

RESEARCH ARTICLE

Forest mycorrhizal dominance depends on historical land use and nitrogen-fixing trees

Nina Wurzburger¹  | Katherine J. Elliott² | Chelcy Ford Miniatt² ¹Odum School of Ecology, University of Georgia, Athens, Georgia, USA²USDA Forest Service, Southern Research Station, Coweeta Hydrologic Laboratory, Otto, North Carolina, USA

Correspondence

Nina Wurzburger

Email: ninawurz@uga.edu

Present address

Chelcy Ford Miniatt, USDA Forest Service, Rocky Mountain Research Station, Albuquerque, New Mexico, USA

Funding information

National Science Foundation, Grant/Award Number: DEB-1440485 and DEB-1637522

Handling Editor: David Moreno Mateos

Abstract

1. Most forests are recovering from human land use, making it critical to understand the effect of disturbance on forest recovery. Forests of the eastern United States have a long history of land use, but it is unknown whether historical disturbances have contributed to their transition from ectomycorrhizal (ECM) to arbuscular mycorrhizal (AM) tree dominance. Disturbance may promote nitrogen (N)-fixing trees in early succession, which can elevate soil N availability even after they die. Higher soil N availability may facilitate the competitive success of AM trees over ECM trees, but such 'N fixer founder effects' have not been empirically tested.
2. Here, we analysed data from three land-use disturbances in a temperate forest historically dominated by ECM trees: selective-cutting (ranging from 0 to 52 m² ha⁻¹), clear-cutting and agricultural abandonment. These disturbances occurred at different times, but long-term data capture 3–7 decades of forest recovery.
3. We found that the AM tree fraction in contemporary forests was 2, 4, and 6-fold higher following selective-cutting, clear-cutting and agricultural abandonment, respectively, compared to forest composition in 1934. Across these disturbances we also observed an increasing abundance of the N fixer black locust immediately following disturbance. Using a simulation model parameterized by data from black locust, we estimated historical rates of symbiotic N fixation to understand the relationship between N fixation and AM dominance in individual plots. We found that N fixation was positively associated with the growth of ECM trees generally, and oak and hickory specifically, only following light selective-cutting (<12 or <18 m² ha⁻¹ basal area extraction, respectively). Following higher levels of selective-cutting and clear-cutting, N fixation was positively associated with the growth of AM trees, particularly red maple and tulip poplar. Agricultural abandonment led to AM dominance regardless of N fixation rates.
4. *Synthesis and applications.* Our findings suggest that common land use practices and black locust, a native N fixer, can reduce the dominance of ECM trees. If N fixers are likely to proliferate following disturbance, we might maintain ECM

This is an open access article under the terms of the [Creative Commons Attribution-NonCommercial-NoDerivs](https://creativecommons.org/licenses/by-nc-nd/4.0/) License, which permits use and distribution in any medium, provided the original work is properly cited, the use is non-commercial and no modifications or adaptations are made.

© 2023 The Authors. *Journal of Applied Ecology* published by John Wiley & Sons Ltd on behalf of British Ecological Society.

dominance by cutting trees at low densities and by applying prescribed fire to remove N.

KEYWORDS

biogeochemistry, ecosystem resilience, land use change, oak decline, symbiotic nitrogen fixation, timber harvest

1 | INTRODUCTION

A majority of the world's forests are recovering from agriculture and timber harvesting (Curtis et al., 2018), making it critical to resolve how prior land use affects ecosystem services. Key ecosystem services—carbon uptake and storage and fresh water supplies—depend on whether forests are resilient to land use, such that they return to their pre-disturbance state, or whether they transition to an alternative stable state (Perring et al., 2016). One potential component of forest resilience are tree species that associate with nitrogen (N)-fixing bacteria. These N fixers tend to proliferate following disturbance, and can alleviate N limitation in the ecosystem by replacing the N lost from disturbance. However, the role of N fixers on forest recovery has not been holistically evaluated within the context of ecosystem resilience, making their role in recovery or state transitions unclear.

The mycorrhizal strategies of trees are central to understanding the role of N fixers in forest resilience. Tree species generally associate with one of two mycorrhizal types (Steidinger et al., 2019) that respond to, and promote, ecosystem properties (Lu & Hedin, 2019; Phillips et al., 2013). Where forests are dominated by arbuscular mycorrhizal (AM) trees, rapid rates of decomposition and nutrient cycling promote more available, mineral forms of nutrients (Phillips et al., 2013) which then favour the AM strategy to scavenge nutrients (Lu & Hedin, 2019). On the other hand, where forests are dominated by ectomycorrhizal (ECM) trees, slower decomposition rates and the dominance of organic forms of nutrients (Phillips et al., 2013) favour the more carbon-costly ECM strategy to mine organic nutrients (Lu & Hedin, 2019). In warm, humid, temperate forests, these mycorrhizal strategies appear to represent alternative stable states (Averill et al., 2022; Steidinger et al., 2019), where the dominant mycorrhizal strategy facilitates a nutrient regime that competitively excludes the other in the long term (Lu & Hedin, 2019). However, state changes are possible—a change to a forest's nutrient regime can make it vulnerable to a mycorrhizal transition.

The role of N fixers in resilience may, therefore, depend on the dominant mycorrhizal association of a forest prior to disturbance. N-fixing trees have the potential to locally enrich soils in N, increasing N availability for at least a decade following their mortality (Von Holle et al., 2013). Such an N enrichment may promote the competitive success of AM trees over ECM trees, suggesting that N fixers can cause biogeochemical founder effects on ecosystem recovery (Lu & Hedin, 2019). Therefore, N fixers may facilitate the resilience of AM dominated forests that undergo disturbance, but they have

the potential to trigger a state change in ECM dominated forests if they elevate N availability sufficiently above the forest's former nutrient regime.

A consideration of how N fixers facilitate forest recovery is particularly relevant to the forests of the eastern United States. These forests have repeatedly experienced land-use disturbances, including agriculture and timber harvesting since European colonization (Pan et al., 2011). Following disturbance, black locust (*Robinia pseudoacacia* L.), an early-successional, N-fixing tree, proliferates in these forests due to the low-N and high-light environment, and can fix a substantial amount of N (Boring & Swank, 1984; Wurzbürger et al., 2022). It is unknown whether black locust can influence subsequent patterns in forest recovery, such as a shift from ECM to AM forest dominance, but evidence suggests this possibility. First, black locust can favour the growth of AM trees over ECM seedlings in early succession (Minucci et al., 2019). Second, black locust can elevate soil N availability for decades following its death (Von Holle et al., 2013), suggesting the potential for lasting effects on forest recovery. Third, other mechanisms that increase soil N availability, atmospheric N deposition and fire exclusion (which provide inputs or prevent losses of N, respectively) are associated with an increase in the abundance of AM trees and a decline in ECM trees in eastern forests (Averill et al., 2018; Jo et al., 2019). Eastern forests may be particularly vulnerable to such a mycorrhizal transition because of the loss of the ECM tree American chestnut (*Castanea dentata* (Marsh.) Borkh.), which co-dominated these forests with oak (*Quercus* spp.) for millennia (Hanberry & Nowacki, 2016), and helped maintain their oligotrophic state (Ellison et al., 2005).

A transition from ECM to AM tree dominance in eastern forests has negative effects on several ecosystem services. Oak, the dominant ECM tree genus across the region, provides high quality timber, supports a complex food web, and is resilient to drought and fire (Nowacki & Abrams, 2008). The decline in oak and increase in AM tree species, such as red maple (*Acer rubrum* L.) and tulip poplar (*Liriodendron tulipifera* L.) can increase drought-induced declines in primary productivity (Hwang et al., 2014, 2020), reduce biodiversity (Elliott et al., 2020), and reduce streamflow (Caldwell et al., 2016; Elliott et al., 2017). Several factors have contributed to the decline in oak and the loss of ECM dominance, including atmospheric N deposition and fire exclusion (Averill et al., 2018; Jo et al., 2019; Nowacki & Abrams, 2008). Yet, prior research has not examined whether land use disturbance affects mycorrhizal dominance, and whether these effects are facilitated by N fixers. Such an analysis requires spatially-resolved tree data that capture forest disturbance and recovery over several decades.

Here, we evaluate the effect of land use on a mixed-deciduous eastern forest previously dominated by ECM trees. We examine one of the longest records of repeated vegetation surveys in a temperate forest to quantify changes in tree above-ground biomass, species composition, and mycorrhizal associations following three land-use disturbances: selective-cutting, clear-cutting and agricultural abandonment. All three of these disturbances were associated with biomass removal, and hence, a loss of N from the ecosystem, the most extreme case being the agricultural experiment, due to the burning of slash and soil tillage. However, the agricultural experiment also included the fertilization of N, which may have partially replenished these N losses. Our long-term data allow us to quantify N fixer abundance and estimate historical rates of symbiotic N fixation using a scaling framework we developed for these forests (Wurzburger et al., 2022). Our data are constrained to one 1626-ha basin, which allows us to examine the effects of land-use disturbances, where temporal changes in climate, atmospheric N deposition, and time-since-fire occurred uniformly across all plots within the basin. We hypothesized that (1) AM tree dominance in contemporary forests would increase from selective-cutting to clear-cutting to agricultural abandonment as the land use disturbance. We expected this pattern because (2) N fixer abundance, and therefore, symbiotic N fixation rates, would increase with increasing N losses and light availability, which both favour black locust's establishment and ability to fix N (Boring et al., 2014), and because (3) N fixers would favour the biomass growth of AM trees more than ECM trees, and lead to a transition from ECM to AM dominance over several decades of forest recovery.

2 | MATERIALS AND METHODS

2.1 | Study area

We analysed previously collected data from the Coweeta Hydrologic Laboratory, an experimental forest of the Southern Research Station, USDA Forest Service, located in the Nantahala National Forest, western North Carolina, USA (latitude 35°03'N, longitude 83°25'W). All Forest Service field studies carried out on Experimental Forest and Range (EFR) sites are covered by a Categorical Exclusion (CE) to the National Environmental Policy Act (NEPA). No permits were required, given the CE on the EFR. The Coweeta Basin encompasses 1626 ha, elevations range from 675 to 1592 m, and slopes range from 30% to over 100%. Mean annual temperature is 12.6°C and mean annual precipitation is 1800 mm (Swift Jr. et al., 1988). Soils are deep sandy loams underlain by folded schist and gneiss (Thomas, 1996).

2.2 | Vegetation plots and forest above-ground biomass

We used previously established permanent plots from three land use disturbances: selective-cutting, clear-cutting and agricultural

abandonment. The selective-cut disturbance represents much of the Coweeta Basin, which was selectively cut (basal area cut ranged 0–52 m² ha⁻¹ across plots) in the early 20th century (mid-point 1921), followed by the mortality of American chestnut in the 1930s–40s (Elliott & Swank, 2008). Following selective-cutting, permanent 0.08 ha (20 m × 40 m) plots were established in 1934 along 13 parallel, approximately north–south transects (330°) at 200 m intervals spanning the Coweeta Basin (Elliott et al., 1997; Swank & Crossley, 1988). We used the 274 plots that were re-measured in all subsequent surveys, the 1970s (1969–1973), 1990s (1988–1993) and 2010s (2009–2013); thus, vegetation survey data span 1934–2010. Plots with no basal area extraction offer a comparison for those that experienced higher levels of selective-cutting, or those that experienced subsequent disturbances.

The clear-cut and agricultural abandonment disturbances occurred subsequently and were established as small watershed-level experiments. In the clear-cut experiment (1976–1977), all boles were cut and removed, but slash remained on-site (Swank & Webster, 2014). Trees were inventoried before clear-cutting in 1974 from 142 (20 m × 40 m) plots, perpendicular to the Coweeta Basin permanent plot network. In subsequent years after clear-cutting (1979, 1984, 1993, 1997, 2008), 24 of these original plots were re-measured by placing two 7 m × 7 m subplots in opposite corners of each 0.08 ha plot (Elliott et al., 1997).

The agricultural abandonment experiment included logging, burning slash, and tilling soils in 1958, followed by planting and maintaining grass with fertilizer and herbicide for multiple years (total N inputs of 273 kg N ha⁻¹) until all treatments were abandoned in 1968 (Elliott et al., 2017; Hibbert, 1969). Vegetation surveys were conducted in 1934, 1982, 1995 and 2012. In 1934, five 0.08 ha permanent plots were measured along the east-side of the watershed; in all subsequent surveys, plots were placed across the entire watershed. In 1982, 34 plots (0.02 ha) were permanently marked continuously along five transects from ridge-top to near stream; these plots were re-measured in 1995 and 2012 (Elliott et al., 1998, 2017).

In all plots, stem diameter was measured on all trees ≥ 2.5 cm at conventional breast height (dbh, 1.37 m) and recorded by species. We used species-specific allometric equations (Martin et al., 1998) to calculate tree above-ground biomass. Because our data are not specific to individual trees over time, we cannot determine which demographic processes explain patterns in forest biomass change. At the time of the most recent surveys, time-since-disturbance was 89 years in the selective-cut plots, 31 years in the clear-cut plots, and 45 years in the agricultural abandonment plots.

2.3 | Symbiotic N fixation estimates

We estimated symbiotic N fixation by black locust by applying a Monte Carlo simulation model that scaled fixation rates from individual trees to plots and over time (Wurzburger et al., 2022) (see Appendix S1). Our simulation model was parameterized by predictions of black locust stem densities in each plot and age-specific

fixation data for black locust. Because these N fixation estimates are based on several assumptions (Wurzburger et al., 2022), we also analysed the effect of black locust using its maximum observed above-ground biomass in each plot (see Section 2.5).

2.4 | Atmospheric N deposition

Bulk N deposition (as ammonium and nitrate) has been quantified annually since 1972 at Coweeta (National Atmospheric Deposition Program, 2019). We estimated the range of N deposition across plots using estimated differences in annual precipitation across the elevational gradient in the Coweeta Basin (Elliott & Swank, 2008).

2.5 | Analysis of forest change

To understand changes in mycorrhizal dominance, we calculated the fraction of above-ground biomass comprised by AM trees, and fit a generalized additive model with a beta distribution (package: *MGCV*; Wood, 2011), including a smoothing spline on time for all three disturbances. To determine whether increases in AM dominance were related to symbiotic N fixation for each disturbance, we then analysed the net biomass change of AM versus ECM tree species as a function of cumulative N fixation in each plot. First, we fit linear mixed-effects models (package: *LME4*; Bates et al., 2015) to above-ground biomass per plot over time since disturbance for all common AM and ECM tree species (present in at least 10% of all plot by time observations; Table S11). We excluded rare species from this analysis because they prevented model convergence and accounted for ~2% of above-ground biomass. Random effects accounted for the repeated sampling of plots and tree species within each mycorrhizal association. We extracted model coefficients to calculate the mean net change in above-ground biomass for each mycorrhizal type over time since disturbance in each plot (Mg ha^{-1}). We then analysed these changes as a function of mycorrhizal type and cumulative N fixation (and their interaction) using linear regression. In the selective-cut disturbance, we included basal area extraction and American chestnut basal area in 1934 as variables (and their potential interaction with cumulative N fixation). We also included a terrain shape metric (Elliott et al., 1999), a continuous, unitless variable derived from GIS, to account for the effect of topographic position on tree growth. We excluded the biomass of black locust and American chestnut in all our analyses of biomass change because their abundances were indirectly (or directly) included as variables in our model. We also excluded eastern hemlock (*Tsuga canadensis* (L.) Carrière) because it has been functionally extirpated from Coweeta over the last two decades. We dropped non-significant interaction terms from final models. We repeated the above analysis using the maximum observed above-ground biomass of black locust in each plot instead of our estimates of cumulative symbiotic N fixation, and found qualitatively similar results (Tables S4–S6). We then determined if biomass change of the two most abundant tree species or

genera of each mycorrhizal association—red maple and tulip poplar (AM) and oak and hickory (*Carya* spp.; ECM) were associated with cumulative N fixation using the same approach.

To understand the relationship between the net change in total above-ground biomass and cumulative N fixation, we analysed data from each disturbance using linear regression following the same approach as above. For all analyses, we square-root transformed variables when necessary to achieve normality and improve the error distributions of models, and backtransformed predictions for graphical presentations. All analyses were conducted in R version 4.0.2 (R Core Team, 2020).

3 | RESULTS

We first examined how mycorrhizal dominance has changed in the Coweeta Basin over the last century and found that the more intense the disturbance, the greater the dominance of AM trees. Following selective-cutting, clear-cutting and agricultural abandonment, the predicted fraction of AM trees increased by 2.2, 3.8, and 6.0-fold since 1934, the beginning of our record (Figure 1a). While these disturbance events differed in their timing (selective-cutting in 1921; clear-cutting in 1977; agricultural abandonment in 1968), our data capture multiple decades of forest recovery, showing a relatively stable forest composition between the two most recent surveys. Thus, over the last century, land use appears to be facilitating a state transition from ECM to AM dominance, with the most rapid, complete transition occurring after agricultural abandonment (Figure 1a,b).

We then explored the role of black locust in these transitions in mycorrhizal dominance, and found that symbiotic N fixation rates were highest immediately following disturbance, and rapidly declined with time (Figure 2). This supports our hypothesis that increasing land use intensity would increase the amount of N fixed, due to the proliferation of black locust. The majority of N fixation occurred in the first two decades following each disturbance, declining below the rate of atmospheric N deposition within three decades.

Next, we tested our hypothesis that N fixation would favour the growth of AM trees more so than ECM trees. In the selective-cut disturbance, mean biomass gains of AM trees and mean biomass losses of ECM trees were greatest under high levels of cutting and high N fixation (mycorrhizal guild by N fixation by basal area extraction interaction: $F_{1,268}=14.22$, $p<0.001$; Figure 3a; Table S1) lending support for this hypothesis. Further, AM, but not ECM, trees gained more biomass in plots where American chestnut was more historically abundant (mycorrhizal guild by chestnut basal area interaction: $F_{1,268}=8.24$, $p=0.004$; Table S1). These relationships between mycorrhizal associations and disturbance suggest that increases in light or other resources after cutting or tree mortality stimulated the growth of AM trees more than N fixation did. However, for ECM trees, net biomass change increased with N fixation under low levels of cutting, but decreased under high levels of cutting (Figure 3a). In the clear-cut disturbance, we found an interaction between the

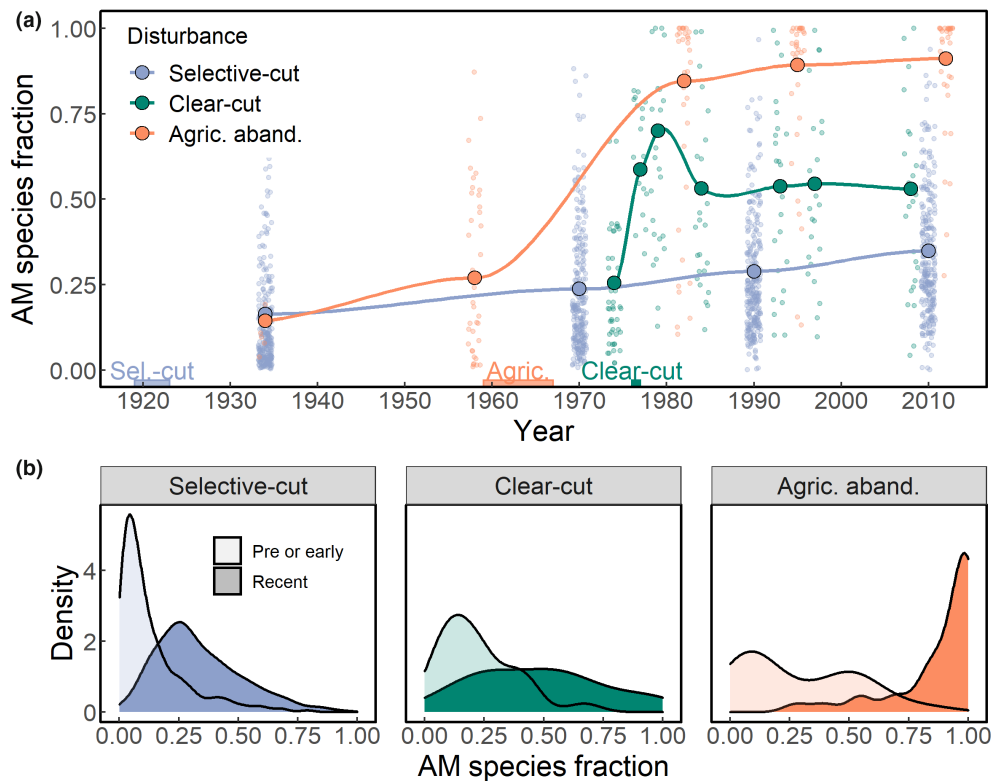


FIGURE 1 Changes in mycorrhizal dominance of Coweeta Basin forests following three land-use disturbances (selective-cutting, clear-cutting and agricultural abandonment), (a) the relative abundance of AM trees (the fraction of aboveground biomass) over time, and (b) shifting density distributions of the AM tree fraction from pre-disturbance (or early following disturbance) to recent surveys. In (a), each small point represents a measure of a long-term vegetation plot, and each curve represents smoothed fit of predictions from generalized additive models. Predicted values of AM fraction from each disturbance were significantly different ($p < 0.05$) from one another in 2008, the year of the most recent survey in the clear-cut disturbance. Repeated surveys, immediately following the clear-cut, capture the pulse of AM species typical of early succession.

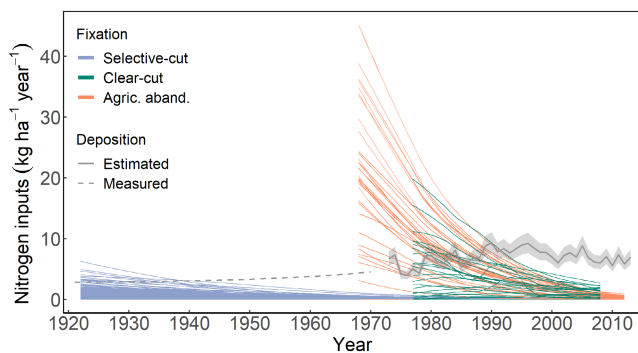


FIGURE 2 Nitrogen inputs from symbiotic N fixation following selective-cutting, clear-cutting and agricultural abandonment, and atmospheric N deposition at Coweeta. Each curve represents the estimated annual symbiotic N fixation rate for an individual long-term vegetation plot from a simulation model parameterized by age-specific fixation data. The solid grey line represents the median of bulk N deposition, and the ribbon represents the maximum and minimum across plots estimated by variation in precipitation across plots. Dashed line represents estimated N deposition, backcast from a relationship between Coweeta N deposition and global N emissions.

mycorrhizal association of trees and N fixation on net biomass change ($F_{1,22} = 5.06$, $p = 0.035$; Figure 3b; Table S2), where AM trees increased, while ECM trees decreased in biomass with increasing N fixation. In the agricultural abandonment disturbance, we found no such interaction with mycorrhizal association and N fixation, but biomass gains of AM trees far exceeded that of ECM trees ($F_{1,32} = 25.79$, $p < 0.0001$; Figure 3c; Table S3). We then conducted the same analyses above, replacing estimates of cumulative symbiotic N fixation with the maximum observation of black locust above-ground biomass in each plot, and found nearly identical results (Tables S4–S6). This verifies a relationship between black locust and mycorrhizal patterns in tree growth, in spite of the uncertainty associated with our estimates of symbiotic N fixation. However, it is possible that black locust may have affected non-fixing trees independently of N effects, for instance by altering pH or increasing phosphorus availability (Houlton et al., 2008).

We next assessed the net biomass change of two species groups—the dominant AM tree species red maple and tulip poplar, and the dominant ECM tree genera oak (*Quercus*) and hickory (*Carya*)—and found that their response largely followed the mycorrhizal responses as above. In the selective-cut disturbance, biomass

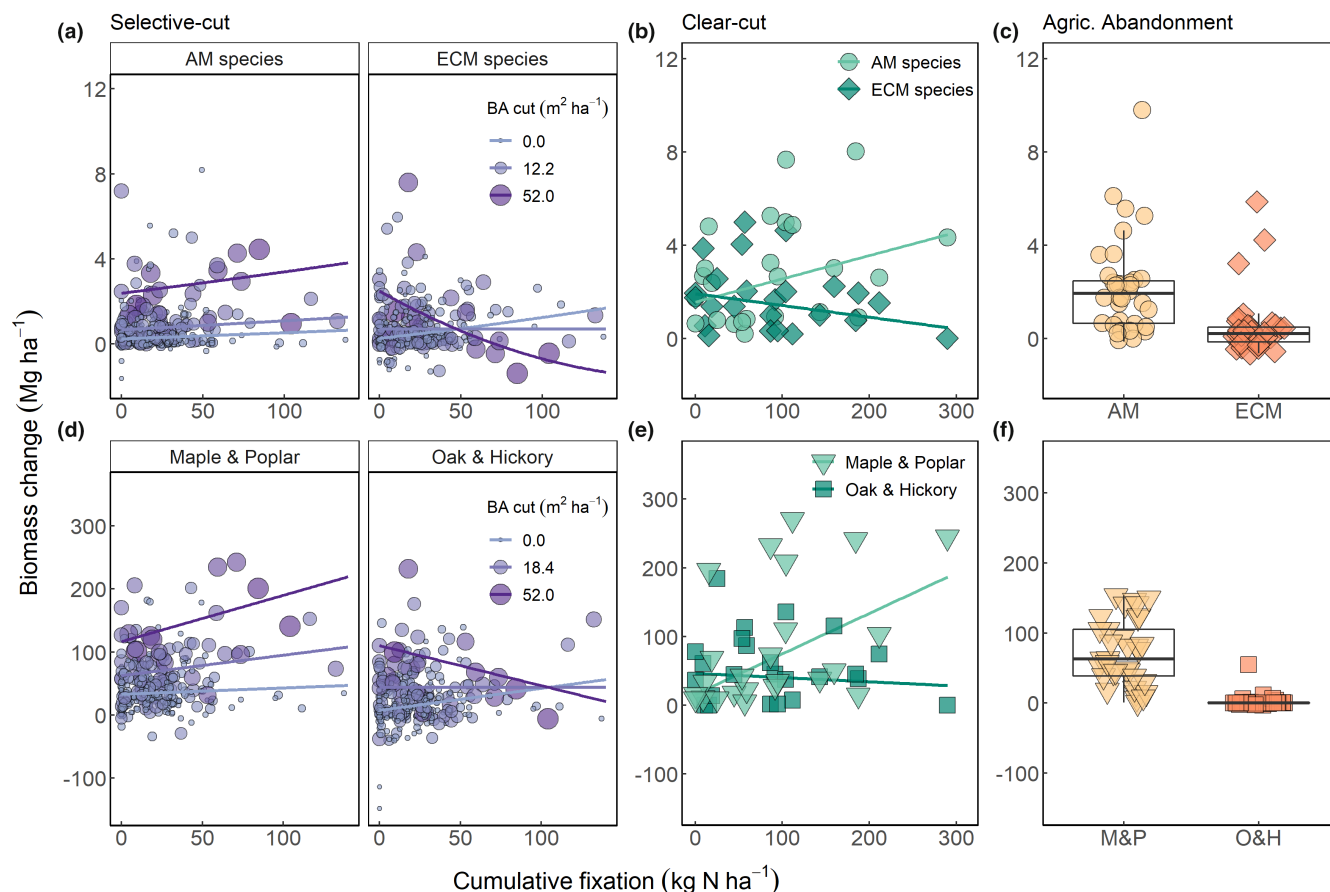


FIGURE 3 Net above-ground biomass change of AM and ECM tree species depends on land-use intensity and symbiotic N fixation. Following selective-cutting, the biomass of AM trees (a) or AM-dominants, maple and tulip poplar (d) increased with N fixation and basal area extraction (BA cut), while the biomass of ECM trees (a) or ECM-dominants, oak and hickory (d) increased with fixation at low levels of basal area extraction but decreased with fixation at high levels of extraction. Following clear-cutting, biomass of AM trees (b) or red maple and tulip poplar (e) increased while that of ECM trees (b) or oak and hickory (e) decreased with increasing fixation. Following agricultural abandonment, biomass of AM trees (c) or red maple and tulip poplar (f) was higher than ECM trees (c) or oak and hickory (f), and was not related to fixation. Each point represents biomass change in a long-term vegetation plot over time since disturbance and in (a), point size and colour denote the amount of basal area extracted from that plot. Lines are predictions of significant interactions ($\alpha < 0.05$), and in (a) and (d), predictions are of minimum and maximum basal area extraction, and an intermediate value where the slope was closest to 0, holding other variables at their median value. Boxplots in (c) and (f) show significant difference ($\alpha < 0.05$) between tree groups.

gains of maple and tulip poplar and the biomass declines of oak and hickory were greatest under high levels of logging and high N fixation (tree group by N fixation by basal area extraction interaction: $F_{1,270} = 11.39$, $p < 0.001$; Figure 3d; Table S7). At the highest level of selective-cutting and symbiotic N fixation, biomass gains of maple and tulip poplar were 10-fold higher than that of oak and hickory. In the clear-cut disturbance, maple and tulip poplar gained biomass while oak and hickory lost biomass under increasing N fixation (tree group by N fixation interaction: $F_{1,22} = 4.83$, $p = 0.039$; Figure 3e; Table S8), translating to a 7-fold difference in biomass gains at the maximum fixation rate. Following agricultural abandonment, maple and tulip poplar biomass gains far exceeded those of oak and hickory, regardless of N fixation (tree group: $F_{1,32} = 134.69$, $p < 0.0001$; Figure 3f; Table S9). Because these species groups accounted for at least 80% of total above-ground forest biomass, their divergent responses explain changes we observed in forest mycorrhizal dominance (Figure 1).

We then determined the relationship between symbiotic N fixation and the net change in total above-ground biomass over time, and found that it depended on land use disturbance. The net change in above-ground biomass was increasingly positive with increasing N fixation following low to moderate levels of selective-cutting, but was negative following high levels of selective-cutting (N fixation by BA extraction interaction: $F_{1,267} = 7.67$, $p = 0.006$; Figure S2a; Table S10). Net changes in forest biomass were not related to cumulative N fixation following clear-cutting ($p = 0.15$; Figure S2b), nor agricultural abandonment ($p = 0.43$; Figure S2c). In addition, biomass gains were greatest in selectively-cut plots that contained the most biomass of American chestnut prior to its mortality ($F_{1,267} = 21.7$, $p < 0.0001$; Table S10), highlighting the potential for changes in tree species and mycorrhizal dominance due to the loss of ECM-associating American chestnut. These findings suggest that N fixation only had a positive effect on net biomass gains under the mildest disturbance—the scenario where ECM tree growth was also facilitated by symbiotic N fixation (Figure 3a).

4 | DISCUSSION

Understanding how human land use affects ecosystem resilience is critical for predicting and managing ecosystem services (Perring et al., 2016; Seidl et al., 2016). While eastern forests of the United States have regrown after repeated disturbances, a transition from ECM to AM tree dominance has the potential to reduce a number of key ecosystem services for the region (Elliott et al., 2017; Nowacki & Abrams, 2008). Our work demonstrates that common land use practices favour the recovery of AM trees over ECM trees, suggesting that widespread and repeated land use (Pan et al., 2011) may have contributed to the transition from ECM to AM dominance in forests of the eastern United States. The increasing dominance of AM trees has been previously attributed to increased N availability due to atmospheric N deposition, fire exclusion and climate change (Averill et al., 2018; Jo et al., 2019). These potential drivers have been similar across forest stands at our study site for at least 90 years, and yet, we find dramatic differences in the abundance of AM trees following different land use disturbances.

In our study, black locust only appeared to facilitate the recovery of ECM forests at low levels of selective-cutting. For example, symbiotic N fixation was positively associated with oak and hickory growth when cutting was less than $18 \text{ m}^2 \text{ ha}^{-1}$, and was positively associated with all ECM spp. when cutting was less than $12 \text{ m}^2 \text{ ha}^{-1}$ (Figure 3a, light violet prediction). Beyond these thresholds of selective-cutting, and following clear-cutting, N fixation was associated with the positive growth of AM trees and not ECM trees (Figure 3a medium and dark violet predictions). Thus, under higher levels of forest cutting, black locust appears to facilitate a transition in mycorrhizal dominance. Yet, after agricultural abandonment, AM trees became dominant regardless of symbiotic N fixation, resulting in a complete mycorrhizal transition (Figures 1 and 3). Nitrogen fertilization of the agroecosystem (total inputs of 273 kg N ha^{-1}) prior to its abandonment and the lack of stumps for regeneration through sprouting may have contributed to this rapid transition in AM dominance.

The strong negative relationship between ECM tree growth and symbiotic N fixation at high levels of selective-cutting (Figure 3a) suggests that ECM trees were increasingly outcompeted by AM trees under higher availability of soil N. Such a response is consistent with theory that the ECM strategy to mine nutrients comes at a high carbon cost, making it unfavourable when competing with AM trees when soil-available N is abundant (Lu & Hedin, 2019). The decline in ECM tree biomass with N fixation in our study was not attributable to the losses of American chestnut or eastern hemlock, which were both functionally extirpated from our study site over the last century (these species were excluded from our analysis) nor did it depend on topographic position. Importantly, under low levels of selective-cutting, ECM tree growth was positively associated with symbiotic N fixation (Figure 3a), suggesting that fixation may facilitate recovery when disturbances mimic the prehistoric disturbance regime. Low levels of cutting create small canopy openings with low to moderate levels of light, similar to the effect of windthrown

trees (Clinton & Baker, 2000) or fire, which were set regularly by Indigenous People (H. R. Delcourt & Delcourt, 1997; P. A. Delcourt & Delcourt, 1998). Frequent fire would have also removed N and prevented the recruitment of AM-associating, fire-intolerant trees (Carpenter et al., 2020; Nowacki & Abrams, 2008), thereby maintaining the forest in an oligotrophic state. This suggests that symbiotic N fixation may have been a natural recovery mechanism for forests dominated by oak, and the ecologically similar American chestnut prior to high-intensity land use.

The biophysical context in which ECM and AM strategies compete over succession has changed because land-use activities, the dominant form of disturbance in eastern forests (Williams & Jackson, 2007), are more intense and frequent than the disturbance regimes under which these ecosystems evolved (P. A. Delcourt & Delcourt, 1998). Disturbance intensity appeared to have a stronger positive effect on the growth of AM trees than did symbiotic N fixation following selective-cutting (Figure 3a), and AM trees grew rapidly following agricultural abandonment regardless of fixation (Figure 3c). This raises the possibility that the AM strategy of scavenging nutrients is most favourable in high-light conditions, particularly for red maple and tulip poplar (Figure 3d–f). Indeed, prior work demonstrates that these two species grow more rapidly in high light than do co-occurring species (Kobe et al., 1995; Kolb et al., 1990), and their growth is further enhanced by the local abundance of black locust in early succession (Minucci et al., 2019). Red maple and tulip poplar produce small, wind-dispersed seeds in comparison to the larger, mammal-dispersed and mast-produced seeds of many ECM trees (Clark et al., 1999). This difference in dispersal may be particularly important following agricultural abandonment, where stump sprouts did not contribute to tree regeneration. These findings suggest that functional traits other than nutrient acquisition strategies, including growth rates in high light and seed dispersal, also contribute to the rise of AM dominance following land use.

Besides land use, other human activities may be weakening the resilience of ECM forests, which depend on, and promote, nutrient-poor conditions (Lu & Hedin, 2019). Atmospheric deposition of N has been elevated for several decades at Coweeta (Figure 2), suggesting the redundancy of fixation and atmospheric deposition in balancing N losses from land use. Further, long-term fire exclusion has prevented a natural loss pathway of N in this system, while inputs from fixation and deposition continue, which may further accumulate N in the ecosystem (Carpenter et al., 2020). Nitrogen-rich conditions may reduce the ecological need for symbiotic N fixation at both tree and ecosystem levels. Our prior work suggests that the nodule biomass of individual black locust trees has declined by ~80% since 1981, independent of forest succession (Wurzburger et al., 2022), which may indicate a down-regulation of N fixation in response to increased soil N availability (Knoepp et al., 2018). Further, the loss of the American chestnut has likely contributed to forest change because chestnut controlled forest nutrient cycles for millennia (Ellison et al., 2005). In support of this, plots with the largest historical abundance of American chestnut had the greatest growth of AM trees (Table S1).

Our work offers a unique case study of how human land use can trigger a transition in mycorrhizal dominance in a temperate forest. Although limited to one large forested basin, our analysis represents over 50,000 tree observations over 76 years and 42 tree species (Table S11), including tree species associated with both northern and southern forests of the eastern US. Further, our long-term forest experiment allows us to observe changes in forest dominance over time due to historical disturbances that have been common across the East—logging at the turn of the 20th century, the losses of American chestnut and eastern hemlock, agricultural abandonment and modern high-intensity timber harvesting. Although spatially extensive data from the Forest Inventory and Analysis (FIA) program of the USDA Forest Service show correlations between AM tree growth with the loss of fire and increasing N deposition, their short-term nature (surveys began in 1970, repeated plot surveys began in 1986) prevent analysis of forest transitions due to these widespread historical disturbances. An analysis of FIA data did not find a relationship between N fixer abundance and AM tree growth (Staccone et al., 2020), perhaps due to the dependency of N fixer effects on historical land use intensity (Figure 3), a metric not provided in FIA data. Nevertheless, it remains critical to determine if and how our findings translate to the broader expanse of eastern forests, and to other temperate and tropical forests that are bistable in mycorrhizal dominance (Lu & Hedin, 2019; Steidinger et al., 2019). Furthermore, determining whether the decadal timescales of our study are sufficient for understanding the mechanisms that underpin forest recovery (Moreno-Mateos et al., 2020) is another critical need.

Our ability to predict mycorrhizal transitions may determine whether we can sustain the ecosystem services provided by forests. Decades of research at our site has shown that a transition from ECM to AM dominance has reduced streamflow and thus freshwater drinking supplies (Caldwell et al., 2016), increased forest water use (Elliott et al., 2017), increased drought-induced declines in primary productivity (Elliott et al., 2015; Hwang et al., 2014, 2020) and reduced biodiversity (Elliott et al., 2020). Our findings suggest that the maintenance of these services depends in part on the interaction of land use with black locust. Stumps of black locust and other undesirable species are treated with herbicide following timber harvest. However, this method is not likely to prevent the proliferation of black locust because it readily recruits from persistent seed bank (Cofer et al., 2018) and sprouts from root stock (Shure et al., 2006) following disturbance. If the presence of black locust is inevitable following disturbance, management actions can minimize its negative effect on ECM forest recovery. For example, in the southern Appalachians, the majority of merchantable tree volume resides in the older, oak-hickory-dominated forests similar to our selectively cut stands. In these stands, our data suggest that no more than 50%–87% of desirable oak trees could be cut, such that black locust would facilitate a return to ECM dominance. With greater cutting levels than this, even more intensive efforts (e.g. direct planting of ECM trees, removal of AM trees, regular prescribed fire) would be necessary to prevent black locust from facilitating a transition to AM dominance and the degradation of ecosystem services. However,

the fact that all forests, even those experiencing mild land use, have increased in AM abundance over the last century (Figure 1), suggests that the long-term success of such strategies might be limited unless combined with other approaches to drastically reduce N in the ecosystem. Regular prescribed fire may be the most practical method for removing N from forests (Tierney, 2017), as it could further promote ECM dominance by reducing the recruitment of fire-intolerant, AM tree species (Nowacki & Abrams, 2008).

Our findings demonstrate how human activities can trigger a transition in forest mycorrhizal dominance, which underscores the vulnerability of eastern forests, and the services they provide to society, to future change. Our findings also offer evidence of 'N founder effects'—N fixers appear to affect trajectories of forest recovery, even decades after their mortality. The ubiquity of N fixers in early successional forests of the world (Sheffer et al., 2015) suggests that N fixers should be considered in a resilience framework that includes their role in ecosystem transitions following land use.

AUTHOR CONTRIBUTIONS

Nina Wurzbürger conceived of the study. Katherine J. Elliott collected and provided data. Nina Wurzbürger and Chelcy Ford Miniati analysed data. Nina Wurzbürger wrote the manuscript with contributions from all authors. Statement on inclusion: This study was conducted in partnership with scientists from the USDA Forest Service, which implemented the land use experiments and oversaw collection of all the long-term data analysed in the study. All authors were engaged with the data analysis and interpretation.

ACKNOWLEDGEMENTS

This research was supported by the U.S. Forest Service Southern Research Station, and the National Science Foundation (NSF) awards DEB-1440485 and DEB-1637522 from the Long-Term Ecological Research (LTER) Program to the Coweeta LTER. Any opinions, findings, conclusions or recommendations expressed in the material are those of the authors and do not necessarily reflect the views of the USDA or NSF.

CONFLICT OF INTEREST STATEMENT

No conflicts to declare.

DATA AVAILABILITY STATEMENT

Data available from USDA Forest Service Research Data Archive <https://doi.org/10.2737/RDS-2023-0013> (Wurzbürger et al., 2023).

ORCID

Nina Wurzbürger  <https://orcid.org/0000-0002-6143-0317>

Chelcy Ford Miniati  <https://orcid.org/0000-0002-3266-9783>

REFERENCES

- Averill, C., Dietze, M. C., & Bhatnagar, J. M. (2018). Continental-scale nitrogen pollution is shifting forest mycorrhizal associations and soil carbon stocks. *Global Change Biology*, 24(10), 4544–4553. <https://doi.org/10.1111/gcb.14368>

- Averill, C., Fortunel, C., Maynard, D. S., van den Hoogen, J., Dietze, M. C., Bhatnagar, J. M., & Crowther, T. W. (2022). Alternative stable states of the forest mycobiome are maintained through positive feedbacks. *Nature Ecology & Evolution*, 6(4), 375–382. <https://doi.org/10.1038/s41559-022-01663-9>
- Bates, D., Mächler, M., Bolker, B., & Walker, S. (2015). Fitting linear mixed-effects models using lme4. *Journal of Statistical Software*, 67(1). <https://doi.org/10.18637/jss.v067.i01>
- Boring, L. R., Elliott, K. J., & Swank, W. T. (2014). Successional forest dynamics 30 years following clearcutting. In *Long-term response of a forest watershed ecosystem: Clearcutting in the southern Appalachians* (pp. 11–35). Oxford University Press.
- Boring, L. R., & Swank, W. T. (1984). The role of black locust (*Robinia pseudo-acacia*) in forest succession. *Journal of Ecology*, 72(3), 749–766. <https://doi.org/10.2307/2259529>
- Caldwell, P. V., Miniati, C. F., Elliott, K. J., Swank, W. T., Brantley, S. T., & Laseter, S. H. (2016). Declining water yield from forested mountain watersheds in response to climate change and forest mesophication. *Global Change Biology*, 22(9), 2997–3012. <https://doi.org/10.1111/gcb.13309>
- Carpenter, D. O., Taylor, M. K., Callahan, M. A., Hiers, J. K., Loudermilk, E. L., O'Brien, J. J., & Wurzbürger, N. (2020). Benefit or liability? The ectomycorrhizal association may undermine tree adaptations to fire after long-term fire exclusion. *Ecosystems*, 24, 1059–1074. <https://doi.org/10.1007/s10021-020-00568-7>
- Clark, J. S., Silman, M., Kern, R., Macklin, E., & HilleRisLambers, J. (1999). Seed dispersal near and far: Patterns across temperate and tropical forests. *Ecology*, 80(5), 1475–1494. [https://doi.org/10.1890/0012-9658\(1999\)080\[1475:SDNAFP\]2.0.CO;2](https://doi.org/10.1890/0012-9658(1999)080[1475:SDNAFP]2.0.CO;2)
- Clinton, B. D., & Baker, C. R. (2000). Catastrophic windthrow in the southern Appalachians: Characteristics of pits and mounds and initial vegetation responses. *Forest Ecology and Management*, 126(1), 51–60. [https://doi.org/10.1016/S0378-1127\(99\)00082-1](https://doi.org/10.1016/S0378-1127(99)00082-1)
- Cofer, T. M., Elliott, K. J., Bush, J. K., & Miniati, C. F. (2018). *Rhododendron maximum* impacts seed bank composition and richness following *Tsuga canadensis* loss in riparian forests. *Ecosphere*, 9(4). <https://doi.org/10.1002/ecs2.2204>
- Curtis, P. G., Slay, C. M., Harris, N. L., Tyukavina, A., & Hansen, M. C. (2018). Classifying drivers of global forest loss. *Science*, 361(6407), 1108–1111. <https://doi.org/10.1126/science.aau3445>
- Delcourt, H. R., & Delcourt, P. A. (1997). Pre-Columbian native American use of fire on southern Appalachian landscapes. *Conservation Biology*, 11(4), 1010–1014.
- Delcourt, P. A., & Delcourt, H. R. (1998). The influence of prehistoric human-set fires on oak-chestnut forests in the southern Appalachians. *Castanea*, 63(3), 10.
- Elliott, K. J., Boring, L. R., & Swank, W. T. (1998). Changes in vegetation structure and diversity after grass-to-forest succession in a southern Appalachian watershed. *The American Midland Naturalist*, 140(2), 219–232.
- Elliott, K. J., Boring, L. R., Swank, W. T., & Haines, B. R. (1997). Successional changes in plant species diversity and composition after clearcutting a southern Appalachian watershed. *Forest Ecology and Management*, 92(1–3), 67–85. [https://doi.org/10.1016/S0378-1127\(96\)03947-3](https://doi.org/10.1016/S0378-1127(96)03947-3)
- Elliott, K. J., Caldwell, P. V., Brantley, S. T., Miniati, C. F., Vose, J. M., & Swank, W. T. (2017). Water yield following forest-grass-forest transitions. *Hydrology and Earth System Sciences*, 21(2), 981–997. <https://doi.org/10.5194/hess-21-981-2017>
- Elliott, K. J., Miniati, C. F., & Medenblik, A. S. (2020). The long-term case for partial-cutting over clear-cutting in the southern Appalachians USA. *New Forests*, 51(2), 273–295. <https://doi.org/10.1007/s11056-019-09731-y>
- Elliott, K. J., Miniati, C. F., Pederson, N., & Laseter, S. H. (2015). Forest tree growth response to hydroclimate variability in the southern Appalachians. *Global Change Biology*, 21(12), 4627–4641. <https://doi.org/10.1111/gcb.13045>
- Elliott, K. J., & Swank, W. T. (2008). Long-term changes in forest composition and diversity following early logging (1919–1923) and the decline of American chestnut (*Castanea dentata*). *Plant Ecology*, 197(2), 155–172. <https://doi.org/10.1007/s11258-007-9352-3>
- Elliott, K. J., Vose, J. M., Swank, W. T., & Bolstad, P. V. (1999). Long-term patterns in vegetation-site relationships in a southern Appalachian forest. *Journal of the Torrey Botanical Society*, 126(4), 320. <https://doi.org/10.2307/2997316>
- Ellison, A. M., Bank, M. S., Clinton, B. D., Colburn, E. A., Elliott, K., Ford, C. R., Foster, D. R., Kloeppel, B. D., Knoepp, J. D., Lovett, G. M., Mohan, J., Orwig, D. A., Rodenhouse, N. L., Sobczak, W. V., Stinson, K. A., Stone, J. K., Swan, C. M., Thompson, J., Von Holle, B., & Webster, J. R. (2005). Loss of foundation species: Consequences for the structure and dynamics of forested ecosystems. *Frontiers in Ecology and the Environment*, 3(9), 479–486. [https://doi.org/10.1890/1540-9295\(2005\)003\[0479:LOFSCF\]2.0.CO;2](https://doi.org/10.1890/1540-9295(2005)003[0479:LOFSCF]2.0.CO;2)
- Hanberry, B. B., & Nowacki, G. J. (2016). Oaks were the historical foundation genus of the east-central United States. *Quaternary Science Reviews*, 145, 94–103. <https://doi.org/10.1016/j.quascirev.2016.05.037>
- Hibbert, A. R. (1969). Water yield changes after converting a forested catchment to grass. *Water Resources Research*, 5(3), 634–640. <https://doi.org/10.1029/WR005i003p00634>
- Houlton, B. Z., Wang, Y.-P., Vitousek, P. M., & Field, C. B. (2008). A unifying framework for dinitrogen fixation in the terrestrial biosphere. *Nature*, 454(7202), 327–330. <https://doi.org/10.1038/nature07028>
- Hwang, T., Band, L. E., Miniati, C. F., Song, C., Bolstad, P. V., Vose, J. M., & Love, J. P. (2014). Divergent phenological response to hydroclimate variability in forested mountain watersheds. *Global Change Biology*, 20(8), 2580–2595. <https://doi.org/10.1111/gcb.12556>
- Hwang, T., Band, L. E., Miniati, C. F., Vose, J. M., Knoepp, J. D., Song, C., & Bolstad, P. V. (2020). Climate change may increase the drought stress of mesophytic trees downslope with ongoing forest mesophication under a history of fire suppression. *Frontiers in Forests and Global Change*, 3, 17. <https://doi.org/10.3389/ffgc.2020.00017>
- Jo, I., Fei, S., Oswald, C. M., Domke, G. M., & Phillips, R. P. (2019). Shifts in dominant tree mycorrhizal associations in response to anthropogenic impacts. *Science Advances*, 5(4), eaav6358. <https://doi.org/10.1126/sciadv.aav6358>
- Knoepp, J. D., See, C. R., Vose, J. M., Miniati, C. F., & Clark, J. S. (2018). Total C and N pools and fluxes vary with time, soil temperature, and moisture along an elevation, precipitation, and vegetation gradient in southern Appalachian forests. *Ecosystems*, 21(8), 1623–1638. <https://doi.org/10.1007/s10021-018-0244-2>
- Kobe, R. K., Pacala, S. W., Silander, J. A., & Canham, C. D. (1995). Juvenile tree survivorship as a component of shade tolerance. *Ecological Applications*, 5(2), 517–532. <https://doi.org/10.2307/1942040>
- Kolb, T. E., Steiner, K. C., McCormick, L. H., & Bowersox, T. W. (1990). Growth response of northern red-oak and yellow-poplar seedlings to light, soil moisture and nutrients in relation to ecological strategy. *Forest Ecology and Management*, 38(1–2), 65–78. [https://doi.org/10.1016/0378-1127\(90\)90086-Q](https://doi.org/10.1016/0378-1127(90)90086-Q)
- Lu, M., & Hedin, L. O. (2019). Global plant-symbiont organization and emergence of biogeochemical cycles resolved by evolution-based trait modelling. *Nature Ecology & Evolution*, 3(2), 239–250. <https://doi.org/10.1038/s41559-018-0759-0>
- Martin, J. G., Kloeppel, B. D., Schaefer, T. L., Kimbler, D. L., & McNulty, S. G. (1998). Aboveground biomass and nitrogen allocation of ten deciduous southern Appalachian tree species. *Canadian Journal of Forest Research*, 28(11), 1648–1659. <https://doi.org/10.1139/x98-146>
- Minucci, J. M., Miniati, C. F., & Wurzbürger, N. (2019). Drought sensitivity of an N₂-fixing tree may slow temperate deciduous forest recovery from disturbance. *Ecology*, 100(12), e02862. <https://doi.org/10.1002/ecy.2862>

- Moreno-Mateos, D., Alberdi, A., Morriën, E., van der Putten, W. H., Rodríguez-Uña, A., & Montoya, D. (2020). The long-term restoration of ecosystem complexity. *Nature Ecology & Evolution*, 4(5), 676–685. <https://doi.org/10.1038/s41559-020-1154-1>
- National Atmospheric Deposition Program. (2019). *National Atmospheric Deposition Program (NRSP-3)*. NADP Program Office Wisconsin State Laboratory of Hygiene.
- Nowacki, G. J., & Abrams, M. D. (2008). The demise of fire and “Mesophication” of forests in the eastern United States. *BioScience*, 58(2), 123–138. <https://doi.org/10.1641/B580207>
- Pan, Y., Chen, J. M., Birdsey, R., McCullough, K., He, L., & Deng, F. (2011). Age structure and disturbance legacy of North American forests. *Biogeosciences*, 8(3), 715–732. <https://doi.org/10.5194/bg-8-715-2011>
- Perring, M. P., De Frenne, P., Baeten, L., Maes, S. L., Depauw, L., Blondeel, H., Carón, M. M., & Verheyen, K. (2016). Global environmental change effects on ecosystems: The importance of land-use legacies. *Global Change Biology*, 22(4), 1361–1371. <https://doi.org/10.1111/gcb.13146>
- Phillips, R. P., Brzostek, E., & Midgley, M. G. (2013). The mycorrhizal-associated nutrient economy: A new framework for predicting carbon-nutrient couplings in temperate forests. *New Phytologist*, 199(1), 41–51. <https://doi.org/10.1111/nph.12221>
- R Core Team. (2020). *R: A language and environment for statistical computing*. R Foundation for Statistical Computing. <https://www.R-project.org/>
- Seidl, R., Spies, T. A., Peterson, D. L., Stephens, S. L., & Hicke, J. A. (2016). REVIEW: Searching for resilience: Addressing the impacts of changing disturbance regimes on forest ecosystem services. *Journal of Applied Ecology*, 53(1), 120–129. <https://doi.org/10.1111/1365-2664.12511>
- Sheffer, E., Batterman, S. A., Levin, S. A., & Hedin, L. O. (2015). Biome-scale nitrogen fixation strategies selected by climatic constraints on nitrogen cycle. *Nature Plants*, 1(12), 15182. <https://doi.org/10.1038/nplants.2015.182>
- Shure, D. J., Phillips, D. L., & Edward Bostick, P. (2006). Gap size and succession in cutover southern Appalachian forests: An 18 year study of vegetation dynamics. *Plant Ecology*, 185(2), 299–318. <https://doi.org/10.1007/s11258-006-9105-8>
- Staccone, A., Kou-Giesbrecht, S., Taylor, B. N., & Menge, D. N. L. (2020). Nitrogen-fixing trees have no net effect on forest growth in the coterminous United States. *Journal of Ecology*, 109, 877–887. <https://doi.org/10.1111/1365-2745.13513>
- Steidinger, B. S., Crowther, T. W., Liang, J., Van Nuland, M. E., Werner, G. D. A., Reich, P. B., Nabuurs, G. J., de-Miguel, S., Zhou, M., Picard, N., Hérault, B., Zhao, X., Zhang, C., Routh, D., & Peay, K. G. (2019). Climatic controls of decomposition drive the global biogeography of forest-tree symbioses. *Nature*, 569(7756), 404–408. <https://doi.org/10.1038/s41586-019-1128-0>
- Swank, W. T., & Crossley, D. A. (1988). *Forest hydrology and ecology at Coweeta*. Springer New York. <https://doi.org/10.1007/978-1-4612-3732-7>
- Swank, W. T., & Webster, J. R. (Eds.). (2014). *Long-term response of a forest watershed ecosystem: Clearcutting in the southern Appalachians*. Oxford University Press.
- Swift, L. W., Jr., Cunningham, G. B., & Douglass, J. E. (1988). Climatology and hydrology. In W. T. Swank & D. A. Crossley, Jr. (Eds.), *Forest hydrology and ecology at Coweeta* (pp. 35–56). Springer-Verlag.
- Thomas, D. J. (1996). *Soil survey of Macon County, North Carolina*. USDA Natural Resources Conservation Service.
- Tierney, J. A. (2017). *Biological nitrogen fixation dynamics during ecosystem recovery in longleaf pine savannas* (M.S. thesis). University of Georgia.
- Von Holle, B., Neill, C., Largay, E. F., Budreski, K. A., Ozimec, B., Clark, S. A., & Lee, K. (2013). Ecosystem legacy of the introduced N₂-fixing tree *Robinia pseudoacacia* in a coastal forest. *Oecologia*, 172(3), 915–924. <https://doi.org/10.1007/s00442-012-2543-1>
- Williams, J. W., & Jackson, S. T. (2007). Novel climates, no-analog communities, and ecological surprises. *Frontiers in Ecology and the Environment*, 5(9), 475–482. <https://doi.org/10.1890/070037>
- Wood, S. N. (2011). Fast stable restricted maximum likelihood and marginal likelihood estimation of semiparametric generalized linear models: Estimation of semiparametric generalized linear models. *Journal of the Royal Statistical Society: Series B (Statistical Methodology)*, 73(1), 3–36. <https://doi.org/10.1111/j.1467-9868.2010.00749.x>
- Wurzburger, N., Elliott, K. J., & Miniat, C. F. (2023). *Long-term changes in forest biomass, tree species composition and nitrogen fixation following land use disturbance*. Forest Service Research Data Archive. <https://doi.org/10.2737/RDS-2023-0013>
- Wurzburger, N., Motes, J. I., & Miniat, C. F. (2022). A framework for scaling symbiotic nitrogen fixation using the most widespread nitrogen fixer in eastern deciduous forests of the United States. *Journal of Ecology*, 110(3), 569–581. <https://doi.org/10.1111/1365-2745.13819>

SUPPORTING INFORMATION

Additional supporting information can be found online in the Supporting Information section at the end of this article.

Appendix S1: Methods for estimating symbiotic N fixation

Figure S1: Stem density of black locust *Robinia pseudoacacia* for (a) selective-cut, (b) clear-cut and (c) agricultural abandonment disturbances over time since disturbance. Points are measures of stem density in each permanent plot at each survey and curves display fitted exponential functions to stem density data, back cast to year of disturbance. The best fit model (lowest AIC_c value) associated with the clear-cut disturbance, which is the most temporally resolved, was applied to the other disturbances.

Figure S2: Net above-ground biomass change of non-fixing trees with increasing symbiotic N fixation by black locust in the same plot. Forest biomass (a) increased with increasing N fixation under low to moderate levels of selective cutting, but decreased under high levels of selective-cutting; and displayed no relationship with fixation following the (b) clear-cut and (c) agricultural abandonment disturbances. Each point represents net biomass change in a long-term vegetation plot over time since disturbance. To visualize the interaction between fixation and the intensity of selective-cutting in panel a, lines represent predictions at the minimum and maximum levels of basal area extraction and the value (20.5 m² ha⁻¹) where the slope is closest to 0, holding other variables at their median. One outlier (453 Mg ha⁻¹) was excluded from panel (a).

Table S1: Results of a linear mixed-effect model of net biomass change from 1921 to 2010 in the selectively cut disturbance. Fixed effects include mycorrhizal guild, cumulative N fixation (since time of disturbance), basal area (BA) of American chestnut in 1934, terrain shape and basal area of extracted trees, random effect of plot. See [Table S11](#) for a list of the 23 species included in analysis. *p* values in bold denote significance at $\alpha=0.05$ or less.

Table S2: Results of linear mixed-effect model of net biomass change in the clear-cut disturbance. Fixed effects include mycorrhizal guild, cumulative N fixation (since time of disturbance) and their interaction. See [Table S11](#) for 18 species included in analysis. *p* values in bold denote significance at $\alpha=0.05$ or less.

Table S3: Results of linear mixed-effect model of net biomass change in the agricultural abandonment disturbance. Fixed effects include mycorrhizal guild, cumulative N fixation (since time of disturbance)

and their interaction. *p* values in bold denote significance at $\alpha=0.05$ or less. See Table S11 for 11 species included in analysis.

Table S4: Results of a linear mixed-effect model of net biomass change from 1921 to 2010 in the selectively-cut disturbance. Fixed effects include mycorrhizal guild, maximum observed black locust above-ground biomass (since time of disturbance), basal area (BA) of American chestnut in 1934 and basal area of extracted trees, random effect of plot. See Table S11 for a list of the 23 species included in analysis. *p* values in bold denote significance at $\alpha=0.05$ or less.

Table S5: Results of linear mixed-effect model of net biomass change in the clear-cut disturbance. Fixed effects include mycorrhizal guild, maximum observed black locust above-ground biomass (since time of disturbance) and their interaction. See Table S11 for 18 species included in analysis. *p* values in bold denote significance at $\alpha=0.05$ or less.

Table S6: Results of linear mixed-effect model of net biomass change in the agricultural abandonment disturbance. Fixed effects include mycorrhizal guild, maximum observed black locust above-ground biomass (since time of disturbance) and their interaction. *p* values in bold denote significance at $\alpha=0.05$ or less. See Table S11 for 11 species included in analysis.

Table S7: Results of a linear mixed-effect model of net biomass change from 1921 to 2010 in the selectively-cut disturbance. Fixed effects include tree group (maple and tulip poplar versus oak and hickory), cumulative N fixation (since time of disturbance), basal area (BA) of American chestnut in 1934 and basal area of extracted trees, random effect of plot. *p* values in bold denote significance at $\alpha=0.05$ or less.

Table S8: Results of linear regression of combined net biomass change of tree group (maple and tulip poplar versus oak and hickory) in the clear-cut disturbance as a function of cumulative N fixation (since time of disturbance). *p* values in bold denote significance at $\alpha=0.05$ or less.

Table S9: Results of linear regression of combined net biomass change of tree group (maple and tulip poplar versus oak and hickory) in the agricultural abandonment disturbance as a function of cumulative N fixation (since time of disturbance). *p* values in bold

denote significance at $\alpha=0.05$ or less.

Table S10: Results of linear mixed-effect model of net forest biomass change from 1921 to 2010 in the selective-cut disturbance as a function of cumulative N fixation (since time of disturbance), basal area (BA) of American chestnut in 1934, and basal area (BA) of extracted trees. *P* values in bold denote significance at $\alpha=0.05$ or less.

Table S11: Tree species observed and their frequency (percent of total plot observations) for each disturbance and their mycorrhizal association type. For our analysis of forest biomass growth by mycorrhizal type, only species observed in more than 10% of plots across time were included (denoted by bold value). Trees were classified as either arbuscular mycorrhizal (AM) or ectomycorrhizal (ECM) based on prior assessments of these genera or species (Brundrett & Tedersoo, 2019). We classified sourwood *Oxydendrum arboreum* as ECM for our analysis. While sourwood can associate with ericoid mycorrhizal fungi (ERM) (Gorman & Starrett, 2003), so can other ECM hosts (Grelet et al., 2010), and our visual assessment of its roots indicated a loose fungal mantle similar to ECM roots. Further, ERM fungi are more functionally similar to ECM than AM fungi (Lambers et al., 2008). All species were included in analysis of net biomass change, with the exception of American chestnut and eastern hemlock (which were functionally extirpated due to an invasive pathogen and pest, respectively) and black locust (which was indirectly included in our model via estimates of symbiotic N fixation). We note that hazel alder *Alnus serrulata* is an N fixer, but it only occurs in riparian areas, and is therefore likely to have a small contribution to symbiotic N fixation in these forests.

How to cite this article: Wurzbürger, N., Elliott, K. J., & Miniat, C. F. (2023). Forest mycorrhizal dominance depends on historical land use and nitrogen-fixing trees. *Journal of Applied Ecology*, 60, 1551–1561. <https://doi.org/10.1111/1365-2664.14443>