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Mixed-Species Plantation Effects on Soil Biological and Chemical Quality and Tree Growth of A Former Agricultural Land

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Citation: Danise, T.; Andriuzzi, W.S.; Battipaglia, G.; Certini, G.; Guggenberger, G.; Innangi, M.; Mastrolonardo, G.; Niccoli, F.; Pelleri, F.; Fioretto, A. Mixed-Species Plantation Effects on Soil Biological and Chemical Quality and Tree Growth of A Former Agricultural Land. *Forests* **2021**, *12*, 842. https://doi.org/10.3390/f12070842

Academic Editor: Timo Domisch

Received: 27 April 2021
Accepted: 18 June 2021
Published: 25 June 2021

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Abstract: Tree planting on abandoned agricultural land could both restore the soil quality and increase the productivity of economically valuable woody species. Here, we assess the impact of mixed-species tree plantations on soil quality at a site in Central Italy where tree intercropping systems were established 20 years ago on a former agricultural land. These intercropping systems include two species of economic interest, *Populus alba* and *Juglans regia*, and one of three different nurse trees, i.e., *Alnus cordata*, *Elaeagnus umbellata*, both of which are N-fixing species, and *Corylus avellana*. We measured tree growth and compared how soil organic matter, soil extracellular enzymes, and nematodes of different feeding groups varied among the intercropping systems and relative to a conventional agricultural field. Our results indicate that tree plantation led to an increase in soil carbon and nitrogen, and enhanced enzyme activities, compared with the agricultural land. The proportion of nematode feeding groups was heterogeneous, but predators were absent from the agricultural soil. Multivariate analysis of soil properties, enzymatic activity, nematodes, and tree growth point to the importance of the presence N-fixing species, as the presence of *A. cordata* was linked to higher soil quality, and *E. umbellata* to growth of the associated valuable woody species. Our findings indicate that intercropping tree species provide a tool for both restoring fertility and improving soil quality.

Keywords: tree plantations; intercropping; soil ecology

1. Introduction

Cropland abandonment is an important process in many regions of the world and one of the dominant processes of land use change in Europe, where abandoned agricultural lands are estimated to cover between 20,277 and 211,814 km^2 by 2040 [1]. Policies and management of these areas have spurred debate regarding the loss of (traditional) agricultural landscapes and potential impacts on biodiversity and ecosystem services [2].

Multiple benefits are recognized to the revegetation of abandoned agricultural land, including hydrological regulation, mitigation of soil erosion, increased soil fertility, higher fungal biomass and decomposition activity, improved water quality, and carbon sequestration [3]. Tree plantation leads to recovery of microbial biomass [4] and to a higher availability of nutrients, such as N and P [5], from soil organic matter (SOM) mineralization. However, different woodland management types may have different ecological effects. Recent literature has shown the advantages of mixed-species plantation in comparison to pure stands in terms of nutrient cycling [6], soil fertility [7], biomass production [8], and carbon sequestration [9]. Mixed plantations are thus considered preferable to monocultures to maintain wood production while also promoting soil quality. The goal of such management is to combine different species to produce specific interactions that will increase stand-level productivity or individual-tree growth rates relative to monocultures. This enables the harvesting of products from different tree species on different rotations while reducing the risk of market shifts, or pest impacts, or some combination of both [10].

Mixed-species plantations containing nitrogen (N)-fixing species have the potential to increase the productivity and the ecophysiological performance of target tree species in comparison to monoculture [11] and to assist in the ecological restoration of degraded land [12]. Yet, there is comparatively scarce literature about the effects of mixed-species plantations on soil quality [13] in terms of chemical and biological characteristics. Symbiotic relationships with fungi and plant-associated microbes play key roles in tree–soil feedbacks [14], and interactions between plants and the complex webs of soil biota, from saprophytic bacteria to invertebrate predators, are also important [15]. The abundance of soil organisms, the composition of their assemblages, and their interaction with environmental factors determine their contribution to ecosystem functioning [16]. Soil enzyme activities are sensitive indicators of functional changes in soil [17] as they have important functions in the C and nutrient (N, P, and S) cycling, as well as in the biochemical degradation of organic pollutants [18].

Soil microbial activity, in turn, is widely affected by soil fauna, including nematodes. Abundant and functionally diverse [19], nematodes are useful indicators of soil food web dynamics [20]. Herbivorous nematodes interact directly with plants by feeding on roots, while bacterivores and fungivores affect the soil microbial community and thereby regulate organic matter decomposition and release of nutrients [21]. These nematodes are fed upon by omnivorous and predatory nematodes, which thereby indirectly affect both plants and microbes. Soil nematodes and other soil invertebrates (e.g., collembola, mites, and enchytraeids) not only react sensitively to soil disturbance and changes in climate [22], but also exert feedback effects on plant growth and plant community structure [23], for instance by releasing nutrients locked up in dead organic matter or in microbes, thus increasing nutrient availability and productivity of plants [24]. As the responses of nematodes to changes and their subsequent effects on microbes and plants vary among nematode taxa and functional groups, the analysis of nematode community composition (e.g., based on diversity and abundance within and across trophic groups [25]) provides a tool for assessing the effects of soil disturbance on soil functions and can be used as a biomonitoring system.

Given the complexity of soil–plant interactions [26], an integrated approach is essential to evaluate soil functioning along with tree productivity under different mixed-species tree plantation types. This could support the identification of best practices to manage former agricultural soils.

We hypothesize that mixed-species afforestation of a former agricultural soil will promote its biological activity compared with an adjacent active agricultural field (AL). We assess this hypothesis using univariate techniques and a multivariate approach based on the covariance between variables related to soil quality (i.e., organic carbon and nitrogen, microbial activity by enzyme analysis, and nematode functional groups) and tree growth of target species (i.e., basal area increment).

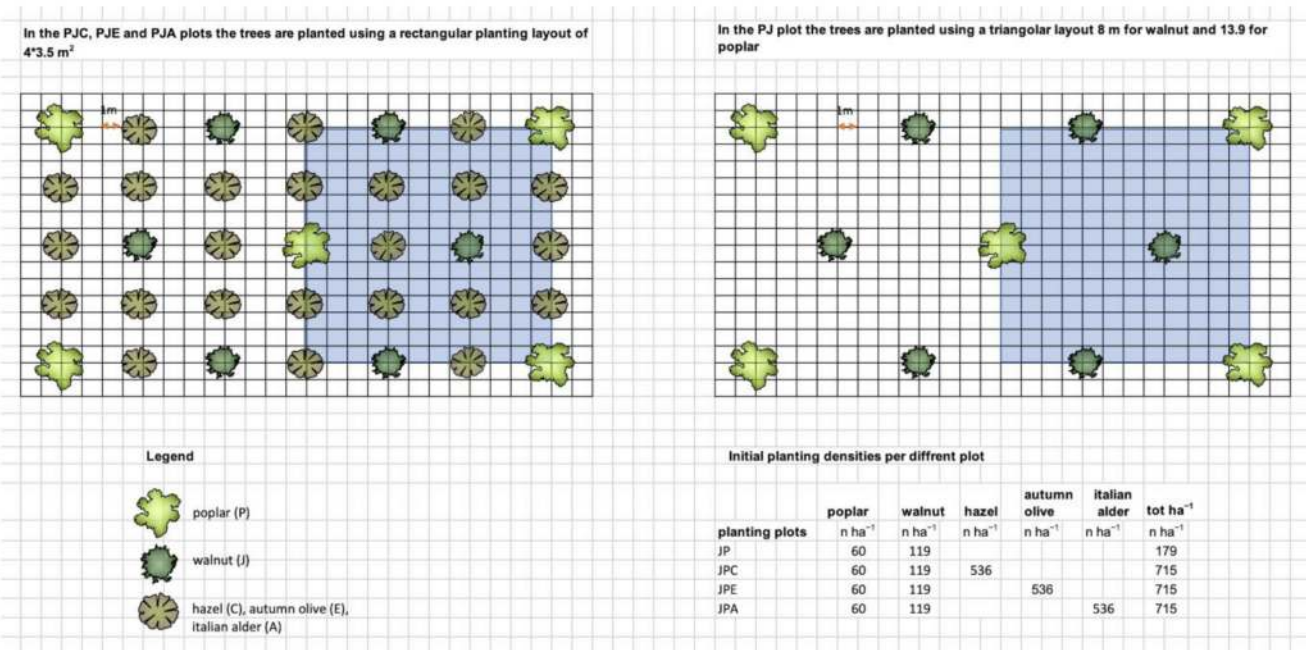
Furthermore, we hypothesize that plots with N-fixing nurse trees differ in terms of soil quality and in target species growth compared with plots without. Additionally,

we expect different effects of native and non-native N-fixing nurse trees, as found by Vilà et al. (2011) [27].

2. Methods

2.1. Study Site

The experimental area is located near Brusciana, Tuscany, Central Italy (43°40′29″ N, 10°55′21″ E), at 35 m a.s.l. The location has a mean annual precipitation of 850 mm and a mean annual temperature of 15 °C. The soils of the site developed on recent (Holocene) fluvial deposits and are Fluventic Haplustepts coarse-loamy, mixed, thermic of the U.S. Soil Taxonomy, according to the 1:250,000 soil map by Regione Toscana (http://sit.lamma.rete.toscana.it/websuoli/, accessed on 5 March 2021). In 1996, an experimental tree plantation was established for the production of wood on agricultural land. This plantation was polycyclic, i.e., different crop trees combining fast growing and rotation periods were planted together: (i) broadleaved crop trees with medium-long rotation (generally 20–30 years); (ii) crop trees with medium-short rotation (generally 10–15 years), namely poplar clones; (iii) crop trees with short rotation (generally 3–7 years). Trees were planted at a fixed distance to reach commercially valuable size [28]. In this trial, two target broadleaved tree species widely used in Italy for timber production, poplar (*Populus alba* L., Salicaceae) and walnut (*Juglans regia* L., Juglandaceae), were planted together, using a triangular layout with a distance of 8 m (179 trees ha^{-1}), and intercropped with N-fixing species, such as *Elaeagnus umbellata* and *Alnus cordata* and with another nurse shrub such as *Corylus avellana*, using a rectangular layout of 3.5 × 4 m^2 (715 trees ha^{-1}) (Figure 1). No thinning was performed in the plantation. Planting plots (≈0.4 ha each) were compared using a randomized blocks design with three replicates:



planting plots	poplar n ha^{-1}	walnut n ha^{-1}	hazel n ha^{-1}	autumn olive n ha^{-1}	italian alder n ha^{-1}	tot ha^{-1} n ha^{-1}
JP	60	119				179
JPC	60	119	536			715
JPE	60	119		536		715
JPA	60	119			536	715

Figure 1. Planting layout of the different afforested plots (PJ, PJC, PJE, and PJA).

1. Poplar and walnut (plots PJ), planted in a high-density mixture.
2. Poplar and walnut intercropped with hazel (*Corylus avellana* L., Betulaceae) (plots PJC), a native shrub frequent in the surrounding forests.
3. Poplar and walnut intercropped with autumn olive (*Elaeagnus umbellata* Thunb., Elaeagnaceae) (plots PJE), an alien N-fixing shrub used in tree farming plantations for its high ability to fix nitrogen in the soil.

4. Poplar and walnut intercropped with Italian alder (*Alnus cordata* (Loisel.) Duby, Betulaceae) (plots PJA), an N-fixing tree species common in Southern Italian forests and widely used in tree farming plantations.

Soil samples were also collected from an adjacent active agricultural field (AL).

2.2. Sampling

The following sampling procedure was carried out in each field replicate (in both afforested and agricultural soil). In November 2018, in the centroid of each of the 0.4 ha planting plots, we designed a regular 12 × 12 m^2 grid. At the central point of each grid and at four microsites 6 m away (in the direction of the cardinal points), we collected mineral soil with a core sampler (diameter 5 cm; length 10 cm) to a depth of 10 cm, resulting in an overall sample size for soil analyses of 75 (i.e., 5 soil cores × 3 field replicates × 5 treatment plots). Within each tree planting plot, four dominant trees of *P. alba* and four trees of *J. regia* were sampled using a 5.1 mm diameter Haglof increment borer, collecting two cores per tree at an angle of 120° from each other.

It should be taken into account that sampling error may have been comparatively large due to small sample size [29].

All the *P. alba* and *J. regia* presented in the plots were alive and of comparable diameter and height (tree height and mean diameter at breast height, respectively, 43 ± 6 cm and 22 ± 3 m *P. alba*; 17 ± 3 cm and 12 ± 2 m, *J. regia*).

2.3. Physical and Chemical Soil Analysis

Once in the laboratory, the soil samples were air-dried and then sieved to remove rock fragments (>2 mm). Soil pH was measured potentiometrically using a 2.5:1 (*v*/*w*) 0.01 M $CaCl_2$ solution to soil ratio. Particle size distribution was determined by the hydrometer method on one combined sample per plot (i.e., three replicates per stand type), obtained by bulking equal aliquots from each plot. Soil bulk density was measured by dividing the weight of the undisturbed core samples, after drying at 105 °C in an oven to constant weight, by the volume of the steel cylinder of the sampler.

Soil organic matter (SOM) was determined gravimetrically by incineration in a muffle furnace at 375 °C for 16 h [30]. Total carbon and nitrogen (TC and TN) were determined by dry combustion on an elemental analyser (Elementar Analysensysteme GmbH, Vario EL III from Elementar, Langenselbold, Germany) [31], while total inorganic carbon (TIC) was measured volumetrically using a Scheibler calcimeter [31]. Total organic carbon (TOC) was calculated as the difference between TC and TIC. All chemical variables were determined in each plot on 5 field cores and 3 laboratory replicates.

2.4. Biological Analysis

Enzyme activities were measured in fresh soil stored at −80 °C [32] by spectrophotometric methods, using 4-nitrophenyl-β-D-glucopyranoside as a substrate for β-glucosidase activity (EC 3.2.1.21) [33], 4-nitrophenyl phosphate bis(cyclohexylammonium) salt as a substrate for acid phosphomonoesterase activity (EC 3.1.3.2) [34], and urea as a substrate for urease activity (EC 3.5.1.5) [35]. Fluorescein diacetate hydrolase (FDAH) activity was determined according to Green et al. (2006) [36]. Each enzyme activity was tested in each plot on 5 field cores and 3 laboratory replicates. Enzyme activities are reported as μmol (for urease in μg) of product developed in one hour per gram of dry matter.

The AI3 index, as validated by Puglisi et al. (2006) [37], is calculated by a linear combination of three different enzymes as follows:

$$\begin{gathered}\text{AI3} = 7.87\ \beta\text{-glucosidase}\ (\mu\text{mol pNP g}^{-1}\ \text{d.w. h}^{-1}) - 8.22\ \text{acid} \\ \text{phosphomonoesterase}\ (\mu\text{mol pNP g}^{-1}\ \text{d.w. h}^{-1}) - 0.49\ \text{urease}\ (\mu\text{g N-NH4}^{+}\ \text{g}^{-1}\ \text{d.w. h}^{-1})\end{gathered} \quad (1)$$

This index is used to assess the quality of soils affected by various forms of degradation such as irrigation with saline water, contamination by heavy metals, organic fertilizer, erosion, and contamination by industrial and/or urban waste, as well as forest soils [32].

High values indicate a low soil biological quality [37], while the index usually assumes negative values in high-quality soils.

2.5. Nematodes

Soil nematodes were extracted from 50 g fresh soil aliquots using Baermann funnels over 3 days. The aliquots were obtained by mixing together 10 g of soil from each field replicate, in order to obtain a pool. Each aliquot was then divided in two replicates of 25 g each. Nematodes collected in water were removed daily and stored at 4 °C prior to identification. Within 5 days of extraction, the nematodes were counted and identified using an optical microscope (Leica DMLS, Wetzlar, Germany; 100× magnification). Based on morphology, they were allocated to the following feeding groups: bacterivores, fungivores, herbivores, omnivores, and predators [38]. A few individuals that could not be assigned with confidence to any of the above were classified as unknown.

2.6. Tree-Ring Measurement and Basal Area Increment of Individual Trees at the Species Level

The core wood samples were air-dried and then polished with a series of successively finer sand-paper grits until rings were clearly visible. Tree rings were measured to the nearest 0.01 mm using a binocular stereoscope and a LINTAB measuring device (Rinntech, Heidelberg, Germany). Cross-dating of the tree-ring series was checked using the program COFECHA [39].

Basal area increment (BAI) of both target species, *J. regia* and *P. alba*, was calculated as the average accumulated increment of the tree rings following the equation:

$$\mathrm{BAIt} = \pi r^2{}_t - \pi r^2{}_{t-1} \quad (2)$$

where BAI at year t is equivalent to the annual ring area, r_t is the stem radius at the end of the annual increment, and r_{t-1} is the stem radius at the beginning of the annual increment. BAI series were used as they minimize the effect of tree size and age on annual growth trends while retaining the high- and low-frequency signals contained in the tree ring series (see Battipaglia et al. (2017) [11] for details), unlike ring width series and with the exception of the first years (3–5 years) of increasing juvenile growth rates.

2.7. Statistical Analyses

One-way analysis of variance (ANOVA) was used to test the effects of the different stands on the response variables, followed by Tukey's post-hoc test ($\alpha = 0.05$). The model residuals were checked for normality and homogeneity of variance. In order to investigate patterns of covariance between soil biological data (i.e., enzyme activities and proportion of nematode functional groups) and tree basal area, we employed a multivariate statistic known as Two-Block Partial Least Squares (2B-PLS). This statistical approach can provide reliable inference when dealing with matrices that have a comparatively low sample size as well as highly correlated variables [40,41]. This technique has recently been applied in several ecological contexts [32,42,43]. Patterns of covariance between the two matrixes can be represented by a scatterplot for the first axis of the 2B-PLS, where the *x*-axis and the *y*-axis represent the two multivariate matrixes, respectively. Patterns of positive or inverse correlation can be asserted both within and between matrixes. All analyses were done in R 3.6.1 [44], using packages 'plsdepot' and 'ggplot2'. Results are reported as mean ± standard error.

3. Results

3.1. Soil Properties

Soil texture was mostly sandy loam. The contribution of the clay fraction was rather homogeneous in the different stands, ranging between 15% and 20% of the total mineral mass (Table 1). However, there were some differences in terms of sand and silt fractions. In particular, PJA stands had the lowest sand content and AL had the highest silt content. In spite of these differences in particle size distributions, soil bulk density was virtually the

same throughout the investigated area. Soil pH was neutral and did not differ significantly between the stands, while it was slightly higher in AL.

Table 1. Soil particle-size distribution, bulk density, and pH of the investigated tree stands and the arable land. Site labels: PJ = white poplar and common walnut; PJC = PJ intercropped with common hazel; PJA = PJ intercropped with Italian alder; PJE = PJ intercropped with autumn olive; AL = agricultural land. Values are mean and standard deviation (n = 5 for soil particle-size distribution and 15 for bulk density and pH). Superscript letters indicate significant differences within means in the column according to one-way ANOVA at $p \leq 0.05$ and Tukey's test.

Stand	Sand	Silt	Clay	Bulk Density	pH_{CaCl2}
	%	%	%	kg dm^{-3}	
PJC	59.5 ± 5.5 ^{a}	25.6 ± 7.9 ^{a}	14.9 ± 2.6 ^{a}	1.4 ± 0.1 ^{a}	7.0 ± 0.2 ^{a}
PJE	53.7 ± 3.8 ^{a,b,c}	29.4 ± 1.4 ^{ab}	16.9 ± 3.2 ^{a}	1.5 ± 0.2 ^{a}	7.1 ± 0.2 ^{a}
PJA	37.5 ± 8.2 ^{b}	42.4 ± 6.7 ^{b}	20.0 ± 2.0 ^{a}	1.4 ± 0.2 ^{a}	7.0 ± 0.2 ^{a}
PJ	43.7 ± 5.3 ^{c}	38.0 ± 4.3 ^{b}	18.3 ± 3.2 ^{a}	1.4 ± 0.1 ^{a}	7.0 ± 0.1 ^{a}
AL	68.7 ± 6.4 ^{a}	15.1 ± 8.2 ^{a}	16.2 ± 1.9 ^{a}	1.5 ± 0.1 ^{a}	7.3 ± 0.2 ^{b}

Overall, soil TC, TN, and SOM contents were higher in the tree stands than in AL (Table 2). Among the stands, PJE had the smallest TC and TOC values, and PJA the highest TC, TN, and TOC. Soils supporting the N-fixing species (PJA and PJE) were the poorest in TIC, while the arable soil (AL) was the richest.

Table 2. Soil chemical properties of the investigated tree stands and the arable land (see Table 1 for site labels). Values are mean and standard error (n = 15). Superscript letters indicate significant differences within means in the column according to one-way ANOVA at $p \leq 0.05$ and Tukey's test.

Stand	SOM mg g^{-1}	TC mg g^{-1}	TN mg g^{-1}	TOC mg g^{-1}	C_{org}/N	TIC mg g^{-1}
PJC	34.6 ± 2.46 ^{a}	32.6 ± 1.19 ^{a,b}	1.3 ± 0.08 ^{b}	17.4 ± 1.17 ^{a,b}	13.4 ± 0.55 ^{a}	15.1 ± 0.40 ^{a,b,c}
PJE	34.0 ± 1.59 ^{a}	28.7 ± 0.73 ^{b,c}	1.4 ± 0.05 ^{ab}	14.5 ± 0.82 ^{b}	10.2 ± 0.27 ^{b}	14.2 ± 0.34 ^{b,c}
PJA	35.0 ± 1.21 ^{a}	33.3 ± 1.86 ^{a,b}	1.8 ± 0.12 ^{a}	19.1 ± 1.83 ^{a}	10.6 ± 0.28 ^{b}	14.2 ± 0.85 ^{c}
PJ	35.8 ± 1.20 ^{a}	32.9 ± 0.87 ^{a}	1.5 ± 0.07 ^{ab}	16.4 ± 0.78 ^{a,b}	11.0 ± 0.41 ^{b}	16.5 ± 1.07 ^{a,b}
AL	20.4 ± 1.06 ^{b}	25.7 ± 0.59 ^{c}	0.8 ± 0.03 ^{c}	8.3 ± 0.22 ^{c}	10.5 ± 0.29 ^{b}	17.4 ± 0.73 ^{a}

3.2. Soil Enzymatic Activities

Fluorescein diacetate hydrolase (FDAH) rates were highest in the presence of nitrogen-fixing species (PJA and PJE stands, with mean values and standard error 0.26 ± 0.028 and 0.22 ± 0.008 µmol fluorescein/g d.w./h, respectively) (Figure 2). The lowest FDAH activity was found in AL (0.14 ± 0.007 µmol fluorescein/g d.w./h), and PJ and PJC were in between (0.19 ± 0.008 and 0.18 ± 0.01 µmol fluorescein/g d.w./h).

The activity of the enzymes β-glucosidase, urease, and acid phosphomonoesterase was about twice as high in the tree stands as in AL (Figure 3).

There was no difference between the tree plantation plots for β-glucosidase (AL: 0.024 ± 0.001; PJ: 0.05 ± 0.003; PJC: 0.04 ± 0.004; PJA: 0.05 ± 0.004; PJE: 0.04 ± 0.003 µmol pNP/g d.w./h) and urease (AL: 8.517 ± 1.114; PJ: 24.303 ± 3.576; PJC: 22.495 ± 2.243; PJA: 34.277 ± 3.561; PJE: 18.568 ± 1.229 µg N-NH_4^+/g d.w./h). In terms of acid phosphomonoesterase, PJ (0.048 ± 0.003 µmol pNP/g d.w./h) and PJC (0.037 ± 0.002 µmol pNP/g d.w./h) were different, while the PJA and PJE plots were similar to each other but different from the other plots (Figure 3).

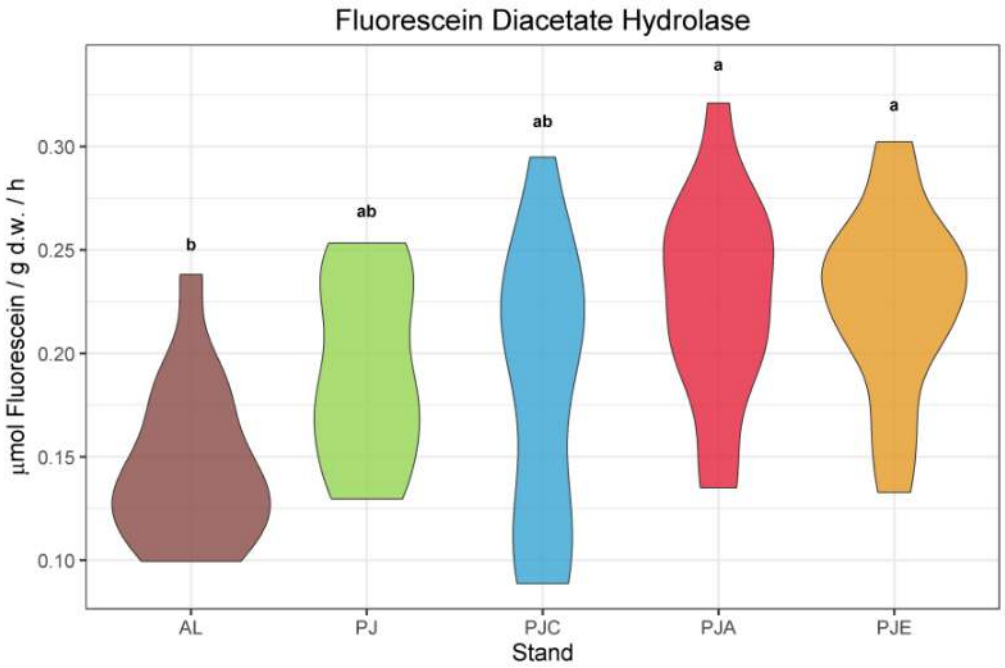


Figure 2. Violin plot of the fluorescein diacetate hydrolase (FDAH) activity in the tree stands and agricultural land plots (see Table 1 for site labels). Different letters indicate significant differences ($p \leq 0.05$, Tukey's post-hoc test following one-way ANOVA).

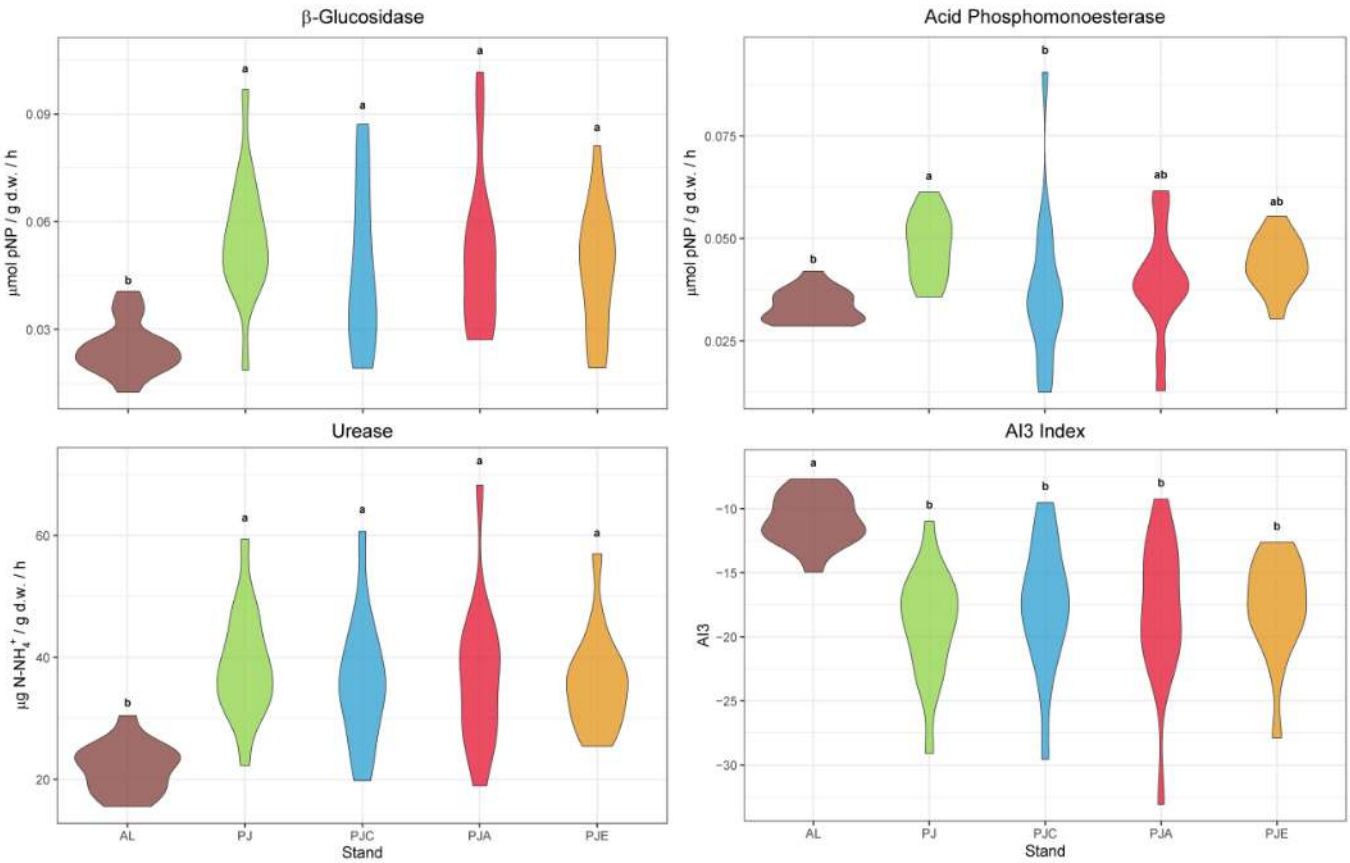


Figure 3. Violin plot of the enzymatic activities in the tree stands and agricultural land plots (see Table 1 for site labels). β-glucosidase; acid phosphomonoesterase; urease; AI3 index. Different letters indicate significant differences ($p \leq 0.05$, Tukey's post-hoc test following one-way ANOVA).

Overall, enzymatic activities were twice as high in the tree plantation plots as in the agricultural field. As a result, the AI3 index calculated from the three enzyme activities was highest in AL, indicating lower soil biological quality (Figure 3). This suggests that tree plantations improved soil quality.

3.3. Nematodes

Herbivores were the nematode functional group with the highest abundance overall, with the maximum relative proportion in PJ (0.59) and PJE (0.66), followed by bacterivores, with the greatest relative abundance in AL (0.49) and PJC (0.45) (Figure 4). The highest proportion of fungivores was found in AL (0.10) and PJ (0.8), while omnivores were more abundant in PJC and PJE (0.05). Generally, predators were the least represented group, ranging from none in AL to 0.04 in PJC. The proportion of unknown was low across land uses (min. 0.01 in AL, PJ, PJE and max 0.03 in PJC). Overall, the agricultural field tended to have higher relative abundance of bacterivores and fungivores compared with the tree plantations, but the differences were not statistically significant.

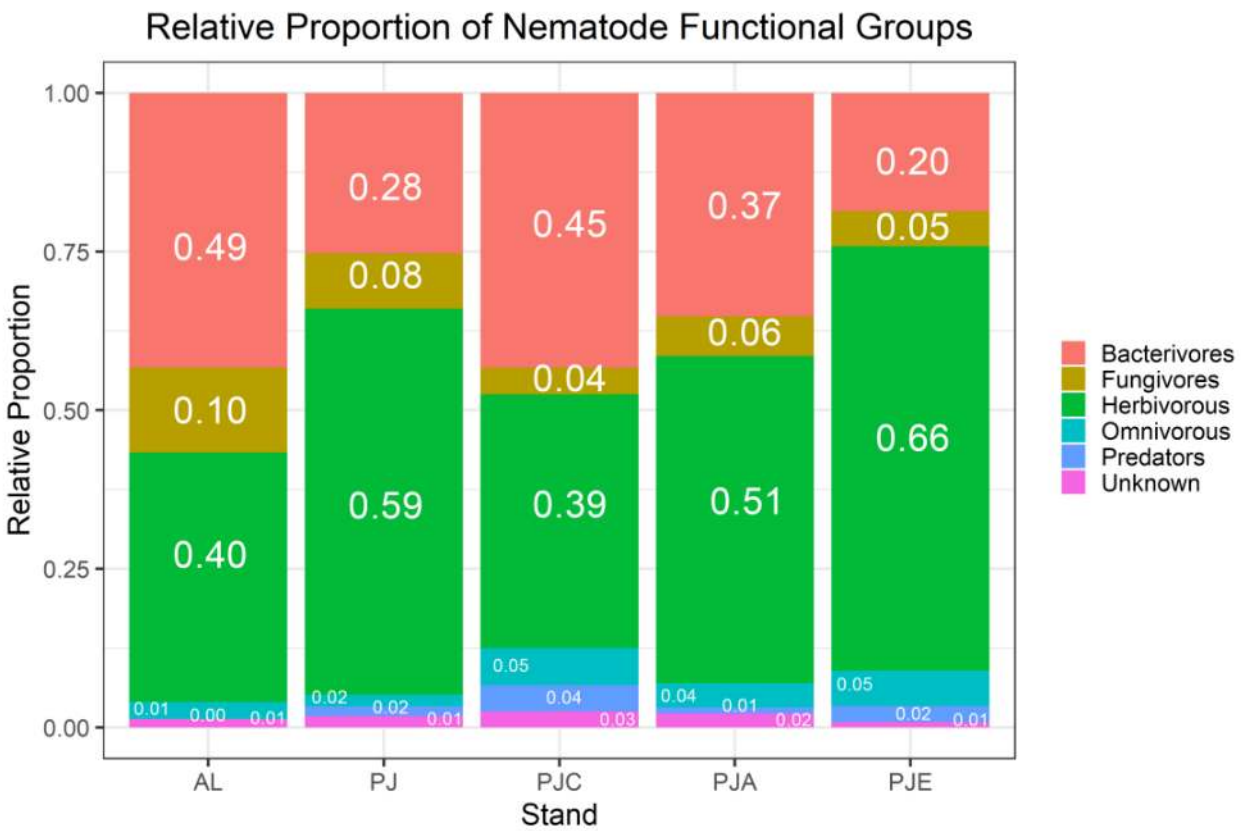


Figure 4. Nematode functional group composition (relative proportion) in the tree stands and agricultural land plots (see Table 1 for site labels).

3.4. Basal Area Increment (BAI)

For both target tree species *J. regia* and *P. alba* (Figure 5a,b), BAI was highest in the intercropping with *E. umbellata* (PJE). PJC and PJA stands had similar BAI (possibly with an outlier in PJA determining this similarity) and distinct from PJE and PJ. Finally, the PJ stand was also distinct from PJE (despite an outlier).

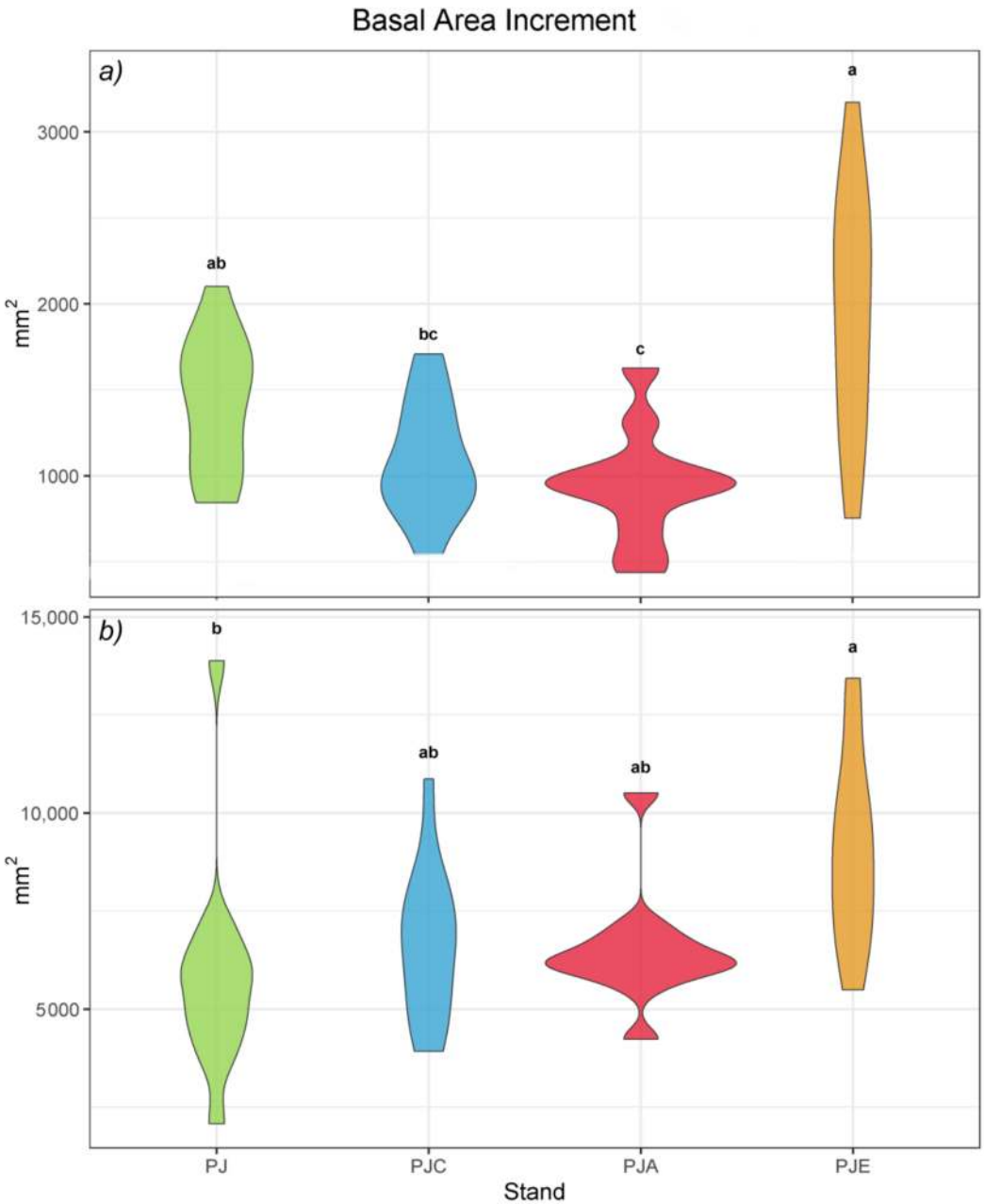


Figure 5. Violin plot of the basal area increment (BAI) of (**a**) *Juglans regia* and (**b**) *Populus alba* trees expressed in mm^2. Different letters indicate significant differences ($p \leq 0.05$, Tukey's post-hoc test following one-way ANOVA). See Table 1 for site labels.

3.5. Two-Block Partial Least Squares Analysis

The 2B-PLS (Figure 6) separated the PJE plot from the other plots. Considering the large degree of variability within plots (as shown by the error bars), on the *y*-axis (i.e., basal area increment) PJA, PJC, and PJ were largely overlapping. On the *x*-axis (i.e., biological data), instead, PJA was marginally distinct from PJC and PJ. The correlation coefficients within and between blocks indicated that PJA (and to a lesser extent PJC and PJ) had lower cumulative basal area compared with the other stands; this was inversely correlated with the enzyme activities of β-glucosidase, acid phosphomonoesterase, and urease as well as the proportion of unknown and bacterivore nematodes. On the other hand, PJE was different, showing the highest cumulative basal area, which was linked to the highest AI3 values and positively correlated with the proportion of predatory and herbivore nematodes. Overall, considering that higher AI3 values are linked to lower soil

quality, a difference between PJA and PJE was found: PJA showed a higher soil quality but lower tree growth, while the opposite was true for PJE. Enzyme activities and functional abundance of nematodes were consistent with this pattern. FDAH and the proportion of omnivore and fungivores nematodes had a low correlation coefficient within Block 1, indicating that these variables contribute poorly to the observed pattern of covariance.

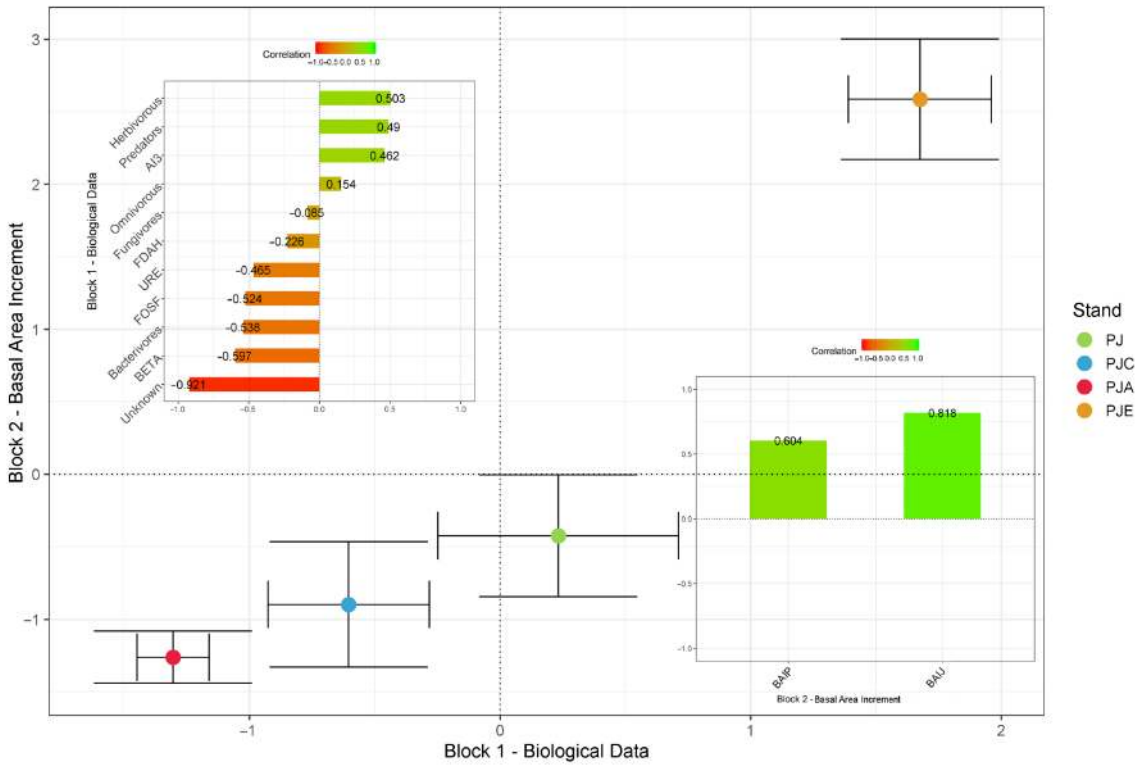


Figure 6. Scatterplot for the first axis of the 2B-PLS. Points are mean ± standard error across Block 1 (biological data) and Block 2 (basal area increment). The insets show the correlation within and between blocks of the variables.

4. Discussion

4.1. Soil Properties

Soil pH may respond rapidly to afforestation because of the change in the base cation cycle and the addition of plant residues [45], which also lead to higher accumulation of organic matter in soil [46]. Therefore, the slight but significantly lower pH we measured in the tree plantation plots compared with the agricultural land was plausibly due to higher plant uptake of base cations [45] and higher organic residue deposition and decomposition, which imply a net input of protons to the soil [47]. N-fixing trees promote soil acidification in mixed stands because of both greater nitrate leaching and release of strongly acidic soil organic matter (SOM) [48]. In our study, however, soil pH clustered around neutrality.

The tree plantation soils showed significant differences from AL in terms of all the other investigated soil abiotic properties, i.e., TC, TOC, TN, and SOM. Previous research highlighted that changes in land use influence soil fertility and quality indicators because of subsequent alterations in abiotic and biotic factors along with SOM stabilization [49]. Land use change alters C and N inputs and dynamics [50], which subsequently regulate microbial processes and soil physical and chemical features [51]. Afforestation of arable lands has been shown to enhance soil C stocks, albeit only in the long term [52]. In fact, whereas tillage destroys the soil structure, enhancing organic matter mineralization and CO_2 emissions [53], afforestation implies a continuous input of above-ground and root litter [46]. In this study, we found differences between the tree stands and the arable field in terms of TOC and TN, as well as SOM content, despite the relatively short time since the tree plantations' establishment. The differences between the different tree mixtures

were smaller. The PJA stands, with alders, showed the highest soil TC, TOC, and TN contents, consistently with Chiti et al. (2007) [54] and Chodak and Niklinska (2010) [55]. Alder is symbiotic with N-fixing actinomycetes and promotes soil N enrichment, leading to nutrient-rich litter and higher soil fertility [56]. Symbiotic N fixation may increase not only soil TN but also TOC [57], as a result of both the slower decomposition rate of humified organic matter and greater C inputs [58]. Innangi et al. (2017) [43] showed that Italian alder leaf litter has low lignin and high ethanol extractable contents, which promote a fast litter decomposition rate and, consequently, a high quantity of soil TOC that can build up as microbial C [59]. The stands without N-fixing species, i.e., PJ and PJC, gave contrasting results in terms of soil C_{org}/TN ratio; we expected this ratio to be higher here than in the other stands, but this was true only for PJC.

4.2. Enzymatic Activities and Soil Quality

All enzymatic activities had lower values in AL compared with tree stands, except the acid phosphomooesterase in PJC (Figures 2 and 3). The enhanced enzymatic activities could be attributed to the increase in soil TOC and TN stocks due to plant residue inputs with afforestation [60], as this provides more substrate for microbial assimilation [61].

Overall, soil microbial activity was not significantly different under N-fixing and non-N-fixing trees, except for FDAH (Figure 2) and acid phosphomonoesterase (Figure 3). Similarly, previous studies found no difference in microbial community activity between the non-N-fixing *Pseudotsuga menziesii* (Mirb.) Franco and N-fixing *Alnus rubra* Bong. trees in the forest of northwestern North America [62], or in microbial biomass C and N under *Acacia mangium* Willd. compared with *Eucalyptus grandis* W. Hill in a 20-month-old mixed-species planting in Brazil [63]. Nonetheless, in the latter case, there was significantly more dehydrogenase enzyme activity under *A. mangium* than *E. grandis*, suggesting some differences in the microbial community activity underneath these N-fixing and non-N-fixing tree species. Our results on soil enzymatic activity are in agreement with Bini at al. (2013) [63] since FDAH, as well as dehydrogenase, has usually been considered a good indicator of microbial biomass in soils [42].

While FDAH was higher in the presence of nitrogen-fixing species, acid phosphomonoesterase increased with *C. avellana.* Acid phosphomonoesterase is involved in P cycling as it catalyzes the hydrolysis of organic P esters to inorganic P [64]. Since the synthesis of phosphomonoesterase may be suppressed by the presence of inorganic P, high phosphomonoesterase activity in PJC could indicate insufficient P supply [55].

The activity of several soil enzymes is considered a reliable indicator of soil quality under different tillage practices and soil management [65]. This is supported by our findings. The AI3 index, based on enzymatic activities, showed that establishment of mixed-tree intercropping systems increases the quality of the soil compared with the agricultural field (Figure 3). Although the mixtures in our study have a low number of species, this result is consistent with the finding by Lange et al. (2015) [66] that tree species diversity can increase SOM and biochemical properties, presumably via higher root inputs and other yet unidentified mechanisms.

4.3. Nematodes

Microfauna grazing on microorganisms, such as nematodes, affects the growth and metabolic activities of microorganisms and alters the microbial community, thus regulating rates of decomposition [67] and nutrient mineralization [68]. Our study showed a heterogeneous pattern among the different forest associations, particularly regarding whether herbivores and bacterivores dominated the community. Yeates and Bongers (1999) [69] showed a dominance of bacterivores for both *Populus* and *Alder* soils (0.43 and 0.80, respectively), while herbivores had lower proportions (0.27 *Populus*; 0.07 *Alder*). In our study, the only intercropping system in line with these results is PJC, where the proportion of bacterivores was slightly higher than that of herbivores. By contrast, in PJE herbivores were more than three times as abundant as bacterivores. Armendàriz and Arpin (1996) [70]

found a greater relative abundance of bacterial feeder nematodes during the first stages of colonization of the soil community succession, while an increase in plant-feeders was observed later. However, the stands in our study were established in the same year and had generally similar soil properties. The greater abundance of herbivores in PJE could be due to the higher tree biomass, the presence of the alien N-fixer *E. umbellata*, and/or due to unmeasured soil or biological properties. Regardless of the causes, the pattern suggests a greater energy flow from living roots to the soil food web in PJE than in PJC. Laboratory experiments and field studies have demonstrated that nematodes that feed on bacteria and fungi play important roles in influencing the turnover of soil microbial biomass and, thus, the availability of plant nutrients [15]. Differences in relative abundance of bacterial-feeding and fungal-feeding nematodes reflects differences in decomposition pathways or channels [71].

Plant-feeding nematodes have been reported to be more sensitive to changes in plant community composition than other groups of nematodes, presumably due to their selective feeding on certain host plants [67,69]. In fact, a global analysis of nematode communities found that edaphic factors (such as sand content and pH) were stronger predictors of communities dominated by bacterivores, whereas vegetation cover was predictive of herbivore-dominated communities [19]. The dominance of bacterivore nematodes in the agricultural soil in our study is in line with results of Yeates and Bongers (1999) [69], who found that bacterivores dominated with a relative proportion of 0.43 in a ploughed corn/soybean rotation cultivated field. Bacterivore abundance generally increases with the incorporation of cover crop residues, often attributed to the increase in bacterial biomass after cover crop additions [72]. However, in the study of Yeates and Bongers (1999) [69], the relative proportion of fungivore nematodes was much higher (0.27) than in the agricultural soil of our study (0.10). The greater relative quantity of other groups could be explained by the fact that fungi and fungivore nematodes are more likely to regulate decomposition of surface residues in no-till soils, whereas bacteria and bacterivorous nematodes regulate decay rates in incorporated residues in conventional-till soils [67], as in our case. Lastly, we found few predatory nematodes overall, but they were notably absent only from the agricultural plots, suggesting a more mature food web in the tree plantations.

4.4. Basal Area Increment

Tree growth is influenced by a wide variety of factors. Although we sampled trees in the centre of each plot, our results could be influenced by the limited extension of the plots and by edge effects, in which conditions near the boundary of two habitats differ from the interior. This can affect tree community composition, structure, and possible tree mortality [73,74]. However, mixed plantations, which are often more productive than monocultures [75], are characterized by pluri-stratified canopies that may increase light availability for the species in the dominant layer [76]. *Elaeagnus umbellata* is a shrub; hence, it does not compete for light with larger, economically valuable tree species, while at the same time enriching soil in nitrogen as it is an N-fixing species. As reported by Forrester et al. (2006) [76], regarding the availability of N-fixing species at stand level, once N has been transferred to the non-N-fixing species, this can result in higher CO_2 assimilation rates [77]. However, our study showed that PJA had lower basal area values for both target species (Figure 5a,b) compared with PJE.

Battipaglia et al. (2017) [11] reported a significant increase in basal area in *Quercus robur* L. growing together with the native N-fixing species *A. cordata*. Mixing valuable tree crops such as *J. nigra* L. or *J. regia* with N-fixing *E. umbellata* can improve tree growth performance of the former species as a result of increased soil N mineralization rates [78]. Panshke et al. (1989) [79] estimated N mineralization rates in a 19-year-old intercropping of black walnut (*J. nigra*) with N-fixing autumn olive (*E. umbellata*.) or black alder (*A. glutinosa*) and in pure walnut plantings at two locations in Illinois, USA. Walnut size was highly correlated with soil N mineralization, particularly soil NO_3-N production in a plot. Total N and TOC contents were smallest in *Elaeagnus* interplantings at both sites, in concordance

with our results, but NO_3-N concentration was higher in the *Elaeagnus* plot than in the *Alnus* plot. Khamzina et al. (2009) [78] also showed that the increase in plant-available soil N was significantly higher in *Elaeagnus angustifolia* L. plots than in *Populus euphratica* Oliv. and *Ulmus pumila* L. plots. All this suggests that *Elaeagnus* favours the formation of N-NO_3, which, in turn, could positively affect the growth of target species.

4.5. Overall Trends

The plots with N-fixing species were markedly different, as highlighted by the 2B-PLS (Figure 6). While PJA was distinguished by its higher enzymatic activity values and higher soil quality as indicated by AI3 index, PJE was distinct because of its lower soil quality but higher BAI values. A key difference between the two N-fixing species lies in the fact that *E. umbellata* is an exotic species. Large differences in microbial community have been reported in ecosystems invaded by exotic N-fixers [80]. Several alien plant species have been shown to decrease local plant species diversity, increase ecosystem productivity, and alter the rate of nutrient cycling [81]. As a strong impact on nutrient cycling affects plant performance (e.g., plant resource allocation, plant competitive ability, plant resistance to herbivores, etc.) and, hence, community structure, Vilà et al. (2011) [27] assumed alien N-fixing plants to have greater community impacts than alien non-N-fixing species. Ge et al. (2018) [82] found that adding root extracts of the alien species *Alternanthera philoxeroides* (Mart.) Griseb. to the soil significantly decreased the soil microbial community activity and the relative abundance of microorganisms. Liang et al. (2016) [83] showed that allelochemicals released by invasive species might not only affect seed germination, root growth, and biomass of native plant species, but also impact soil enzymatic activities, soil microbial community, and soil functions, thus both directly and indirectly affecting the growth and development of native plants. However, further analyses are needed to clarify the reasons for the different results we obtained in the presence of a native and non-native N-fixing species.

5. Conclusions

Our results suggest that the afforestation of arable soils is a valid strategy for improving soil quality. Soil C, N, and enzymatic activities were all greater in the tree plantations than in the agricultural soil. As expected, the presence of N-fixing species had an impact on both the above-ground and below-ground compartments. As shown by the multivariate analysis, the non-native N-fixing *Elaeagnus umbellata* improved the growth of both *Populus alba* and *Juglans regia*, the target species, and supported a relatively high abundance of herbivorous nematodes. The native N-fixing species *Alnus cordata*, on the other hand, was associated with higher soil quality than *E. umbellata*. These findings suggest potential trade-offs to consider when selecting which tree species to use in mixture plantations, as some promote the growth of the target tree species while others promote soil chemical and biological quality. Nevertheless, our results support the use of mixed-tree plantations in the afforestation of former agricultural land both to restore soil quality and ensure an economic yield. We suggest that increasing tree growth and reducing the rotation length of the target species in the mixed plantation may be sufficient to offset the increased costs associated with planting and managing a mixed-species plantation.

Author Contributions: T.D. conceptualized the research design, was involved in the soil sampling and analyses, and led the writing of the manuscript. G.C. was involved in the soil sampling and analyses. G.G. was involved in the soil sampling and analyses. M.I. was involved in the soil sampling and analyses, and performed the multivariate statistical analyses. G.M. was involved in the soil sampling and analyses. F.N. was involved in the dendroecological sampling and analyses. F.P. was involved in the dendroecological sampling, contributed to field design and identification, and undertook the formal identification of the plant material used in the study. W.S.A. was involved in nematode analyses. G.B. was involved in the dendroecological sampling and analyses, and contributed to field design and identification. A.F. conceptualized the research design, and was involved in the soil sampling and analyses. All authors contributed to writing and revisions of the text. All authors have read and agreed to the published version of the manuscript.

Funding: This research received no external funding.

Data Availability Statement: The data presented in this study are available on request from the corresponding author.

Acknowledgments: We declare that the study protocol complies with relevant institutional, national, and international guidelines and legislation. Furthermore, we declare that we received formal permission from the tree farm owners, the Bini family, to collect *Populus alba* and *Juglans regia* tree cores. No voucher specimens have been deposited in a publicly available herbarium as the plants came from a tree nursery and were recognized on field by the same personnel who planted the trees.

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

References

1. van der Zanden, E.H.; Verburg, P.H.; Schulp, C.J.E.; Verkerk Johannes, P. Trade-offs of European agricultural abandonment. *Land Use Policy* **2017**, *62*, 290–301. [CrossRef]
2. Stoate, C.; Bàldi, A.; Beja, P.; Boatman, N.D.; Herzon, I.; van Doorn, A.; de Snoo, G.R.; Rakosy, L.; Ramwell, C. Ecological impacts of early 21st century agricultural change in Europe—A review. *J. Environ. Manag.* **2009**, *91*, 22–46. [CrossRef] [PubMed]
3. Keesstra, S.; Nunes, J.; Novara, A.; Finger, D.; Avelar, D.; Kalantari, Z.; Cerdà, A. The superior effect of nature based solutions in land management for enhancing ecosystem services. *Sci. Total Environ.* **2018**, *610–611*, 997–1009. [CrossRef] [PubMed]
4. van der Wal, A.; Van Veen, J.A.; Pijl, A.S.; Summerbell, R.C.; de Boer, W. Constraints on development of fungal biomass and decomposition processes during restoration of arable sandy soils. *Soil Biol. Biochem.* **2006**, *38*, 2890–2902. [CrossRef]
5. Benayas, M.R.; Martins, A.; Nicolau, M.; Schulz, J.J. Abandonment of agricultural land : An overview of drivers and consequences. *Perspect. Agric. Vet. Sci. Nutr. Nat. Resour.* **2007**, *2*, 1–14. [CrossRef]
6. Pardos, M.; del Río, M.; Pretzsch, H.; Jactel, H.; Bielak, K.; Bravo, F.; Brazaitis, G.; Defossez, E.; Engel, M.; Godvod, K. The greater resilience of mixed forests to drought mainly depends on their composition: Analysis along a climate gradient across Europe. *For. Ecol. Manag.* **2020**, *481*, 118687. [CrossRef]
7. Danise, T.; Innangi, M.; Curcio, E.; Fioretto, A.; Guggenberger, G. Fast Spectrophotometric Method as Alternative for CuO Oxidation to Assess Lignin in Soils with Different Tree Cover. *Forests* **2020**, *11*, 1262. [CrossRef]
8. Marron, N.; Epron, D. Are mixed-tree plantations including a nitrogen-fixing species more productive than monocultures? *For. Ecol. Manag.* **2019**, *441*, 242–252. [CrossRef]
9. Molina-Valero, J.A.; Camarero, J.J.; Álvarez-González, J.G.; Cerioni, M.; Hevia, A.; Sánchez-Salguero, R.; Martin-Benito, D.; Pérez-Cruzado, C. Mature forests hold maximum live biomass stocks. *For. Ecol. Manag.* **2021**, *480*, 118635. [CrossRef]
10. Liu, C.L.C.; Kuchma, O.; Krutovsky, K.V. Mixed-species versus monocultures in plantation forestry: Development, benefits, ecosystem services and perspectives for the future. *Glob. Ecol. Conserv.* **2018**, *15*, e00419. [CrossRef]
11. Battipaglia, G.; Pelleri, F.; Lombardi, F.; Altieri, S.; Vitone, A.; Conte, E.; Tognetti, R. Effects of associating *Quercus robur* L. and *Alnus cordata* Loisel. on plantation productivity and water use efficiency. *For. Ecol. Manag.* **2017**, *391*, 106–114. [CrossRef]
12. Kelty, M.J. The role of species mixtures in plantation forestry. *For. Ecol. Manag.* **2006**, *233*, 195–204. [CrossRef]
13. Chifflot, V.; Rivest, D.; Olivier, A.; Cogliastro, A.; Khasa, D. Molecular analysis of arbuscular mycorrhizal community structure and spores distribution in tree-based intercropping and forest systems. *Agric. Ecosyst. Environ.* **2009**, *131*, 32–39. [CrossRef]
14. Yang, G.; Wagg, C.; Veresoglou, S.D.; Hempel, S.; Rillig, M.C. How soil biota drive ecosystem stability. *Trends Plant. Sci.* **2018**, *23*, 1057–1067. [CrossRef]
15. Bardgett, R.D.; Cook, R.; Yeates, G.W.; Denton, C.S. The influence of nematodes on below-ground processes in grassland ecosystems. *Plant. Soil* **1999**, *212*, 23–33. [CrossRef]
16. Bardgett, R.D.; Van Der Putten, W.H. Belowground biodiversity and ecosystem functioning. *Nature* **2014**, *515*, 505–511. [CrossRef]
17. Bastida, F.; Zsolnay, A.; Hernández, T.; García, C. Past, present and future of soil quality indices: A biological perspective. *Geoderma* **2008**, *147*, 159–171. [CrossRef]
18. Masto, R.E.; Chhonkar, P.K.; Singh, D.; Patra, A.K. Alternative soil quality indices for evaluating the effect of intensive cropping, fertilisation and manuring for 31 years in the semi-arid soils of India. *Environ. Monit Assess.* **2008**, *136*, 419–435. [CrossRef]
19. van den Hoogen, J.; Geisen, S.; Routh, D.; Ferris, H.; Traunspurger, W.; Wardle, D.A.; De Goede, R.G.M.; Adams, B.J.; Ahmad, W.; Andriuzzi, W.S.; et al. Soil nematode abundance and functional group composition at a global scale. *Nature* **2019**, *572*, 194–198. [CrossRef]
20. Bongers, T.; Bongers, M. Functional diversity of nematodes. *Appl. Soil Ecol.* **1998**, *10*, 239–251. [CrossRef]
21. Ayuke, F.O.; Brussaard, L.; Vanlauwe, B.; Six, J.; Lelei, D.K.; Kibunja, C.N.; Pulleman, M.M. Soil fertility management : Impacts on soil macrofauna, soil aggregation and soil organic matter allocation. *Appl. Soil Ecol.* **2011**, *48*, 53–62. [CrossRef]
22. A'Bear, A.D.; Jones, T.H.; Boddy, L. Potential impacts of climate change on interactions among saprotrophic cord-forming fungal mycelia and grazing soil invertebrates. *Fungal Ecol.* **2014**, *10*, 34–43. [CrossRef]
23. van der Putten, W.H.; Bardgett, R.D.; Bever, J.D.; Bezemer, T.M.; Casper, B.B.; Fukami, T.; Kardol, P.; Klironomos, J.N.; Kulmatiski, A.; Schweitzer, J.A.; et al. Plant—Soil feedbacks : The past, the present and future challenges. *J. Ecol.* **2013**, *101*, 265–276. [CrossRef]

24. Zou, K.; Thébault, E.; Lacroix, G.; Barot, S. Interactions between the green and brown food web determine ecosystem functioning. *Funct. Ecol.* **2016**, *30*, 1454–1465. [CrossRef]
25. Mekonen, S.; Petros, I.; Hailemariam, M.; Conservation, B. The Role of Nematodes in the Processes of Soil Ecology and Their Use as Bioindicators. *Agric. Biol. J. N. Am.* **2017**, *8*, 132–140. [CrossRef]
26. Kulmatiski, A.; Beard, K.H.; Stevens, J.R.; Cobbold, S.M. Plant-soil feedbacks: A meta-analytical review. *Ecol. Lett.* **2008**, *11*, 980–992. [CrossRef]
27. Vila', M.; Espinar, J.L.; Hejda, M.; Hulme, P.E.; Jarosìk, V.; Maron, J.L.; Pergl, J.; Schaffner, U.; Sun, Y.; Pysek, P. Ecological impacts of invasive alien plants : A meta-analysis of their effects on species, communities and ecosystems. *Ecol. Lett.* **2011**, *14*, 702–708. [CrossRef]
28. Pelleri, F.; Ravagni, S.; Bianchetto, E.; Bidini, C. Comparing growth rate in a mixed plantation (walnut, poplar and nurse trees) with different planting designs: Results from an experimental plantation in northern Italy. *Ann. Silvic. Res.* **2013**, *37*, 13–21.
29. Cunningham, S.C.; Roxburgh, S.H.; Paul, K.I.; Patti, A.F.; Cavagnaro, T.R. Generating spatially and statistically representative maps of environmental variables to test the efficiency of alternative sampling protocols. *Agric. Ecosyst. Environ.* **2017**, *243*, 103–113. [CrossRef]
30. Pribyl, D.W. A critical review of the conventional SOC to SOM conversion factor. *Geoderma* **2010**, *156*, 75–83. [CrossRef]
31. MIPAF. Metodi di Analisi Chimica del Suolo. In *Collana di Metodi Analitici per L'agricoltura*; Sequi, P., Ed.; Franco Angeli: Milan, Italy, 2000.
32. Danise, T.; Innangi, M.; Curcio, E.; Fioretto, A. Covariation between plant biodiversity and soil systems in a European beech forest and a black pine plantation: The case of Mount Faito, (Campania, Southern Italy). *J. For. Res.* **2021**. [CrossRef]
33. Eivazi, F.; Tabatabai, M.A. Glucosidases and galactosidases in soils. *Soil Biol. Biochem.* **1988**, *20*, 601–606. [CrossRef]
34. Eivazi, F.; Tabatabai, M.A. Eivazi Phosphates in soils. *Soil Biol. Biochem.* **1977**, *9*, 167–172. [CrossRef]
35. Kandeler, E. Short-Term Assay of Soil Urease Activity Using Colorimetric Determination of Ammonium. *Soil Biol. Biochem.* **1988**, *6*, 68–72. [CrossRef]
36. Green, V.S.; Stottb, D.E.; Diacka, M. Assay for fluorescein diacetate hydrolytic activity: Optimization for soil samples. *Soil Biol. Biochem.* **2006**, *38*, 693–701. [CrossRef]
37. Puglisi, E.; Del Re, A.A.M.; Rao, M.A.; Gianfreda, L. Development and validation of numerical indexes integrating enzyme activities of soils. *Soil Biol. Biochem.* **2006**, *38*, 1673–1681. [CrossRef]
38. Yeates, G.W.; Bongers, T.; Goede, R.G.M.D.E.; Freckman, D.W.; Georgieva, S.S. Feeding Habits in Soil Nematode Families and Genera–An Outline for Soil Ecologists. *J. Nematol.* **1993**, *25*, 315–331.
39. Holmes, R.L. Computer-assisted quality control in tree-ring dating and measurement. *Tree-Ring Bull.* **1983**, *43*, 51–67.
40. Barker, M.; Rayens, W. Partial least squares for discrimination. *J. Chemom.* **2003**, *17*, 166–173. [CrossRef]
41. Carrascal, L.M.; Galván, I.; Gordo, O. Partial least squares regression as an alternative to current regression methods used in ecology. *Oikos* **2009**, *118*, 681–690. [CrossRef]
42. Innangi, M.; Balestrieri, R.; Danise, T.; d'Alessandro, F.; Fioretto, A. From soil to bird community: A Partial Least Squares approach to investigate a natural wooded area surrounded by urban patchwork (Astroni crater, southern Italy). *Ecol. Modell.* **2019**, *394*, 1–10. [CrossRef]
43. Innangi, M.; Danise, T.; d'Alessandro, F.; Curcio, E.; Fioretto, A. Dynamics of Organic Matter in Leaf Litter and Topsoil within an Italian Alder (*Alnus cordata* (Loisel.) Desf.) Ecosystem. *Forests* **2017**, *8*, 240. [CrossRef]
44. Team, R.C. A Language and Environment for Statistical Computing. R Foundation for Statistical Computing. 2019. Available online: https://www.r-project.org/ (accessed on 5 March 2020).
45. Hong, S.; Piao, S.; Chen, A.; Liu, Y.; Liu, L.; Peng, S.; Sardans, J.; Sun, Y.; Peñuelas, J.; Zeng, H. Afforestation neutralizes soil pH. *Nat. Commun.* **2018**, *9*, 520. [CrossRef]
46. Laganiere, J.; Angers, D.A.; Pare, D. Carbon accumulation in agricultural soils after afforestation : A meta-analysis. *Glob. Chang. Biol.* **2010**, *16*, 439–453. [CrossRef]
47. Berthrong, S.T.; Jobbágy, E.G.; Jackson, R.B. A global meta-analysis of soil exchangeable cations, pH, carbon, and nitrogen with afforestation. *Ecol. Appl.* **2009**, *19*, 2228–2241. [CrossRef]
48. Liming, J. The Review of Mixtures of Nitrogen fixing and Non nitrogen fixing Tree Species. *World For. Res.* **1998**, *11*, 20–26.
49. Ramesh, T.; Bolan, N.S.; Kirkham, M.B.; Wijesekara, H.; Kanchikerimath, M.; Rao, C.S.; Sandeep, S.; Rinklebe, J.; Ok, Y.S.; Choudhury, B.U. Soil organic carbon dynamics: Impact of land use changes and management practices: A review. In *Advances in Agronomy*; Elsevier: Amsterdam, The Netherlands, 2019; Volume 156, pp. 1–107. ISBN 0065-2113.
50. Kooch, Y.; Rostayee, F.; Hosseini, S.M. Effects of tree species on topsoil properties and nitrogen cycling in natural forest and tree plantations of northern Iran. *Catena* **2016**, *144*, 65–73. [CrossRef]
51. Li, X.G.; Li, Y.K.; Li, F.M.; Ma, Q.; Zhang, P.L.; Yin, P. Changes in soil organic carbon, nutrients and aggregation after conversion of native desert soil into irrigated arable land. *Soil Tillage Res.* **2009**, *104*, 263–269. [CrossRef]
52. Le Noë, J.; Matej, S.; Magerl, A.; Bhan, M.; Erb, K.; Gingrich, S. Modeling and empirical validation of long-term carbon sequestration in forests (France, 1850–2015). *Glob. Chang. Biol.* **2020**, *26*, 2421–2434. [CrossRef]
53. Wei, X.; Shao, M.; Gale, W.; Li, L. Global pattern of soil carbon losses due to the conversion of forests to agricultural land. *Sci. Rep.* **2014**, *4*, 6–11. [CrossRef]

54. Chiti, T.; Certini, G.; Puglisi, A.; Sanesi, G.; Capperucci, A.; Forte, C. Effects of associating a N-fixer species to monotypic oak plantations on the quantity and quality of organic matter in minesoils. *Geoderma* **2007**, *138*, 162–169. [CrossRef]
55. Chodak, M.; Niklinska, M. The effect of different tree species on the chemical and microbial properties of reclaimed mine soils. *Biol Fertil Soils* **2010**, *46*, 555–566. [CrossRef]
56. Parsapour, M.K.; Kooch, Y.; Hosseini, S.M.; Alavi, S.J. Litter and topsoil in *Alnus subcordata* plantation on former degraded natural forest land : A synthesis of age-sequence. *Soil Tillage Res.* **2018**, *179*, 1–10. [CrossRef]
57. Johnson, D.W.; Curtis, P.S. Effects of forest management on soil C and N storage: Meta analysis. *For. Ecol. Manag.* **2001**, *140*, 227–238. [CrossRef]
58. Shi, S.; Han, P.; Zhang, P.; Ding, F.; Ma, C. The impact of afforestation on soil organic carbon sequestration on the Qinghai Plateau, China. *PLoS ONE* **2015**, *10*, e0116591. [CrossRef]
59. Cotrufo, M.F.; Wallenstein, M.D.; Boot, C.M.; Denef, K.; Paul, E. The Microbial Efficiency-Matrix Stabilization (MEMS) framework integrates plant litter decomposition with soil organic matter stabilization: Do labile plant inputs form stable soil organic matter? *Glob. Chang. Biol.* **2013**, *19*, 988–995. [CrossRef]
60. Zhang, H.; Xiong, X.; Wu, J.; Zhao, J.; Zhao, M.; Chu, G.; Hui, D.; Zhou, G.; Deng, Q.; Zhang, D. Changes in Soil Microbial Biomass, Community Composition, and Enzyme Activities After Half-Century Forest Restoration in Degraded Tropical Lands. *Forests* **2019**, *10*, 1124. [CrossRef]
61. Allison, S.D.; Vitousek, P.M. Responses of extracellular enzymes to simple and complex nutrient inputs. *Soil Biol. Biochem.* **2005**, *37*, 937–944. [CrossRef]
62. Boyle, S.A.; Yarwood, R.R.; Bottomley, P.J.; Myrold, D.D. Bacterial and fungal contributions to soil nitrogen cycling under Douglas fir and red alder at two sites in Oregon. *Soil Biol. Biochem.* **2008**, *40*, 443–451. [CrossRef]
63. Bini, D.; Dos Santos, C.A.; Bouillet, J.-P.; de Morais Gonçalves, J.L.; Cardoso, E.J.B.N. *Eucalyptus grandis* and *Acacia mangium* in monoculture and intercropped plantations: Evolution of soil and litter microbial and chemical attributes during early stages of plant development. *Appl. Soil Ecol.* **2013**, *63*, 57–66. [CrossRef]
64. Tan, X.; Chang, S.X.; Kabzems, R. Soil compaction and forest floor removal reduced microbial biomass and enzyme activities in a boreal aspen forest soil. *Biol. Fertil. Soils* **2008**, *44*, 471–479. [CrossRef]
65. Raiesi, F.; Kabiri, V. Identification of soil quality indicators for assessing the effect of different tillage practices through a soil quality index in a semi-arid environment. *Ecol. Indic.* **2016**, *71*, 198–207. [CrossRef]
66. Lange, M.; Eisenhauer, N.; Sierra, C.A.; Bessler, H.; Engels, C.; Griffiths, R.I.; Mellado-Vázquez, P.G.; Malik, A.A.; Roy, J.; Scheu, S.; et al. Plant diversity increases soil microbial activity and soil carbon storage. *Nat. Commun.* **2015**, *6* (Suppl. 6707). [CrossRef]
67. Neher, D.A. Ecology of plant and free-living nematodes in natural and agricultural soil. *Annu. Rev. Phytopathol.* **2010**, *48*, 371–394. [CrossRef]
68. Bjørnlund, L.; Liu, M.; Rønn, R.; Christensen, S.; Ekelund, F. Nematodes and protozoa affect plants differently, depending on soil nutrient status. *Eur. J. Soil Biol.* **2012**, *50*, 28–31. [CrossRef]
69. Yeates, G.W.; Bongers, T. Nematode diversity in agroecosystems. *Agric. Ecosyst. Environ.* **1999**, *74*, 113–135. [CrossRef]
70. Armendáriz, I.; Arpin, P. Nematodes and their relationship to forest dynamics: I. Species and trophic groups. *Biol. Fertil. soils* **1996**, *23*, 405–413. [CrossRef]
71. Urzelai, A.; Hernandez, J.A.; Pastor, J. Biotic indices based on soil nematode communities for assessing soil quality in terrestrial ecosystems. *Sci. Total Environ.* **2000**, *247*, 253–261. [CrossRef]
72. Dupont, S.T.; Ferris, H.; Horn, M. Van Effects of cover crop quality and quantity on nematode-based soil food webs and nutrient cycling. *Appl. Soil Ecol.* **2009**, *41*, 157–167. [CrossRef]
73. Babst, F.; Alexander, M.R.; Szejner, P.; Bouriaud, O.; Klesse, S.; Roden, J.; Ciais, P.; Poulter, B.; Frank, D.; Moore, D.J.P. A tree-ring perspective on the terrestrial carbon cycle. *Oecologia* **2014**, *176*, 307–322. [CrossRef]
74. de Paula, M.D.; Groeneveld, J.; Huth, A. The extent of edge effects in fragmented landscapes: Insights from satellite measurements of tree cover. *Ecol. Indic.* **2016**, *69*, 196–204. [CrossRef]
75. Jingjing, L.; Crowther, T.W.; Picard, N.; Wiser, S.; Mo, Z.; Alberti, G.; Schulze, E.D.; McGuire, A.D.; Bozzato, F.; Pretzsch, H. Positive biodiversity-productivity relationship predominant in global forests. *Science* **2021**, *354*, 196.
76. Forrester, D.I.; Bauhus, J.; Cowie, A.L.; Vanclay, J.K. Mixed-species plantations of *Eucalyptus* with nitrogen-fixing trees: A review. *For. Ecol. Manag.* **2006**, *233*, 211–230. [CrossRef]
77. Guerrieri, R.; Mencuccini, M.; Sheppard, L.J.; Saurer, M.; Perks, M.P.; Levy, P.; Sutton, M.A.; Borghetti, M.; Grace, J. The legacy of enhanced N and S deposition as revealed by the combined analysis of δ13C, δ18O and δ15N in tree rings. *Glob. Chang. Biol.* **2011**, *17*, 1946–1962. [CrossRef]
78. Khamzina, A.; Lamers, J.P.A.; Vlek, P.L.G. Nitrogen fixation by *Elaeagnus angustifolia* in the reclamation of degraded croplands of Central Asia. *Tree Physiol.* **2009**, *29*, 799–808. [CrossRef]
79. Paschke, M.W.; Dawson, J.O.; David, M.B. Soil nitrogen mineralization in plantations of *Juglans nigra* interplanted with actinorhizal *Elaeagnus umbellata* or *Alnus glutinosa*. *Plant Soil* **1989**, *118*, 33–42. [CrossRef]
80. Hoogmoed, M.; Cunningham, S.C.; Baker, P.; Beringer, J.; Cavagnaro, T.R. N-fixing trees in restoration plantings: Effects on nitrogen supply and soil microbial communities. *Soil Biol. Biochem.* **2014**, *77*, 203–212. [CrossRef]
81. Ehrenfeld, J.G. Ecosystem consequences of biological invasions. *Annu. Rev. Ecol. Evol. Syst.* **2010**, *41*, 59–80. [CrossRef]

82. Ge, Y.; Wang, Q.; Wang, L.; Liu, W.; Liu, X.; Huang, Y.; Christie, P. Response of soil enzymes and microbial communities to root extracts of the alien *Alternanthera philoxeroides*. *Arch. Agron. Soil Sci.* **2018**, *64*, 708–717. [CrossRef]
83. Liang, J.; Reynolds, T.; Wassie, A.; Collins, C.; Wubalem, A. Effects of exotic *Eucalyptus* spp. plantations on soil properties in and around sacred natural sites in the northern Ethiopian Highlands. *AIMS Agric. Food* **2016**, *1*, 175–193. [CrossRef]