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# Determinants of Deadwood Biomass under the Background of Nitrogen and Water Addition in Warm Temperate Forests

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Abstract: Climate change is exacerbating the vulnerability of temperate forests to severe disturbances, potentially increasing tree mortality rates. Despite the significance of this issue, there has been a lack of comprehensive research on tree survival across extensive forest areas under the background of global climate change. To fill this gap, we conducted a detailed analysis of tree survival within a canopy nitrogen and water addition experimental platform in central China, utilizing data from two censuses and evaluating contributing factors. Our findings revealed 283 dead trees within the plots, predominantly of very small diameters (1–10 cm). The distribution of these dead trees varied among subplots, influenced by both biotic and abiotic factors. Notably, three dominant tree species were responsible for 64.8% of the deadwood biomass. The study determined that both the breast diameter and the quantity of dead trees, affected by surrounding trees and environmental conditions, played a critical role in deadwood biomass accumulation. This research offers an in-depth examination of deadwood biomass patterns in a temperate forest, highlighting the need to consider both experiment treatments and abiotic elements like topography in studies of forest ecosystem carbon. The insights gained from this study enhance our understanding of warm temperate forests' role in the global carbon cycle and offer valuable guidance for forest conservation and management strategies.

**Keywords:** canopy nitrogen addition; canopy water addition; abiotic and biotic factors; temperate forest; tree survival



1. Introduction

Forests are serving as the lungs of the Earth by absorbing carbon dioxide and releasing oxygen, thus playing a crucial role in mitigating climate change [1,2]. They are biodiversity hotspots. Forests also provide essential ecosystem services, including water regulation, soil protection, and nutrient cycling, which are critical for agriculture and human survival [3]. Despite their importance, forests worldwide are under significant threat due to global climatic changes [4,5]. This not only contributes to biodiversity loss but also increases greenhouse gas emissions, exacerbating climate change. Forests are also threatened by invasive species, which can outcompete native flora and fauna, and the increasing frequency and severity of wildfires, often exacerbated by climate change and poor management practices [6–8]. Additionally, changing precipitation patterns and temperatures are affecting forest health and productivity, making them more susceptible to pests and diseases.



Citation: Hong, L.; Tang, S.; Li, T.; Fu, L.; Song, X.; Duan, G.; Fu, J.; Ma, L. Determinants of Deadwood Biomass under the Background of Nitrogen and Water Addition in Warm
Temperate Forests. *Forests* **2024**, *15*, 1464. https://doi.org/10.3390/f15081464

Academic Editors: Andrzej Węgiel and Adrian Łukowski

Received: 20 May 2024 Revised: 20 July 2024 Accepted: 14 August 2024 Published: 20 August 2024



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The increase in atmospheric nitrogen deposition is a pressing environmental issue that has garnered significant attention in recent years. This phenomenon is primarily driven by human activities, such as the burning of fossil fuels, industrial processes, and agricultural practices, which release large amounts of reactive nitrogen into the atmosphere. The current situation regarding increased rainfall patterns is becoming another significant concern across the globe, as it is closely linked to the broader impacts of climate change [9]. This phenomenon is characterized by more intense and frequent precipitation events in various regions, disrupting traditional weather patterns and posing challenges to both natural ecosystems and human societies [10].

The consequences of increased atmospheric nitrogen deposition are manifold and impact various ecosystems in profound ways [11,12]. Nitrogen deposition can initially stimulate forest growth because nitrogen is a critical nutrient for plants. However, excessive nitrogen can lead to a series of detrimental ecological effects. For instance, it can cause soil acidification, which in turn can harm root systems and reduce the availability of other essential nutrients [13]. Moreover, nitrogen saturation in forests can lead to imbalances in plant species composition by favoring fast-growing species over others, thereby reducing biodiversity. Additionally, increased nitrogen levels can affect the interactions between plants and mycorrhizal fungi, which are crucial for nutrient uptake in many trees [14]. It can also exacerbate the susceptibility of forests to pests, diseases, and the effects of climate change, such as drought stress. In terms of broader ecological impacts, excessive nitrogen deposition can lead to increased emissions of nitrous oxide, a potent greenhouse gas, from soils, thus contributing to global warming.

The increase in precipitation, a significant aspect of climate change, has profound implications for forest ecosystems worldwide. While forests require water to thrive, the alteration of precipitation patterns and the occurrence of more intense and frequent rainfall events can disrupt the delicate balance of these ecosystems [15]. Excessive rainfall can lead to soil saturation, reducing the oxygen available to tree roots and potentially causing root asphyxiation and reduced growth. Furthermore, heavy and consistent downpours increase the risk of soil erosion, stripping away the fertile top layer of soil and depleting the nutrients essential for forest health [16]. This erosion can also lead to the leaching of vital nutrients from the soil, further impoverishing the forest floor and affecting plant growth. Increased precipitation can also alter the hydrological dynamics within a forest, affecting both plant and animal species. For example, changes in water availability can impact the prevalence and distribution of certain tree species, leading to shifts in forest composition over time [17]. Additionally, higher moisture levels create favorable conditions for the proliferation of pests and diseases, which can further stress and damage trees, sometimes leading to widespread forest decline. Moreover, the increase in rainfall can affect the forest's capacity to sequester carbon. While more water can stimulate growth and carbon uptake in some instances, the negative impacts of excessive moisture and nutrient loss may counteract these benefits, potentially diminishing the forest's role as a carbon sink.

The survival of trees in forest ecosystems is influenced by a complex interplay of factors that can either bolster their growth and resilience or pose significant threats to their well-being [18]. Key among these factors are climatic conditions, such as temperature and precipitation patterns, which directly affect the availability of water and the suitability of habitats for different tree species [19]. Extreme weather events, including droughts, heavy rainfall, and storms, can cause immediate damage to trees and alter the environmental conditions necessary for their survival. Soil quality is another critical determinant of tree health and survival [20]. Nutrient availability, pH levels, and soil structure influence root development and the ability of trees to uptake water and essential minerals.

Current experiments on nitrogen addition in forest understories are pivotal for understanding the complex dynamics of forest ecosystems in response to increased atmospheric nitrogen deposition, a consequence of industrial and agricultural activities [21,22]. These experiments aim to simulate the effects of nitrogen enrichment on various aspects of forest ecology, including plant diversity, soil chemistry, and microbial communities. By doing

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so, researchers can gain insights into how forests might change in the coming decades and develop strategies to mitigate potential negative impacts [2,23]. One of the primary advantages of these experiments is their ability to provide direct, empirical evidence on the ecological consequences of nitrogen addition. They can help identify critical thresholds for nitrogen inputs beyond which forest health and biodiversity may be compromised [3,24]. Furthermore, such experiments can inform forest management practices, guiding efforts to maintain ecosystem balance and function in the face of changing environmental conditions. However, there are several limitations and drawbacks to these experiments. One crucial point is that previous nitrogen and water addition experiments have mostly focused on understorey additions, overlooking the role of the forest canopy.

In this study, we investigated the factors that affect the survival of trees in warm temperate forests under the interaction of nitrogen and water addition, aiming to provide a certain basis for forest management and protection. The development of this study was based on the forest canopy nitrogen and water addition experimental platform located in Jigongshan National Nature Reserve, Henan Province. Our hypothesis is as follows: (1) Does the addition of nitrogen and water affect the death of forest trees? (2) Are the effects of nitrogen and water addition on the biomass of dead trees in forests the same?

### 2. Materials and Methods

# 2.1. Study Site

The study site is located in the Jigongshan National Nature Reserve ( $31^{\circ}46'-31^{\circ}52'$  N,  $114^{\circ}01'-114^{\circ}06'$  E), Xinyang City, Henan Province, China. The study area has a warm temperate climate. The annual average rainfall is 1119 mm and temperature is 15.2 °C. The soil type is yellow-brown soil. The forest type is mixed deciduous forest, and the forest is 50 years old (thinned in 1970) [25]. The stand density of the community was 446 stems ha<sup>-1</sup>.

# 2.2. Experimental Design

We have established a complete dual-factor random block design that includes four blocks. The vegetation, terrain, soil, and other environmental factors within the four blocks are relatively consistent. Each block has been randomly assigned with four treatments (907 m² per plot), including control (CK), canopy N addition (addition N 25 kg·ha $^{-1}$ ·yr $^{-1}$ ), canopy water addition (addition of water equivalent to 30% of the local precipitation, 336 mm·yr $^{-1}$ ), and canopy N and water addition. A 20 m buffer zone is established between any two treatments, and a cement board with a depth of 50 cm is buried between the buffer zones to prevent mutual contamination between the treatments (Figure 1).

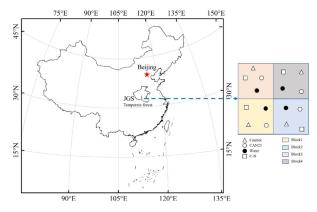


Figure 1. Location of the study site and experiment design.

In 2012, an iron tower with a height of 35 m (5–8 m above the canopy) was erected in the middle of each treatment except for CK, and the base of the tower was reinforced with cement to enhance stability. A 360-degree rotating nozzle is installed at the top of

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the tower, with a spraying range of 17 m, ensuring that N solution and water are evenly sprayed within the treatment area. The treatment process began in 2013 and started in April each year, with final treatment in October. The required solution for treatment is sourced from the collected local rainwater. In case of insufficient rainwater, water is taken from local lakes. Before canopy addition of N, it is necessary to test the solution (collected rainwater or local lake water) and then add high-purity NH<sub>4</sub>NO<sub>3</sub> to achieve the target N concentration. The canopy N addition treatment should be carried out on a sunny, windless evening or early morning in the middle of the month (7 times a year) to avoid uncertainties caused by evaporation and local rainstorms. Unlike the canopy addition of N, to prevent surface runoff caused by heavy rainfall, canopy water addition is distributed once a week (four times a month, a total of 28 times from April to October). Canopy addition of N and water is a combination of the two (Figure 2).



Figure 2. Facilities for nitrogen and water application in forest canopies.

## 2.3. Quadrat Establishment and Field Methods

Before the experimental treatment, a 20 m square vegetation survey plot was established under each treatment. In 2012, a baseline ecological survey was conducted on these 16 plots according to standards, including species composition and community structure. All individuals with a breast height diameter (DBH)  $\geq 1$  cm within the plots were surveyed, measuring and recording the species name, DBH, tree height, and crown width of each tree individual. Additionally, each was numbered, tagged, and the coordinates within the plot (relative position) were marked. In 2017, five years after the experimental treatment, the plant community was re-surveyed. During the re-survey of the trees, an effort was made to locate all individuals from the initial survey based on their numbers and coordinates, record the new survey data, and mark their status: alive (top-broken, fallen, etc.) or dead (standing dead, fallen, stump, disappeared, etc.). For new individuals (those with a DBH < 1 cm in the initial survey or not present but with a DBH  $\geq 1$  cm in the re-survey), the survey and recording were conducted according to the previous standards, and they were tagged and numbered. The numbering continued in sequence from before to avoid duplication.

### 2.4. Biotic and Abiotic Factors

To enhance the assessment of factors influencing tree mortality, the biotic and abiotic factors were estimated. The habitat variables for each plot, including soil characteristics and topographical features, were evaluated based on the criteria used in a prior investigation conducted in the same area. This included all topographical variables such as elevation, aspect, convexity, and slope, which were recorded for every subplot. For the analysis of soil properties, a total of 48 soil samples from the top 20 cm layer were collected. These samples were then analyzed for eight key soil properties: pH, organic matter, total nitrogen (N), available N, total phosphorus (P), available P, total potassium (K), and available K, following the methodologies outlined in previous research [26].

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#### 2.5. Deadwood Biomass Estimates

Before estimating the biomass of each deadwood individual, the decay grade coefficient of each was determined by visual assessment. If a tree had only recently died, the decay grade coefficient was set to 5/5. The decay grade coefficient was set to 0/5 if a tree was completely decomposed (and had disappeared in the second census). Then, the deadwood biomass was calculated by multiplying the decay grade coefficient and biomass of each tree.

The biomass of each tree was estimated using the allometric equation for mixed forests [27]:

$$B = a \times (DBH)^b$$

where a and b are statistical parameters (see Table 1 for equations and summary statistics). B is the total dry weight including trunks and branches and was obtained by multiplying the biomass of each tree by the decay grade coefficient. Finally, the deadwood biomass storage ( $DB_d$ ) was estimated by sum the biomass of each tree within each plot within our study site.

Table 1.	Allometric re	egression e	equations	and	summary	statistics.

DBH-Class	Equations	Adjusted R <sup>2</sup>	Standard Error of the Mean	R. Error (%)
$DBH \le 5 cm$	$WT = 0.05549 \times D^{2.87776}$	0.91164	0.60826	-0.23
	$WB = 0.01124 \times D^{3.16237}$	0.81933	0.30284	0.00
	$WL = 0.01551 \times D^{2.32693}$	0.86555	0.08602	0.42
	$WR = 0.02838 \times D^{2.65348}$	0.90495	0.22077	-0.27
$5 < DBH \le 10 \text{ cm}$	$WT = 0.11701 \times D^{2.36933}$	0.88428	2.05700	0.04
	$WB = 0.01621 \times D^{2.93859}$	0.76490	1.79321	0.63
	$WL = 0.04169 \times D^{1.90082}$	0.68922	0.44047	0.39
	$WR = 0.04977 \times D^{2.19517}$	0.95730	0.32819	-0.16
$10 < DBH \le 20 cm$	$WT = 0.10769 \times D^{2.34891}$	0.77761	4.15734	4.55
	$WB = 0.00385 \times D^{3.15093}$	0.88184	3.81171	3.69
	$WL = 0.00372 \times D^{2.65113}$	0.82848	0.96151	0.57
	$WR = 0.03538 \times D^{2.29567}$	0.81687	3.46518	0.45
DBH > 20 cm	$WT = 0.03541 \times D^{2.65146}$	0.97844	36.71034	-2.34
	$WB = 0.00583 \times D^{2.94383}$	0.85965	52.85291	-1.61
	$WL = 0.07709 \times D^{1.55399}$	0.71000	4.94167	-0.30
	$WR = 0.01128 \times D^{2.67850}$	0.92962	24.5010	-1.11

WT = weight of trunk, WB = weight of branch, WL = weight of leaf, WR = weight of root.

# 2.6. Statistical Analyses

The statistical significance of our findings was determined to be significant with a value of (p < 0.05). Given that the data deviated from a normal distribution, we applied a transformation method to standardize it. To identify differences among levels within factors, we employed the Tukey honest significant difference (Tukey HSD) test. To address multicollinearity among soil variables, principal component analysis (PCA) was utilized for the soil attributes, with the first two principal components representing concentration variables that captured 83.5% of the total variance in soil properties. For modeling the variations in deadwood biomass due to biotic and abiotic factors within each plot, we used structural equation modeling, which includes the number of dead trees, species richness, basal area, and soil physicochemical properties. These analyses were performed using R software version 4.2.1.

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## 3. Results

# 3.1. Species Composition and Community Characteristics of Trees

During 2012 and 2017, the total count of standing trees in the study area fell from 3314 to 3031, marking the death of 283 trees (which constituted 8.5% of the total tree population in 2012). Within each  $20 \times 20$  m plot, the number of deceased trees varied from 9 to 24, with an average mortality of  $17 \pm 3$  (standard error) trees per plot (Table 2). The variety of tree species observed declined from 38 to 35, with 18 species recorded among the dead trees within the study area. The diversity of dead tree species within plots varied, ranging from 4 to 15 species, with an average of  $1 \pm 0.2$  species per plot. Deadwood was found across a broad spectrum of DBH sizes (1–65 cm), indicating that large trees were also affected. Nonetheless, a significant majority, 80.6%, of the dead trees had a very small DBH, as detailed in Table 3 and illustrated in Figure 3. The results of one-way ANOVA showed that adding nitrogen or water would affect the biomass of dead trees, but it would not affect their mortality.

**Table 2.** Deadwood density, abundance, and biomass ( $DB_d$  in t) across DBH ranges within the 20 ha subtropical forest plot.

DBH Ranges	cm	Abundance	DB <sub>d</sub>
Very small	1–10 cm	201	8.2
Small	10-30 cm	50	24.9
Medium	30–50 cm	30	16.3
Large	>50 cm	2	13.4
	Total	283	62.8

**Table 3.** Biomass storage (DB<sub>d</sub> in t) by dead tree species of the top 10 individuals in a 20 ha subtropical forest plot.

	Species	$DB_d$
1	Quercus acutissima Carruth.	11.7
2	Liquidambar formosana Hance	9.9
3	Quercus variabilis Blume	7.7
4	Acer buergerianum Miq.	6.6
5	Celtis sinensis Pers.	6.3
6	Lindera glauca (Siebold & Zucc.) Blume	4.9
7	Prunus tomentosa Thunb.	4.5
8	Diospyros lotus L.	3.5
9	Vernicia fordii (Hemsl.) Airy Shaw	3.3
10	Quercus glauca Thunb.	1.9

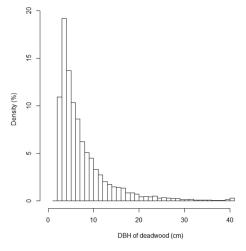


Figure 3. Distribution of breast diameter of dead trees in the study site.

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# 3.2. Drivers of Tree Mortality at Community Level

Nitrogen and water addition had negative effects on deadwood biomass storage indirectly via influencing community characteristics in the study area, because there are no significant differences between nitrogen addition treatment or water addition treatment with CK, respectively. Specifically, N addition indirectly influenced deadwood biomass via negative effects on abundance, soil properties, and basal area (Figure 4). However, water addition indirectly increased and decreased the deadwood biomass of this temperate forest via increasing deadwood richness and decreasing abundance, basal area, and soil properties. The impact pathways of nitrogen and water addition on the biomass of dead trees in warm temperate forests are different.

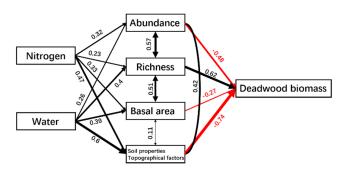


Figure 4. Path analysis of the effects of nitrogen and water addition on the deadwood biomass.

#### 4. Discussion

Despite the wealth of research on deadwood's contributions to the carbon cycle and its importance in the regeneration of trees within diverse forest ecosystems, the specific roles and effects of deadwood in the forests of southern China have not been thoroughly investigated [28]. Deadwood biomass is a vital component of forest ecosystems, with its influence shaped by factors like species composition, community structure, and the landscape's physical features [29,30]. Research has shown that the nature of the wood can greatly affect carbon storage, particularly when comparing forests within the same climatic regions. Although previous studies have explored deadwood's roles in carbon storage, biodiversity, and tree regeneration, there is an increasing demand for more detailed studies on how deadwood biomass is affected by climate change, diseases, insect infestations, and land-use changes, all of which are key drivers in the transformation of global forest ecosystems [31–33].

Tree mortality plays a crucial role in determining the structure, composition, and evolutionary processes of forest ecosystems [34]. It significantly impacts the composition of species, which in turn affects community characteristics, along with the accumulation of nutrients and biomass [35]. Consequently, comprehending tree mortality is vital for understanding the dynamics of forests. This research contributes to the existing knowledge by analyzing the variations in deadwood biomass over a span of five years, highlighting its consequences for the health and management of forests [36,37].

In this research, we categorized individual trees based on their diameter at breast height (DBH) into different groups. We discovered that the volume of deadwood plays a significant role in the total deadwood biomass. Notably, the mortality of larger trees seemed to have the most pronounced effect on deadwood biomass, although these observations did not achieve statistical significance [38–40]. This observation is consistent with prior studies which have shown that although large trees make up only 3%–5% of the total tree population, they account for over 65% of the total deadwood biomass. Our results support these previous findings, underscoring the critical role that deadwood from large trees has in the buildup of deadwood biomass [26]. Specifically, we found that the local variations in deadwood biomass within the  $20 \times 20$  m plots were mainly due to these large trees. In forests with low wood density, trees tend to grow larger, which leads to an increase in deadwood biomass. In our analysis, the trees with the lowest dry density made up a small

percentage (2.6% and 2.5%) of the total tree population, yet they contributed to 45%–49% of the biomass used for timber construction and a similar share of the total deadwood biomass [19,41]. The study indicates that the structure of the forest, especially the basal area of trees with the largest diameters and, to a lesser extent, the average wood density at the stand level, might explain the variations in deadwood biomass observed across the study sites [5,9,42]. We observed significant variability in biomass, with the variation in dry wood biomass across different locations being associated with density differences and specific site conditions.

Furthermore, the research pinpointed several abiotic factors that play a crucial role in the ecology of dead trees, including elevation, openness, and characteristics of the soil. Although forest environments typically experience minimal local fluctuations, a significant reduction in various attributes of dead trees was noted. The elevation at which trees were found ranged from 230 m to 470 m, showcasing a broad spectrum of diversity [29,33,37]. The terrain of the area under study was more complex than that of other locations such as Changbai Mountain, Pasoh, and Barro Colorado Island, where similar investigations have taken place. This complexity, coupled with the diversity of dead trees at lower elevations, resulted in notable differences in mortality rates [43]. The diversity among dead trees was further limited by the narrow species range and average tree diameter. Differences in altitude might shed light on these disparities; higher elevations typically face harsher conditions, including stronger winds and lower temperatures, which can markedly affect tree growth, structural stability, and the functioning of ecosystems [7,44]. Moreover, other abiotic elements such as the curvature of the slope, directionality, and soil nutrient content are likely to have significant impacts on the life cycle of dead trees.

Our research showed that the warm temperate forest under investigation was more dynamic than other tropical and subtropical forests that have been studied in the past. For example, a study carried out in a protected 25-hectare area in northeastern China reported similar dynamics but with significantly lower mortality rates [21,30]. Additionally, the density observed in a permanent 5-hectare forest plot in northern China was lower than the terrestrial mortality rates found in our study, a difference that might be explained by that forest being in an earlier stage of development. The pace of forest dynamics is affected by a variety of factors, including the landscape's topography, its geological features, climatic conditions, the biological and successional stages it is currently in, and the impact of human activities [5]. The forest we studied stands out due to its numerous steep inclines and a more intricate and diverse ecosystem, which contributes to more vigorous population dynamics.

The decrease in the biomass of standing trees due to the conversion into deadwood biomass throughout the five-year duration of our study falls within the expected range of variability for forest health, as supported by existing literature [11,38]. Furthermore, the carbon stored in the growing deadwood biomass could potentially be released as CO<sub>2</sub>. While there have been efforts to measure deadwood biomass and identify its determinants in China, more extensive and prolonged research is necessary to determine if these forests are experiencing a temporary state of balance. Such research is vital for pinpointing factors that bolster the health and resilience of this subtropical forest, as well as forests worldwide. In light of predicted global changes and the increasing focus on forest carbon storage capabilities, our results indicate that forest management practices should consider dead trees as integral elements of forest dynamics [4,20,41]. Future studies should delve into the microbial ecosystems within deadwood and examine the interactions between dead and living biomass in subtropical forests.

Both empirical and theoretical research have yielded diverse perspectives on the correlation between tree size and survival rates, with a consistent observation being the increased mortality rates among smaller trees [37,38,40]. Typically, epiphytes, or trees that grow on other trees, exhibit higher mortality due to their reduced ability to cope with environmental stressors. Our results are in line with forest studies that suggest higher mortality rates are more common in larger tree categories, possibly because these larger trees, with their extensive canopies, encounter less competition from surrounding trees.

This diminished competition grants them improved access to vital environmental resources such as water, nutrients, and sunlight, owing to the uneven nature of competition [1,5,43]. Conversely, smaller trees face higher mortality rates due to severe interspecific competition, rendering them more vulnerable to environmental shifts. Larger trees, often found in more advantageous environmental settings through a phenomenon known as environmental filtering, question the relevance of metabolic ecology theory in this subtropical forest context. This theory, which is frequently applied to tropical forests, argues that populations of varying sizes absorb and utilize energy at comparable rates, a premise that may be valid in some forests but not necessarily in diverse, natural mixed forests [45].

#### 5. Conclusions

In our study, we explored the spatial distribution and determinants of deadwood biomass over a period of five years in a subtropical forest environment. Our analysis revealed that the composition and characteristics of the tree community, particularly of those trees that have died, varied across different treatments of the study site. From 2012 to 2017, the carbon sequestered in deadwood biomass amounted to 1.5 tons. The changes in tree density and diameter at breast height (DBH) were identified as key factors influencing the accumulation of deadwood biomass within the research plot. The biomass of dominant tree species accounts for the vast majority of dead wood biomass, indicating that the addition of nitrogen and water can also harm dominant tree species. The findings from our research contribute to a deeper understanding of carbon storage processes within temperate forest ecosystems and can assist in improving and validating models of carbon cycling.

**Author Contributions:** Writing—original draft preparation, L.H.; formal analysis, S.T.; validation, T.L.; methodology, L.F.; data curation, X.S.; writing—review and editing, G.D.; Supervision, J.F.; Conceptualization, L.M. All authors have read and agreed to the published version of the manuscript.

**Funding:** This work was supported by the NSFC-Henan Joint Fund (grant number U1904204), National Natural Science Foundations of China (No. 31971653), Xinyang Academy of Ecological Research Open Foundation (2023DBS02, 2020DBS03, 2023XYQN02), Central Plains Scholars Workstation Sponsored Project "Research on the Purification Function of Forests on the Environmental Air in Xinyang City" (234400510026), Henan Provincial Natural Science Foundation Project (232102320120, 242102320331), and Background investigation and data integration of forest ecosystem status (2021FY100702).

**Data Availability Statement:** The data that support the fundings of this study are available from the corresponding author.

**Acknowledgments:** We acknowledge the work of the principal investigators and their field assistants in collecting the field data on the experiment plots. We would like to thank Joseph Elliot at the University of Kansas for her assistance with the English language and grammatical editing of the manuscript.

Conflicts of Interest: The authors declare no conflicts of interest.

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