_2 efflux ranged 0.4–12.2 µmol m^{-2} s^{-1} and was highly variable within plots (38% CV). Our measured values of E_{soil} did not differ significantly between treatments ($F_{1,6} = 0.38$, $P = 0.56$), but declined significantly over time ($F_{2,12} = 5.55$, $P = 0.02$; Figure 5A) across both treatments. We found that soil moisture negatively affected efflux, whereas temperature positively affected efflux in the HWA plots in 2004. Within plots, variability in soil moisture was high (21% CV), whereas variability in soil temperature was relatively consistent (<2% CV). In the HWA plots in 2004, soil moisture ranged from 15 to 62% and soil temperature ranged from 12.7 to 18.9°C. Over the course of the study, neither soil moisture nor temperature varied significantly between treatments, but both varied significantly among years ($F_{2,12} = 12.97$, $P < 0.01$ and $F_{2,12} = 29.66$, $P < 0.01$, respectively). In 2006, soil moisture was significantly lower than it was in 2004 and 2005, reflecting the pattern of annual precipitation for the three study years (2,160, 2,320, and 1,691 mm). Soil temperature increased significantly from 2004 to 2005, and decreased in 2006. Because soil moisture and temperature varied significantly over the study period, we estimated E_{soil} given the abiotic conditions in 2005 and 2006 from the predictive model that was parameterized from 2004 data (before HWA infestation was observed) (Figure 5B). The model had the following form and parameters ($R^2 = 0.89$, $P < 0.01$):

$$\hat{E}_{soil} = 0.5266 \cdot e^{(0.152 \cdot T_{soil})} \cdot e^{(-0.015 \cdot \theta)}. \qquad (2)$$

The coefficients for T_{soil} and θ were significantly different than zero. The normalized coefficients for T_{soil} and θ were 0.868 and −0.003, indicating that the relative effect of T_{soil} was stronger than that of θ. Values of $\hat{E}_{soil}$ revealed that abiotic variables were not entirely responsible for driving patterns in E_{soil} between treatments and over time. Therefore, the relative difference between E_{soil} and $\hat{E}_{soil}$ over

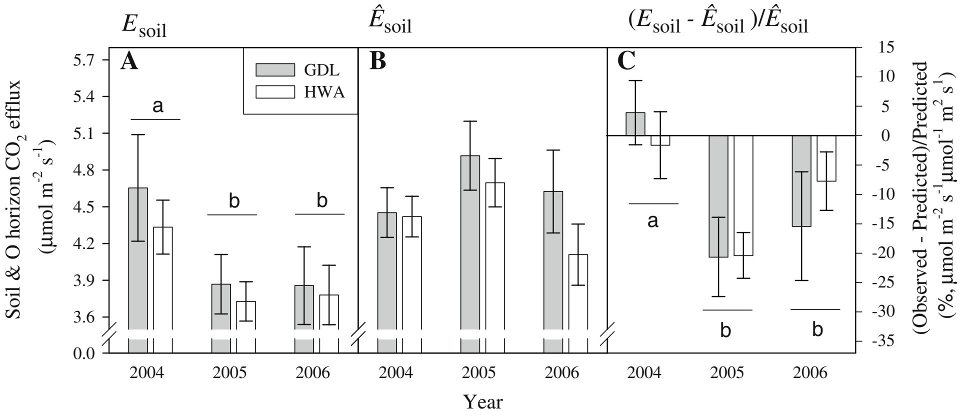

Figure 5. Observed mean soil CO$_2$ efflux (E_{soil}) from soil plus O horizon from the girdled (GDL) and HWA treatments (**A**). Predicted responses ($\hat{E}_{soil}$) in (**B**), and the deviance from predicted values (**C**). Bars denote standard error. Different letters in (**A**) and (**C**) denote significant differences among years. Girdling occurred in July 2004. HWA infestation was observed in December 2004

time was a means to better isolate treatment effects from abiotic effects (Figure 5C). We found that observed values of E_{soil} in the HWA and GDL plots did not differ between treatments ($F_{1,6} = 1.27$, $P = 0.30$), but were significantly lower than those predicted in 2005 and 2006 (time effect, $F_{2,12} = 7.90$, $P < 0.01$; Figure 5C).

DISCUSSION

Understanding how the loss of a foundation species affects the forest carbon cycle is critical for predicting the short- and long-term impacts of disturbance on forest ecosystems. The goal of the present study was to compare the decline of hemlock undergoing a girdling treatment with that of hemlock infested with HWA and to track the process of hemlock mortality to changes in the forest carbon cycle. We expected that the decline of girdled trees would out-pace that of HWA-infested trees because girdling terminates photosynthate supply, and because of the documented gradual decline of hemlock from HWA in the northeast. Relative to hardwood species in neighboring stands, the BAI of girdled hemlock declined rapidly over the course of our study. However, the BAI of HWA-infested trees also reduced during this period. Across both treatments, hemlock trees declined by approximately 85% relative to neighboring hardwoods in the girdled and HWA-infested stands between the first and third years after girdling and HWA infestation. In contrast, co-occurring hardwoods in the same stands increased by approximately 40% during the same time period. Although there are no data from hemlock stands prior to HWA infestation to function as a baseline, the reductions in hemlock productivity were concurrent with HWA infestation; and these reductions in productivity had direct consequences on the carbon cycle, which in most cases were similar to those induced by the girdling of trees.

Canopy litter fall progressed differently over time for each treatment. In the girdled treatment, most of the canopy leaves fell in the 2 years following girdling, and the crowns were bare by the end of the second year. These results are similar to those of other studies of evergreens retaining two to three cohorts of needles; studies report elevated litter inputs within a year, and complete litter fall within 2 years of girdling (Schroeer and others 1999; Yorks and others 2003). In the girdled plots, litter fall of fine branches peaked the year after the greatest inputs of needle fall, showing the general progression toward mortality. In contrast, leaf and fine branch litter fall in the HWA-infested plots were relatively consistent in the first and second years after infestation. Other studies have reported either reduced needle production or a termination of needle production with HWA infestation (McClure 1991; McClure and Cheah 1999). Even if hemlock ceased producing new needles at the onset of HWA infestation in the first year of the study, declines in litter fall would remain undetected until 4 years later, because hemlock needle longevity is approximately 3 years (Whitney 1982). In other research plots in the Coweeta basin, hemlock trees had 80% crown loss by the third year of HWA infestation (K. Elliot, unpublished data; USDA 2005). These results suggest that reductions in hemlock needle production would be apparent in litter fall rates in the fourth year of HWA infestation.

Although we detected contrasting patterns in the annual rate of leaf litter fall between girdled and HWA-infested stands, we did not observe a difference in Oi horizon mass between treatments at the end of the study. In fact, in spite of annual differences in inputs, cumulative leaf litter inputs were similar between treatments over the study period. The elevated mass of the Oi horizon in the girdled plots compared to the HWA plots may reflect the momentum of increasing fine branch litter fall we observed the second year after girdling hemlock. In the future, we expect dramatic and persistent increases in O horizon mass and hence, soil carbon content, from continuous inputs of branch and stem litter (Orwig and Foster 1998; Eschtruth and others 2006). Our measures of total O horizon mass are within, or above the range reported for southern Appalachian hemlock forests with an *R. maximum* component (1,606–2,240 g m^{-2}) (Boettcher and Kalisz 1990), a species that can promote O horizon formation greater than 3,000 g m^{-2} (Wurzburger and Hendrick 2007). Over the long term, the substantial O horizon masses created by hemlock are likely to be maintained, or enhanced, if *R. maximum* is present in the subcanopy. With the replacement of hemlock by hardwood species, however, O horizon mass will likely reduce slowly over time.

Mirroring the decline in hemlock BAI, very fine root biomass decreased over time in our study plots. Although the treatments were not significantly different from each other, the decline in very fine root biomass in the girdled plots (38% ± 10%) was greater than that in the HWA-infested plots (22% ± 18%). Hemlock accounted for 32–66% of very fine root biomass from a subset of samples collected in the first year prior to treatment conditions. Therefore, we suggest that the decline in very fine root biomass reflects a significant loss of the hemlock component of very fine root biomass in both treatments. In support of this idea, very fine root biomass in neighboring mixed hardwood stands (<3% hemlock), remained constant over the same time period (Nuckolls 2007). Because girdling terminates belowground carbon allocation, we expected a substantial decrease in hemlock very fine root biomass in the girdled treatment, reflecting the mortality and decomposition of hemlock roots after the trees depleted their carbohydrate stores. In contrast, in HWA-infested plots, we expected that a decline in very fine root biomass would reflect a reduction in hemlock belowground production. Although decreased root production is not surprising for infested hemlock, the magnitude of the decline in biomass suggests a rapid termination in very fine root production within 2 years of HWA infestation. Surprisingly, a decline in standing biomass was not detected in the fine, medium or coarse root size categories for either treatment, suggesting that these root classes are slower to die or decompose. Detrital inputs from these larger root categories are likely to be delayed relative to inputs of very fine root mass. Interestingly, despite large inputs of root litter over the course of hemlock mortality, standing stocks of root biomass may be reduced for only a brief period in these mixed forests as the root production of co-occurring species increases during hemlock decline (Schroeer and others 1999; Jones and others 2003).

Soil CO_2 efflux reflects primary productivity directly, through root respiration, and indirectly, through root exudation and mortality; and we therefore expected that girdling and HWA infestation would lead to reductions in E_{soil}. As predicted, E_{soil} declined in the first and second years after girdling and HWA infestation; however, climate also varied significantly over time. By estimating values of E_{soil} under these climatic conditions, we more effectively isolated the response of E_{soil} to girdling or HWA infestation. In the first year after girdling and HWA infestation, E_{soil} was approximately 20% lower than our predictions for both treatments. Similar to our results, Binkley and others (2006) found that girdling half of a forest stand reduced E_{soil} by 20%. In other studies where investigators girdled all overstory trees, E_{soil} was reduced by 31–60% in the first year (Scott-Denton and others 2006; Ekberg and others 2007). The lack of a treatment difference in our study suggests that within 1 year of HWA infestation the decline of hemlock results in a similar reduction in E_{soil} as does terminating photosynthate supply via girdling. Reduced belowground productivity and associated reductions in root respiration and exudation may explain the strong decline in E_{soil} within 1 year. The reduction in E_{soil} and the concurrent decline in BAI and standing very fine root biomass suggest rapid declines in hemlock productivity from HWA infestation. E_{soil} may increase over pre-infestation levels after mortality of infested trees occurs, when coarse root detritus and enhanced O horizon masses further increase decomposition rates.

CONCLUSIONS

We observed declines in BAI production, very fine root biomass and E_{soil} as rapidly as 1 year after HWA infestation, and these responses were generally similar in magnitude and timing as those induced by girdling. Therefore, HWA infestation rapidly impacts the carbon cycle in these stands. No studies have directly compared the decline of hemlock stands from HWA infestation in the northeast with those in the southern Appalachians. Our results suggest that hemlock decline is progressing more quickly in the southern extent compared to the northern extent of this species' range, wherein studies report protracted decline lasting more than 9 years (Eschtruth and others 2006). The reasons for this may include differing climatic extremes throughout the species' range, such as the infrequent occurrence of severe winter temperatures (−25°C) in the southern Appalachians that suppress HWA populations (Skinner and others 2003). We know little about how the loss of hemlock will influence forest processes and biogeochemical cycling in the southern Appalachians. Evidence from the northeast suggests that HWA itself promotes nutrient inputs into these systems (Stadler and others 2005); however, these effects may be contingent upon prolonged (that is, a decade or longer) infestation of the insect in slowly declining stands, a phenomenon that may not occur in the southern Appalachians. Therefore, the patterns of enhanced nutrient inputs observed in the northeast may not be a logical anticipation within the next decade for hemlock stands in the southern extent of the species' range. A more likely scenario for the southeast is that accelerated inputs of detritus from dead trees will impact carbon and nutrient cycling, and the subsequent competition for nutrients among regenerating species. *R. maximum* may be favored in soils of high organic content and low nutrient availability, which may characterize much of the landscape after widespread hemlock mortality in the southern Appalachians. The potential for *R. maximum* to replace hemlock, and the strong influence of this shrub on nutrient cycling and forest succession, will further distinguish moderate to longer term forest processes in the southern Appalachians from those in the northeast.

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