

RESEARCH ARTICLE

# Impact of understory vegetation on soil carbon and nitrogen dynamic in aerially seeded *Pinus massoniana* plantations

Ping Pan<sup>1</sup>, Fang Zhao<sup>2</sup>, Jinkui Ning<sup>1</sup>, Ling Zhang<sup>1\*</sup>, Xunzhi Ouyang<sup>1\*</sup>, Hao Zang<sup>1</sup>

**1** College of Forestry, Jiangxi Agricultural University, Nanchang, China, **2** College of Tourism and Territorial Resources, Jiujiang University, Jiujiang, China

\* [lingzhang09@126.com](mailto:lingzhang09@126.com), [lingzhang@jxau.edu.cn](mailto:lingzhang@jxau.edu.cn) (LZ); [oyxz\\_2003@hotmail.com](mailto:oyxz_2003@hotmail.com) (XO)



**OPEN ACCESS**

**Citation:** Pan P, Zhao F, Ning J, Zhang L, Ouyang X, Zang H (2018) Impact of understory vegetation on soil carbon and nitrogen dynamic in aerially seeded *Pinus massoniana* plantations. PLoS ONE 13(1): e0191952. <https://doi.org/10.1371/journal.pone.0191952>

**Editor:** Jorge Paz-Ferreiro, RMIT University, AUSTRALIA

**Received:** March 21, 2017

**Accepted:** January 15, 2018

**Published:** January 29, 2018

**Copyright:** © 2018 Pan et al. This is an open access article distributed under the terms of the [Creative Commons Attribution License](https://creativecommons.org/licenses/by/4.0/), which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited.

**Data Availability Statement:** All relevant data are within the paper.

**Funding:** This study was financially supported by the National Natural Science Foundation of China (No. 31360181 and 31160159) and Jiangxi Agricultural University (No. 9232306047). The funders had no role in study design, data collection and analysis, decision to publish, or preparation of the manuscript.

**Competing interests:** The authors have declared that no competing interests exist.

## Abstract

Understory vegetation plays a vital role in regulating soil carbon (C) and nitrogen (N) characteristics due to differences in plant functional traits. Different understory vegetation types have been reported following aerial seeding. While aerial seeding is common in areas with serious soil erosion, few studies have been conducted to investigate changes in soil C and N cycling as affected by understory vegetation in aerially seeded plantations. Here, we studied soil C and N characteristics under two naturally formed understory vegetation types (*Dicranopteris* and graminoid) in aerially seeded *Pinus massoniana* Lamb plantations. Across the two studied understory vegetation types, soil organic C was significantly correlated with all measured soil N variables, including total N, available N, microbial biomass N and water-soluble organic N, while microbial biomass C was correlated with all measured variables except soil organic C. *Dicranopteris* and graminoid differed in their effects on soil C and N process. Except water-soluble organic C, all the other C and N variables were higher in soils with graminoids. The higher levels of soil organic C, microbial biomass C, total N, available N, microbial biomass N and water-soluble organic N were consistent with the higher litter and root quality (C/N) of graminoid vegetation compared to *Dicranopteris*. Changes in soil C and N cycles might be impacted by understory vegetation types via differences in litter or root quality.

## Introduction

Soil carbon (C) and nitrogen (N) play important role in impacting soil capacity to maintain biological productivity [1, 2] and regulating atmospheric C and N compositions [3–5]. Among soil C and N components, soil microbial biomass C (MBC) and N (MBN) are labile constituents in soil organic C (SOC) and total N (TN) and indicators of changes in soil C and N pools [6]. In addition, both soil water-soluble organic C (WSOC) and N (WSON) are sensitive to land use, forest management, and habitat disturbance. These labile constituents of soil organic matter are considered to be important indicators of soil capacity in maintaining biological productivity [7, 8].

In forest ecosystems, understory vegetation can be an important factor impacting soil C and N processes [9] as has been documented by previous studies [10, 11]. While understory

vegetation represents only a small portion of forest biomass, it plays an important role in maintaining biodiversity, ecosystem stability, and sustainable productivity of forest ecosystems [12]. However, the dependence of soil C and N processes on understory vegetation types are not thoroughly understood [13]. Studies have shown that understory vegetation types impact soil temperature and moisture [14], microbial richness and composition [15–18], and C/N ratios [11, 13, 19]. For example, Wu et al. [20] reported that removing understory vegetation from *Eucalyptus* forest decreased root biomass and soil organic matter input, and hence altered soil microbial community structure. Similarly, Wan et al. [21] evaluated the effect of the understory vegetation (*Dicranopteris dishotoma*) on the growth of subtropical forest canopy vegetation (*Eucalyptus*) and found decreased soil moisture, soil organic matter, and soil pH after understory vegetation was removed. So, it is critical to understand the effects of understory vegetation on soil C and N cycling, and further, potential effects on changes in atmospheric C and N compositions [22, 23], especially in areas with intensive reforestation and management of understory vegetation.

In the mid-20<sup>th</sup> century, the southern part of Jiangxi Province, China, was a region with large-scale intensive soil erosion and increased runoff, especially the Xing'guo County area. To control runoff and soil erosion and restore forest vegetation, aerial seeding of *P. massoniana* has been widely implemented since the 1970s [24]. Aerial seeding plantations characterized by faster restoration played important role in improving the ecological environment of the region. Considering the importance of understory vegetation in aerially seeded plantations, their effects on soil element cycling process could not be ignored. In the process of forest development, two understory vegetation types (*Dicranopteris* and graminoid) have formed. As the root system of *Dicranopteris* consists of clustering rhizomes, it can grow on the surface of soil through horizontal spread, effectively reducing water loss and mitigating soil erosion [25]. The root systems of graminoids differ from those of *Dicranopteris*. In addition, the dominant graminoid species *Paspalum thunbergii* may have symbiotic relationship with N-fixing soil bacteria as its congeners, which will potentially impact litter C and N characteristics and soil C and N cycling [26]. However, the effect of understory vegetation on soil C and N cycling process in aerially seeded plantations has not been investigated.

Here, we conducted a study in Xing'guo County to understand how soil C and N cycling vary in aerially seeded *P. massoniana* plantations [24] that differ in understory vegetation types to investigate the following questions: 1) How are soil C and N components related to each other in aerially seeded *P. massoniana* plantations that differ in understory vegetation? 2) How do vegetation functional characteristics vary between *Dicranopteris* and graminoid dominated understories? 3) How do soil C and N characteristics vary among plantations with understories dominated by *Dicranopteris* vs. graminoids? The results will provide a scientific reference for the management of understory vegetation in aerially seeded *P. massoniana* plantations.

## Materials and methods

### Study area

Xing'guo County (115°01'~115°51' E, 26°03'~26°42' N) located in the central southern part of Jiangxi Province, subtropical China, is characterized by a mid-subtropical warm and humid climate. The mean annual temperature is 18.9°C and annual precipitation is 1,539 mm, with a distinct wet season from April to June [27]. The frost-free period lasts for 280–300 days. Soils are classified as Udic Ferralsols developed from granite weathering [28]. This area is rich in forest resources. Presently, the main forests here are evergreen broadleaf forests, *P. massoniana* forests, and *Cunninghamia lanceolata* plantations [27, 29]. The area of aerially seeded *P.*

*massoniana* plantations is 64,000 ha, accounting for 29.5% of the county's forest land area [27]. These plantations have understories dominated by either *Dicranopteris linearis* or *Paspalum thunbergii*, respectively, which were formed 10–15 years later after *P. massoniana* were sowed. The dominated proportion of *Dicranopteris* and graminoid vegetation were more than 90% and 85%, respectively.

### Plot establishment and soil sampling

In August 2012, a reconnaissance survey was conducted in aerially seeded *P. massoniana* plantations. The plantations selected for this study had not been disturbed by anthropogenic activities since aerially seeding during 1986 and 1991, and had understories dominated by *Dicranopteris* or graminoid species (Fig 1). The slope faced south with an intermediate slope (S1 Table). The understory had a density of 60–80% cover. Plots were established paired and randomly in *P. massoniana* plantations with understory vegetation dominated by *Dicranopteris* or graminoid species (S1 Table; Fig 1). The plot size was 400 m<sup>2</sup> (20 × 20 m), with nine replications for each understory vegetation type. No permits were required.

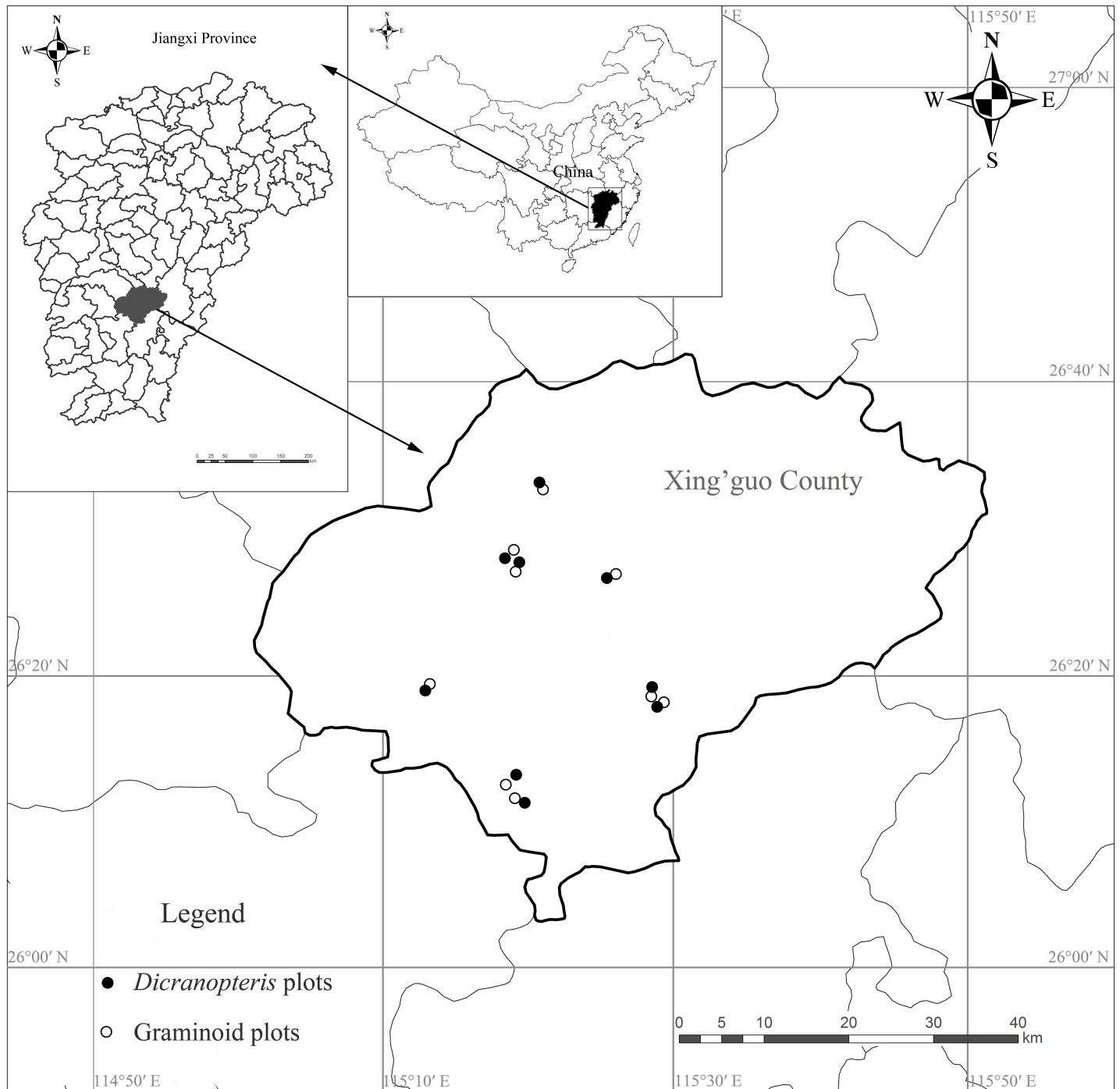
Soil samples were collected from the top 20 cm soil layer from three sampling points at the upper, middle, and lower part of each plot using a soil auger (6 cm in diameter). Soil samples of each plot were mixed together to obtain one composite sample. Approximately 1 kg of the mixed sample was then divided into two portions. One portion was preserved at a temperature of 4°C for the measurement of MBC, MBN, available N (AN), WSOC and WSON. The other portion was air-dried and processed for the determination of SOC and TN.

### Understory vegetation litter, root and biomass collection

Three randomly distributed subplots (1 m × 1 m) were established for collection of litter, root and total biomass in each plot. In July 2012, plant litter and above- and below-ground biomass were each collected by vegetation types. Root collection was performed by excavating soils within the subplots. Specifically, subplot soil within the 0–30 cm soil layer was all collected and passed through 0.5 mm sieve to obtain all fine roots [30]. Soil attached to roots was removed by washing. Plant samples were taken back to the lab and dried to obtain dry biomass by plots. Total biomass (TB) was obtained by summing above- and below-ground biomass. Root to shoot ratio (RSR) was calculated based on dry weight of below- and above-ground biomass. Subsamples of litter and root were processed for determination of C and N concentrations [31].

### Plant and soil C and N measurements

Soil organic C and plant sample C were measured by the potassium dichromate oxidation-external heating method [32]. Carbon oxidation occurred in potassium dichromate solution and C content was obtained by subsequent titration. Total N was digested by concentrated sulfuric acid and determined by the Kjeldahl method [32]. Carbon to N ratio (C/N) was calculated based on C and N concentrations. Soil NH<sub>4</sub><sup>+</sup> and NO<sub>3</sub><sup>-</sup> (AN) were extracted by 2 mol L<sup>-1</sup> potassium chloride solution produced by deionized water and determined by the colorimetric method [8]. Microbial biomass C and MBN were measured by the chloroform fumigation—potassium sulfate extraction method [33, 34]. After extraction, MBC and MBN were measured with a TOC-1020A analyzer (Elementar, Germany). The TOC-1020A analyzer was also used for measurement of WSOC after extraction with deionized water (10 g soil with 20 ml water, shaking at 25 °C for 15 min) and filtration using 0.45 μm polytetrafluoroethylene filters [35]. Water-soluble organic N was obtained by calculating the difference between total soluble N and inorganic N.



**Fig 1. Sampling locations of the study area (Xing'guo County, Jiangxi province, China).**

<https://doi.org/10.1371/journal.pone.0191952.g001>

### Statistical analyses

We conducted pairwise correlation analysis to examine the correlations among soil C and N properties both across and within understory vegetation types to examine differences in these correlations induced by understory vegetation types. We performed principal component analysis (PCA) to understand the overall pattern of soil C and N variation among plots. We

**Table 1. Differences in functional traits (mean ± 1 s.e.) between two understory vegetation types.** Significant results of paired *t* tests are in bold. Root C/N, root C to N ratio; Litter C/N, litter C to N ratio. TB, total biomass, g m<sup>-2</sup>; RSR, root to shoot ratio.

Variables	<i>Dicranopteris</i>	Graminoid	<i>t</i> ratio	<i>P</i>
Root C/N	<b>36.02±0.49</b>	<b>33.43±0.42</b>	<b>-5.27</b>	<b>0.0008</b>
Litter C/N	<b>40.65±0.48</b>	<b>33.55±0.39</b>	<b>-7.33</b>	<b>&lt;0.0001</b>
TB	250.10±4.69	240.28±4.29	<b>-2.96</b>	<b>0.0180</b>
RSR	<b>1.10±0.01</b>	<b>1.04±0.02</b>	<b>-2.35</b>	<b>0.0468</b>

<https://doi.org/10.1371/journal.pone.0191952.t001>

conducted paired *t* tests to examine differences in soil C and N characteristics between forests with two different kinds of understory vegetation types. No transformations were conducted since data met the assumptions of ANOVA. Statistical analyses were conducted in JMP 9.0 (SAS Institute, Cary, NC, USA).

## Results

### Differences in functional traits between two understory vegetation types

Both litter and roots produced by graminoid vegetation had lower C/N relative to that produced by *Dicranopteris* (Table 1). In addition, *Dicranopteris* was higher in TB and RSR compared to graminoid (Table 1).

### Soil C and N correlations across understory vegetation types

Microbial biomass C was not correlated with SOC (Tables 2 and 3) but was only correlated with WSOC (Table 3). The PCA plot showed that, except WSOC, all the other variables contributed to the first principal component (Fig 2). The first principal component based on all

**Table 2. Soil C and N variables (means ± 1 se) in plots with different understory vegetation types.** SOC, soil organic carbon, g kg<sup>-1</sup>; MBC, microbial biomass carbon, mg kg<sup>-1</sup>; WSOC, water-soluble organic carbon, mg kg<sup>-1</sup>; TN, total nitrogen, mg kg<sup>-1</sup>; AN, available nitrogen, mg kg<sup>-1</sup>; MBN, microbial biomass nitrogen, mg kg<sup>-1</sup>; WSON, water-soluble organic nitrogen, mg kg<sup>-1</sup>. Statistical results are shown in Figs 3 and 4.

Type	Plot	SOC	MBC	WSOC	TN	AN	MBN	WSON
<i>Dicranopteris</i>	1	7.97±0.67	146.20±3.54	219.21±1.22	79.55±4.66	16.33±1.55	23.50±1.62	25.85±1.56
	3	10.21±1.61	190.22±5.23	300.38±3.08	84.41±2.03	22.01±1.27	33.11±1.35	40.25±1.75
	5	7.54±1.05	97.40±2.58	218.56±4.04	80.20±4.24	15.12±1.21	19.66±1.82	28.70±3.03
	7	2.55±0.54	259.38±4.62	234.80±3.38	75.79±3.28	15.38±1.10	24.27±1.71	20.53±1.78
	9	5.33±0.59	189.29±4.53	249.73±5.63	77.98±3.60	14.75±0.91	28.88±2.76	28.60±1.13
	11	5.13±0.06	96.42±3.31	147.14±5.00	73.69±2.11	15.00±1.08	17.35±1.85	32.81±1.24
	13	8.65±0.47	103.13±4.59	190.25±3.62	75.48±3.44	14.74±0.80	24.27±2.29	23.20±1.22
	15	4.42±0.16	179.82±2.40	226.36±3.43	72.10±1.16	10.32±1.19	22.74±1.28	37.15±1.56
	17	3.42±0.06	241.67±1.73	227.66±1.23	71.44±1.52	16.32±1.27	25.02±1.88	42.28±1.20
Graminoid	2	9.17±0.64	204.61±3.32	229.80±3.84	82.01±1.15	26.52±1.81	41.62±2.01	47.67±2.15
	4	16.84±1.00	267.90±4.04	243.64±1.40	96.45±2.83	53.83±1.39	67.74±0.94	63.23±1.70
	6	6.53±0.83	173.10±5.08	209.62±4.49	76.66±2.93	20.44±2.53	52.31±0.99	43.22±1.78
	8	9.27±1.21	223.56±5.81	257.60±3.84	87.65±2.72	29.79±0.47	40.79±1.30	43.03±1.32
	10	4.28±0.50	165.94±5.35	253.38±5.05	72.09±1.91	15.42±2.78	26.58±2.41	20.59±2.19
	12	6.62±0.92	177.52±3.62	234.42±3.83	86.80±2.32	17.36±2.28	32.72±1.85	38.00±1.23
	14	13.35±0.62	337.33±11.91	257.07±5.30	89.13±3.45	32.72±1.56	57.69±3.44	62.38±2.27
	16	8.29±0.63	199.81±1.85	244.29±2.00	87.89±2.07	15.46±2.33	19.67±3.98	40.72±2.16
	18	13.36±1.53	317.08±2.54	248.63±7.30	95.22±2.59	36.94±2.22	53.15±1.54	48.05±2.81

<https://doi.org/10.1371/journal.pone.0191952.t002>

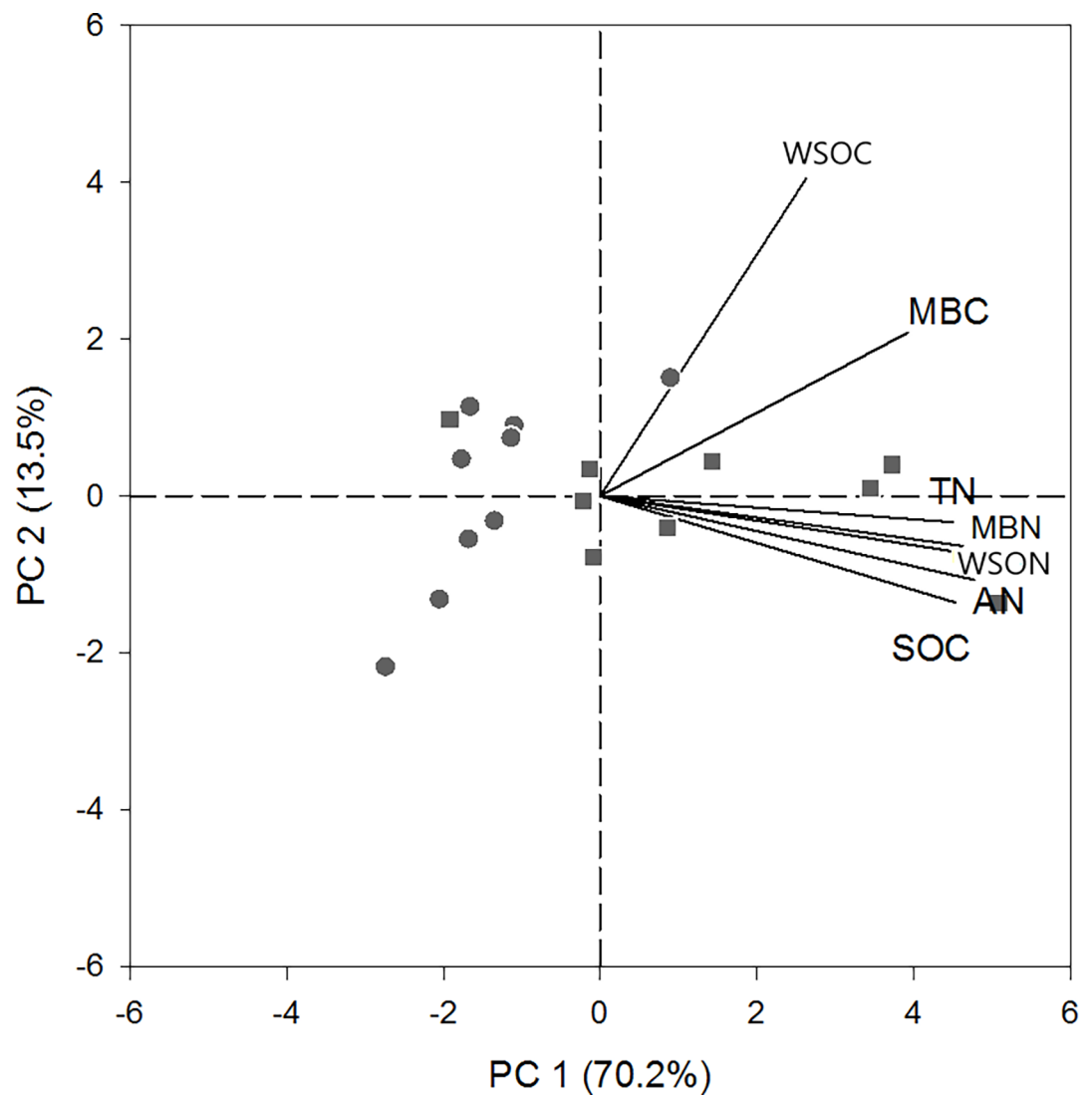
**Table 3. Pairwise correlation coefficients (R) of soil carbon and nitrogen variables in the studied plots.** SOC, soil organic carbon, g kg<sup>-1</sup>; MBC, microbial biomass carbon, mg kg<sup>-1</sup>; WSOC, water-soluble organic carbon, mg kg<sup>-1</sup>; TN, total nitrogen, mg kg<sup>-1</sup>; AN, available nitrogen, mg kg<sup>-1</sup>; MBN, microbial biomass nitrogen, mg kg<sup>-1</sup>; WSON, water-soluble organic nitrogen, mg kg<sup>-1</sup>.

Variables	MBC	WSOC	TN	AN	MBN	WSON
SOC	0.47	0.33	0.88**	0.88**	0.77**	0.75**
MBC		0.57*	0.57*	0.64**	0.68**	0.66**
WSOC			0.45	0.32	0.35	0.31
TN				0.81**	0.69**	0.70**
AN					0.89**	0.79**
MBN						0.81**

\*, P<0.05

\*\*, P<0.01.

<https://doi.org/10.1371/journal.pone.0191952.t003>



**Fig 2. Principal components analysis (PCA) of C and N variables based on measurements from all plots.** Filled circles, *Dicranopteris*; filled squares, graminoids.

<https://doi.org/10.1371/journal.pone.0191952.g002>

**Table 4. Pairwise correlation coefficients (R) of soil carbon and nitrogen variables in plots dominated by *Dicranopteris* or graminoids.** SOC, soil organic carbon, g kg<sup>-1</sup>; MBC, microbial biomass carbon, mg kg<sup>-1</sup>; WSOC, water-soluble organic carbon, mg kg<sup>-1</sup>; TN, total nitrogen, mg kg<sup>-1</sup>; AN, available nitrogen, mg kg<sup>-1</sup>; MBN, microbial biomass nitrogen, mg kg<sup>-1</sup>; WSON, water-soluble organic nitrogen, mg kg<sup>-1</sup>.

Vegetation type	Variables	MBC	WSOC	TN	AN	MBN	WSON
<i>Dicranopteris</i>	SOC	-0.56	0.26	0.76*	0.55	0.33	0.01
	MBC		0.56	-0.17	0.14	0.52	0.20
	WSOC			0.58	0.52	0.88**	0.34
	TN				0.72*	0.51	-0.11
	AN					0.60	0.23
	MBN						0.25
Graminoid	SOC	0.85**	0.34	0.86**	0.94**	0.79*	0.90**
	MBC		0.58	0.73*	0.70*	0.66	0.76*
	WSOC			0.35	0.27	0.01	0.11
	TN				0.74*	0.49	0.73*
	AN					0.86**	0.79*
	MBN						0.81**

\*,  $P < 0.05$

\*\*,  $P < 0.01$ .

<https://doi.org/10.1371/journal.pone.0191952.t004>

variables explained 70.2% of the variations among all plots (Fig 2). The cumulative percentage of the first two components was 83.7% (Fig 2).

### Understory vegetation effects on soil C and N characteristics

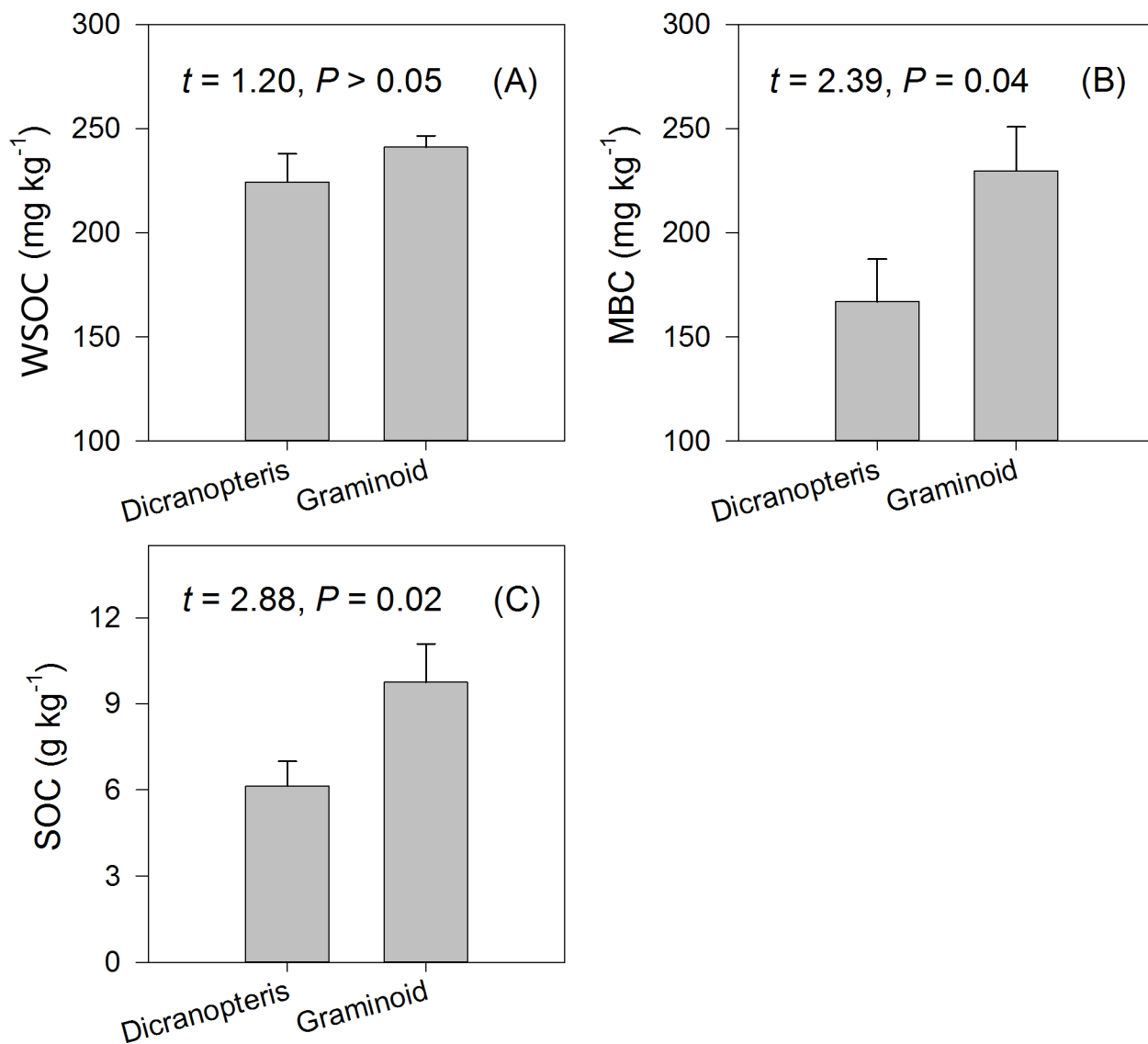
Correlation analysis of soil C and N variables under the two different vegetation types showed that both SOC and AN were correlated significantly with TN while WSOC was correlated with MBN for *Dicranopteris* plots (Table 4). For plots dominated by graminoid species, SOC, MBC, TN, AN, and WSON were correlated with each other while MBN was correlated with SOC, AN, and WSON (Table 4).

Except for WSOC ( $t = 1.20, P > 0.05$ ), all the measured soil C and N variables were significantly higher in soils in plots dominated by graminoid species compared to those in plots dominated by *Dicranopteris* (Figs 3 and 4). Specifically, SOC, MBC, AN, MBN, and WSON were 58.8%, 37.5%, 77.5%, 79.3%, and 45.6% higher, respectively, in graminoid plots compared to *Dicranopteris* plots (Figs 3 and 4).

## Discussion

### Correlations between WSOC and MBC differ across and within understory vegetation types

Except WSOC, most of the variables correlated with each other across understory vegetation types [36]. Water soluble organic C has been commonly used as an indicator for microbial activities as it is readily available for microbes [37–39], which is consistent with the significant correlation between WSOC and MBC in this study (Table 3). When analyzed separately by understory vegetation, no correlations were observed between WSOC and MBC (Table 4). Although the study area had 18 plots, all plots were characterized by similar slope, tree diameter, canopy density and stand density (S1 Table), suggesting different effects of understory vegetation on MBC or WSOC. In addition, results of the PCA showed that WSOC was the only variable correlated positively with the second principal component, while all other variables positively correlated with the first component (Fig 2). Based on the spatial distribution of the



**Fig 3. Dependence of soil C variables on understory vegetation types.** Results of paired  $t$  tests are shown.

<https://doi.org/10.1371/journal.pone.0191952.g003>

data points with different treatments as well as MBC and WSOC, the results further indicated labile soil C variable (WSOC or MBC) was differently altered across understory vegetation types (Fig 3).

### Soil MBC and N variables increased by graminoids vegetation

Understory vegetation participates in the nutrient cycling of soil C and N process through several different ways. The graminoid vegetation has a fibrous root system, with higher decomposition rates compared with the rhizomes of *Dicranopteris*, which would potentially increase soil organic matter inputs [40, 41]. Additionally, due to differences in functional traits, understory vegetation types might alter litter decomposition rates and microbial activities by affecting soil moisture, temperature, and other environmental factors [42, 43]. Changes in the litter decomposition environment might alter litter decomposition rate and hence C and N releasing rate [44, 45]. Moreover, litter produced by different plant species or genotypes might differ in



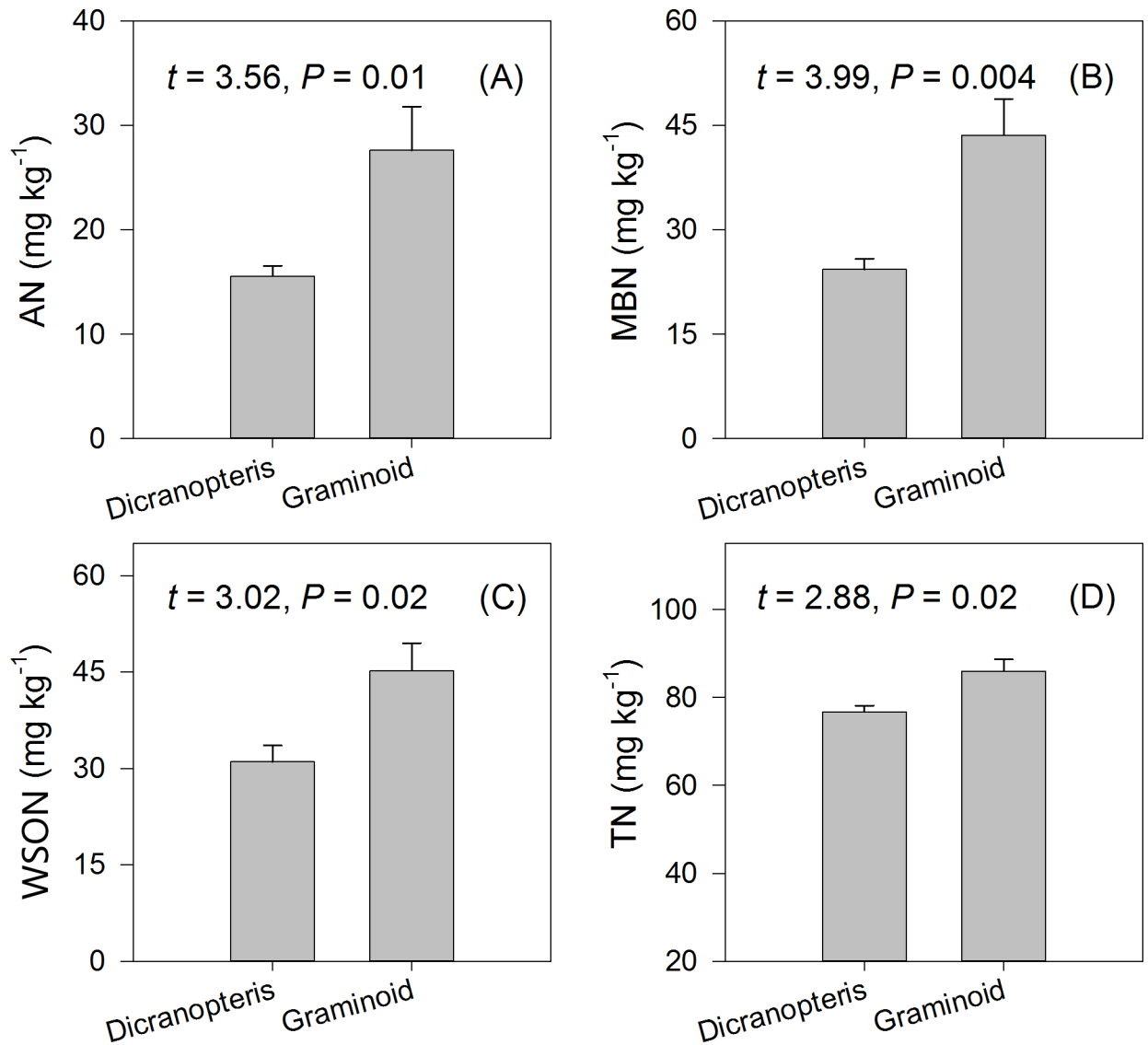


Fig 4. Dependence of soil N variables on understory vegetation types. Results of paired *t* tests are shown.

<https://doi.org/10.1371/journal.pone.0191952.g004>

litter quality (e.g., lower C/N or higher N concentration) [46]. Both litter and root C/N of graminoids were significantly lower relative to *Dicranopteris* (Table 1). Higher litter quality (e.g., lower C to N ratio) might be associated with faster litter decomposition and hence faster nutrient releasing rate [44, 46], indicating graminoid vegetation might be more beneficial to enhancing nutrient cycling rates in areas it dominated. Due to differences in their functional traits, the quantity and quality of C and N released from decomposing litter and roots might differ among vegetation types [47, 48]. For example, Fu *et al.* [49] studied the effect of four shrub-grass types on SOC and TN in the Loess Plateau and found higher SOC in soils with vegetation characterized by higher aboveground biomass and underground root density. Chen *et al.* [50] proposed that soil N recycling rate and availability in soils with graminoid *Agropyron desertorum* was higher than that in soils with *Artemisia* plants due to differences in litter quality.

Except for WSOC, we found significantly increased SOC, MBC, AN, MBN, and TN in soils dominated by graminoids compared those with *Dicranopteris*, which is consistent with the higher litter and root quality of the graminoids (Table 1). In this study, the dominant species in plots with graminoids is *Paspalum thunbergii*, which is characterized by N fixing ability. The N-fixing ability might induce higher N levels in *P. thunbergii* soils [26], which would further increase the litter quality produced by *P. thunbergii*. Indeed, both litter and root N concentration were higher in graminoid areas, accompanied by higher soil N availability [44]. Stokdyk and Herrman [18] reported N-rich *Frangula alnus* leaf litter enhances soil N mineralization. Therefore, if there are no differences in soil abiotic factors, higher litter quality might be followed by faster C and N return rate and hence soil C and N availability. In addition, the positive effects of graminoids on soil C and N components were also consistent with the lower RSR of graminoids relative to *Dicranopteris*, indicating higher soil nutrient availabilities in these graminoid soils. Hence, the colonization of graminoid in plantations dominated by *Dicranopteris* should be promoted to improve the soil quality of aerially seeded *Pinus massoniana* plantations.

While soil microbial community and enzyme activity could have contributed to changes in soil C and N processes [51–53], which should be considered in further studies examining understory vegetation effects on soil C and N process. In addition, this study was conducted in areas with similar erosion levels before the colonization of both studied understory vegetation types. Areas with different erosion levels might also differ in understory vegetation types and soil element cycling. Future studies with manipulation levels of soil erosion and understory vegetation types would be necessary in understanding the effects of understory vegetation types on soil C and N cycling in aerially seeded plantations.

## Conclusion

Our results suggest that understory vegetation generated different effects on soil C and N processes. Graminoid understory vegetation with higher litter and root quality increased soil C and N components. Considering their significant contribution to atmospheric compositions and mitigations of global climate change, variation in soil C and N as affected by different understory vegetation should be considered in future studies, especially those in degraded areas where aerial seeding afforestation has been widely implemented with different understory vegetation types.

## Supporting information

**S1 Table. Description of the studied plots (DBH: Diameter at breast height; N: Tree number).**

(DOCX)

## Acknowledgments

This study was financially supported by the National Natural Science Foundation of China (No. 31360181 and 31160159) and Postdoctoral Science Foundation of Jiangxi Agricultural University (No. 9232306047).

## Author Contributions

**Conceptualization:** Ping Pan, Fang Zhao, Jinkui Ning, Xunzhi Ouyang.

**Data curation:** Ping Pan, Ling Zhang, Xunzhi Ouyang.

**Formal analysis:** Ping Pan, Ling Zhang, Xunzhi Ouyang.

**Funding acquisition:** Ling Zhang, Xunzhi Ouyang.

**Investigation:** Ping Pan, Ling Zhang, Xunzhi Ouyang, Hao Zang.

**Methodology:** Ping Pan, Fang Zhao, Jinkui Ning, Xunzhi Ouyang.

**Project administration:** Jinkui Ning.

**Resources:** Ping Pan, Jinkui Ning, Ling Zhang, Xunzhi Ouyang.

**Software:** Ping Pan, Ling Zhang, Xunzhi Ouyang.

**Supervision:** Xunzhi Ouyang.

**Validation:** Xunzhi Ouyang.

**Visualization:** Ping Pan, Fang Zhao, Jinkui Ning, Ling Zhang, Xunzhi Ouyang.

**Writing – original draft:** Ping Pan, Jinkui Ning, Ling Zhang, Xunzhi Ouyang.

**Writing – review & editing:** Ping Pan, Fang Zhao, Jinkui Ning, Ling Zhang, Xunzhi Ouyang, Hao Zang.

## References

1. Reich PB, Hobbie SE, Lee T, Ellsworth DS, West JB, Tilman D, et al. Nitrogen limitation constrains sustainability of ecosystem response to CO<sub>2</sub>. *Nature*. 2006; 440(7086): 922–925. <https://doi.org/10.1038/nature04486> PMID: 16612381
2. Ren W, Tian H, Tao B, Huang Y, Pan S. China's crop productivity and soil carbon storage as influenced by multifactor global change. *Global Change Biology*. 2012; 18(9): 2945–2957. <https://doi.org/10.1111/j.1365-2486.2012.02741.x> PMID: 24501069
3. Merbold L, Eugster W, Stieger J, Zahniser M, Nelson D, Buchmann N. Greenhouse gas budget (CO<sub>2</sub>, CH<sub>4</sub> and N<sub>2</sub>O) of intensively managed grassland following restoration. *Global Change Biology*. 2014; 20(6): 1913–1928. <https://doi.org/10.1111/gcb.12518> PMID: 24395474
4. Raich JW, Potter CS. Global Patterns of Carbon-Dioxide Emissions from Soils. *Global Biogeochemical Cycles*. 1995; 9(1): 23–36.
5. Shrestha RK, Strahm BD, Sucre EB. Greenhouse gas emissions in response to nitrogen fertilization in managed forest ecosystems. *New Forests*. 2015; 46(2): 167–193.
6. Roy A, Singh KP. Dynamics of microbial biomass and nitrogen supply during primary succession on blastfurnace slag dumps in dry tropics. *Soil Biology & Biochemistry*. 2003; 35(3): 365–372.
7. Wang QK, Wang SL. Response of labile soil organic matter to changes in forest vegetation in subtropical regions. *Applied Soil Ecology*. 2011; 47(3): 210–216.
8. Deng BL, Li ZZ, Zhang L, Ma YC, Li Z, Zhang WY, et al. Increases in soil CO<sub>2</sub> and N<sub>2</sub>O emissions with warming depend on plant species in restored alpine meadows of Wugong Mountain, China. *Journal of Soils and Sediments*. 2016; 16(3): 777–784.
9. Sohng J, Singhakumara BMP, Ashton MS. Effects on soil chemistry of tropical deforestation for agriculture and subsequent reforestation with special reference to changes in carbon and nitrogen. *Forest Ecology and Management*. 2017; 389: 331–340.
10. Ge J, Xie Z, Xu W, Zhao C. Controls over leaf litter decomposition in a mixed evergreen and deciduous broad-leaved forest, Central China. *Plant and Soil*. 2017; 412(1–2): 345–355.
11. Inoue T, Fukuzawa K, Watanabe T, Yoshida T, Shibata H. Spatial pattern of soil nitrogen availability and its relationship to stand structure in a coniferous-broadleaved mixed forest with a dense dwarf bamboo understory in northern Japan. *Ecological Research*. 2017: 1–15.
12. Chastain RA, Currie WS, Townsend PA. Carbon sequestration and nutrient cycling implications of the evergreen understory layer in Appalachian forests. *Forest Ecology and Management*. 2006; 231(1): 63–77.
13. Minick KJ, Strahm BD, Fox TR, Sucre EB, Leggett ZH, Zerpa JL. Switchgrass intercropping reduces soil inorganic nitrogen in a young loblolly pine plantation located in coastal North Carolina. *Forest Ecology and Management*. 2014; 319: 161–168.

14. Gurlevik N, Kelting D, Allen HL. Nitrogen mineralization following vegetation control and fertilization in a 14 year old loblolly pine plantation. *Soil Science Society Of America Journal*. 2004; 68(1): 272–281.
15. Wu J, Liu Z, Wang X, Sun Y, Zhou L, Lin Y, et al. Effects of understorey removal and tree girdling on soil microbial community composition and litter decomposition in two Eucalyptus plantations in South China. *Functional Ecology*. 2011; 25(4): 921–931.
16. Sierra J. Temperature and soil moisture dependence of N mineralization in intact soil cores. *Soil Biology & Biochemistry*. 1997; 29(s 9–10): 1557–1563.
17. Blazier MA, Hennessey TC, Deng S. Effects of fertilization and vegetation control on microbial biomass carbon and dehydrogenase activity in a juvenile loblolly pine plantation. *Forest Science*. 2005; 51(5): 449–459.
18. Stokdyk JP, Herrman KS. Effects of *Frangula alnus* on soil microbial communities and biogeochemical processes in Wisconsin forests. *Plant and Soil*. 2016; 409(1–2): 65–75.
19. Matsushima M, Chang SX. Effects of understorey removal, N fertilization, and litter layer removal on soil N cycling in a 13-year-old white spruce plantation infested with Canada bluejoint grass. *Plant and Soil*. 2007; 292(1): 243–258.
20. Wu J, Liu Z, Huang G, Chen D, Zhang W, Shao Y, et al. Response of soil respiration and ecosystem carbon budget to vegetation removal in Eucalyptus plantations with contrasting ages. *Scientific Reports*. 2014; 4(203): 6262.
21. Wan S, Zhang C, Chen Y, Zhao J, Wang X, Wu J, et al. The understorey fern *Dicranopteris dichotoma* facilitates the overstorey Eucalyptus trees in subtropical plantations. *Ecosphere*. 2014; 5(5): art51-art.
22. Knops JMH, Bradley KL, Wedin DA. Mechanisms of plant species impacts on ecosystem nitrogen cycling. *Ecology Letters*. 2002; 5(3): 454–466.
23. Tateno M, Chapin FS. The logic of carbon and nitrogen interactions in terrestrial ecosystems. *American Naturalist*. 1997; 149(4): 723–744.
24. Xiao X, Wei X, Liu Y, Ouyang X, Li Q, Ning J. Aerial seeding: an effective forest restoration method in highly degraded forest landscapes of sub-tropic regions. *Forests*. 2015; 6(6): 1748–1762.
25. Liu YC, Jing LQ, Wang HQ, Ze qing MA, Jia XW. Characteristics of biomass allocation of *Dicranopteris dichotoma*. *Chinese Journal of Ecology*. 2008; 27(5): 705–711.
26. Barea JM, Brown ME. Effects on plant growth produced by *Azotobacter paspali* related to synthesis of plant growth regulating substances. *Journal of Applied Bacteriology*. 1974; 37(4): 583–593. PMID: [4611996](https://pubmed.ncbi.nlm.nih.gov/4611996/)
27. Ding S, Ying X, Lv D, Ouyang X. Effects of understorey vegetation coverage on soil quality of aerial seeding *Pinus massoniana* stands in south of Jiangxi province. *Research of Soil and Water Conservation*. 2014; 21(3): 31–36.
28. Teng Q, Sun B, Fu X, Li S, Cui Z, Cao H. Analysis of nifH gene diversity in red soil amended with manure in Jiangxi, South China. *Journal Of Microbiology*. 2009; 47(2): 135–141.
29. Zhao F, Ouyang X. Assessing relative contributions of various influencing factors to soil organic carbon in aerielly-seeded *Pinus massoniana* plantations. *Acta Ecologica Sinica*. 2016; 36(9): 2637–2645.
30. Cheng D, Zhong Q, Niklas KJ, Ma Y, Yang Y, Zhang J. Isometric scaling of above- and below-ground biomass at the individual and community levels in the understorey of a sub-tropical forest. *Annals Of Botany*. 2015; 115(2): 303. <https://doi.org/10.1093/aob/mcu238> PMID: [25564468](https://pubmed.ncbi.nlm.nih.gov/25564468/)
31. Jacob CE, Tozzi E, Willenborg CJ. Neighbour presence, not identity, influences root and shoot allocation in pea. *PLoS ONE*. 2017; 12(3): e0173758. <https://doi.org/10.1371/journal.pone.0173758> PMID: [28291827](https://pubmed.ncbi.nlm.nih.gov/28291827/)
32. Lu RK. *Methods of Soil and Agro-chemical Analysis*. Beijing (in Chinese): China Agricultural Science & Technology Press; 2000.
33. Brookes PC, Landman A, Pruden G, Jenkinson DS. Chloroform fumigation and the release of soil nitrogen: A rapid direct extraction method to measure microbial biomass nitrogen in soil. *Soil Biology and Biochemistry*. 1985; 17(6): 837–842.
34. Jenkinson DS, Powlson DS. The effects of biocidal treatments on metabolism in soil. *Soil Biology and Biochemistry*. 1976; 8(3): 209–213.
35. Fang H, Cheng S, Yu G, Xu M, Wang Y, Li L, et al. Experimental nitrogen deposition alters the quantity and quality of soil dissolved organic carbon in an alpine meadow on the Qinghai-Tibetan Plateau. *Applied Soil Ecology*. 2014; 81: 1–11.
36. Yimer F, Ledin S, Abdelkadir A. Soil organic carbon and total nitrogen stocks as affected by topographic aspect and vegetation in the Bale Mountains, Ethiopia. *Geoderma*. 2006; 135(135): 335–344.

37. Jensen LS, Mueller T, Magid J, Nielsen NE. Temporal variation of C and N mineralization, microbial biomass and extractable organic pools in soil after oilseed rape straw incorporation in the field. *Soil Biology and Biochemistry*. 1997; 29(7): 1043–1055.
38. Liang BC, Gregorich EG, Schnitzer M, Voroney RP. Carbon mineralization in soils of different textures as affected by water-soluble organic carbon extracted from composted dairy manure. *Biology and Fertility Of Soils*. 1996; 21(1–2): 10–16.
39. Martin-Olmedo P, Rees RM. Short-term N availability in response to dissolved-organic-carbon from poultry manure, alone or in combination with cellulose. *Biology and Fertility Of Soils*. 1999; 29(4): 386–393.
40. Zhao J, Wan S, Li Z, Shao Y, Xu G, Liu Z, et al. Dicranopteris-dominated understory as major driver of intensive forest ecosystem in humid subtropical and tropical region. *Soil Biology and Biochemistry*. 2012; 49: 78–87.
41. Bradleycook JI, Petrenko CL, Friedland AJ, Virginia RA, editors. Soil carbon storage and temperature sensitivity associated with shrub and graminoid vegetation in Kangerlussuaq, Greenland. AGU Fall Meeting; 2014.
42. Yildiz O, Cromack K, Radosevich SR, Martinezghersa MA, Baham JE. Comparison of 5th- and 14th-year Douglas-fir and understory vegetation responses to selective vegetation removal. *Forest Ecology and Management*. 2011; 262(4): 586–597.
43. Wang F, Zou B, Li H, Li Z. The effect of understory removal on microclimate and soil properties in two subtropical lumber plantations. *Journal of Forest Research*. 2014; 19(1): 238–243.
44. Zhang L, Wang H, Zou J, Rogers WE, Siemann E. Non-Native plant litter enhances soil carbon dioxide emissions in an invaded annual grassland. *PLoS ONE*. 2014; 9(3): e92301. <https://doi.org/10.1371/journal.pone.0092301> PMID: 24647312
45. Zhang L, Ma X, Wang H, Liu S, Siemann E, Zou J. Soil respiration and litter decomposition increased following perennial forb invasion into an annual grassland. *Pedosphere*. 2016; 26(4): 567–576.
46. Zhang L, Zou J, Siemann E. Interactive effects of elevated CO<sub>2</sub> and nitrogen deposition accelerate litter decomposition cycles of invasive tree (*Triadica sebifera*). *Forest Ecology and Management*. 2017; 385: 189–197.
47. Langley JA, Hungate BA. Mycorrhizal controls on belowground litter quality. *Ecology*. 2003; 84(9): 2302–2312.
48. Sauheitl L, Glaser B, Dippold M, Leiber K, Weigelt A. Amino acid fingerprint of a grassland soil reflects changes in plant species richness. *Plant and Soil*. 2010; 334(1): 353–363.
49. Fu X, Shao M, Wei X, Horton R. Soil organic carbon and total nitrogen as affected by vegetation types in Northern Loess Plateau of China. *Geoderma*. 2010; 155(1): 31–35.
50. Chen J, Stark JM. Plant species effects and carbon and nitrogen cycling in a sagebrush-crested wheat-grass soil. *Soil Biology & Biochemistry*. 2000; 32(1): 47–57.
51. Ramirez KS, Craine JM, Noah F. Consistent effects of nitrogen amendments on soil microbial communities and processes across biomes. *Global Change Biology*. 2012; 18(6): 1918–1927.
52. Gallo M, Amonette R, Lauber C, Sinsabaugh RL, Zak DR. Microbial community structure and oxidative enzyme activity in nitrogen-amended north temperate forest soils. *Microbial Ecology*. 2004; 48(2): 218–229. <https://doi.org/10.1007/s00248-003-9001-x> PMID: 15546042
53. Khalili B, Ogunseitan OA, Goulden ML, Allison SD. Interactive effects of precipitation manipulation and nitrogen addition on soil properties in California grassland and shrubland. *Applied Soil Ecology*. 2016; 107: 144–153.