



Full Length Article

Stand Characteristics and Soil Properties in Japanese Red Pine (*Pinus densiflora*) Pure Forests with Different Disease Severity Index in Kunyushan Mountains Region, China

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Abstract

Pine needle blight (*Pestalotiopsis funereal* Desm. Steyaert) has caused major damage to the Japanese red pine (*P. densiflora* Sieb. et Zucc.) pure forests in the Kunyushan Mountains region of China. In this study, six *P. densiflora* stand types (ST- 1-6) were selected and surveyed to determine the relationships between the stand characteristics, soil properties and the disease occurrence. Our results showed that the stem densities in the sampled plots of infected stands (ST- 2-6) differed significantly from those in healthy plots (ST- 1) ($P < 0.01$), with similar patterns found for diameter at breast height (DBH) and basal area at breast in infected plots versus healthy ones ($P < 0.05$). Soil bulk densities among the six stand types were significantly different ($P < 0.05$), being lowest in ST- 1. Generally, soil water-related physical properties, except for bulk density and maximum moisture, did not differ remarkably among the stands. Regarding their soil chemical properties, organic matter and total nitrogen were higher in ST- 1, on average, whereas the total potassium was lower there than infected stands. The differences in organic matter, total nitrogen and total potassium among the six stand types were all significant ($P < 0.05$). Furthermore, the redundancy analysis suggested that 12 of 19 examined soil variables that were strongly related with the ordinations of stand characteristics. Key among these soil factors was available nitrogen, bulk density, pH, organic matter and total nitrogen. Taken together, these empirical results demonstrate the response of pine stands to disease, and are potentially valuable for guiding applications of ecological control of pine needle blight. © 2019 Friends Science Publishers

Keywords: Japanese red pine pure forests; Pine needle blight; Redundancy analysis; Soil properties; Stand type; Stand characteristics

Introduction

Japanese red pine (JRP; *Pinus densiflora*) is one of the most economically and environmentally important coniferous forest species in the Kunyushan Mountains region, where pure forests of this tree account for ca. 70% forested hectares (Du, 2011; Sun *et al.*, 2015). The health and stability of these JRP-dominated stands are closely related to the successional dynamics and resilience of local forest ecosystems. However, the JRP forests have long been under constant threat from pine needle blight disease caused by *Pestalotiopsis funerea*, which can infect many needles, making it a major disease affecting JRP foliage (Orlikowski *et al.*, 2014). In particular, 1-year-old leaf needles are most vulnerable to its impact. However, the disease also harms 2-year-old needles, withering their tips, and even affecting the stability of the ecosystem when the disease becomes serious (Li *et al.*, 2012). Being a globally

widespread fungus, *P. funerea* is reportedly an opportunistic invader of conifers (Gonthier and Nicolotti, 2002).

Pestalotiopsis species can cause a variety of diseases in plants, one of which is pine needle blight (Espinoza *et al.*, 2008). To date, scholars in China and abroad have conducted comprehensive studies into the pathological and physiological characteristics of *P. funerea* (e.g., Kozłowska *et al.*, 2002; Zhou *et al.*, 2002; Ivanová, 2016). This pathogenic fungus is white at the initial stage in PDA medium, and then gradually turns pale yellow, becoming pink or brown as the mycelia mature. The black conidia are transmitted by rain or wind and complete their initial infection via conidiophores (Kozłowska *et al.*, 2002; Ivanová, 2016). Other studies have reported on ways to prevent this disease, including chemical control, biological control, and ecological control (Miyakado, 1986; Zhou *et al.*, 2002; Liang *et al.*, 2016). The regularity of pine needle blight epidemics is well studied, but few reports exist on the

relationship between soil properties and this disease (Dijk *et al.*, 1992). To fill this knowledge gap, it is important to survey the variation in local soil conditions and tree growth dynamics in JRP forests that have been infected by pine needle blight. As far as we know, this is the first study to compare the differences of stand characteristics and soil properties of JRP pure forests after suffering pine needle blight in the Kunyushan Mountains region in China.

The objective of this study was to investigate potential differences among stand characteristics and soil properties in healthy JRP pure forests and those damaged by pine needle blight. We addressed the following two questions: (1) In what way do stand characteristics and soil properties differ among the JRP stand types? (2) What is the relative importance of different soil properties to tree growth in JRP pure forests varying in disease severity?

Materials and Methods

Study Area

The Kunyushan Mountains lie in the Jiaodong Peninsula of eastern Shandong Province, China (121°41'34"–121°48'04"E; 37°11'50"–37°17'22"N). The climate in this region is moderate, due to the warm-temperate monsoon climate, with a mean annual temperature of 12.3°C. There is a frost-free period of 200–220 days, with an average annual precipitation and a mean annual relative humidity of 800–1200 mm and 62.6%, respectively. The soil type is mostly brown soil and sandy loam. Tree of JRP are the main indigenous conifers found here; they have less demanding site conditions, and are naturally distributed from the piedmont up to highest areas of the mountain, at 800 m above sea level.

Site and Stand Characteristics Survey

This work was guided on the "Observation Methodology for Long-term Forest Ecosystem Research" of National Standards of the People's Republic of China (GB/T 33027-2016). A total of 136 temporary plots (each 30 m × 30 m) were established in JRP pure forest (from May to August, 2017), sharing the same soil type and other similar site conditions. Temporary markers were set using eight tagged trees—one per plot corner and another four lying midway along each plot's four borders—and surrounded with red ropes. Site features and stand factors of each plot were then recorded in the field, including elevation, slope degree and canopy cover. Each tree with diameter at breast height (DBH) ≥ 2 cm was numbered in every JRP pure forest plot. For each tree, its height, crown width, DBH, and canopy density were recorded. Canopy density was measured by a CI-110 Plant Canopy Digital Imager (CID Inc., Vancouver, USA). The stand density was expressed as the number of trees on per hectare basis. The elevation of each plot was obtained from a portable GPS.

From the DBH measurements we derived basal area at breast height, and then stand volume in the sampled plots was calculated as follows (Gao *et al.*, 2015a):

$$G_{1.3} = \pi D^2 / 4 \quad (1)$$

$$M = G_{1.3} (h + 3) fa \quad (2)$$

In the above equations, $G_{1.3}$ is the basal area at breast height in $\text{m}^2 \cdot \text{ha}^{-1}$, D is the diameter at breast height (DBH) in cm, M is the stand volume in $\text{m}^3 \cdot \text{ha}^{-1}$, h is the tree height in m and fa is the experimental form value ($= 0.42$).

Disease Severity Index

Disease status of JRP pure forest in each plot was described by a disease severity index (DSI). This DSI for pine needle blight was measured by the "five-spot" method, in that two trees were taken from each of the four corners and the center of each plot. These 10 JRP trees were divided into upper, middle and lower layers, for which one branch was taken from each direction of east, south, west and north. The needles of each sampled branch were presumed as approximately cylindrical—with the same area at the needle bottom as at its tip—so the ratio of the blight's spotted area to the leaf area could be converted into the proportion of needle length afflicted with disease. We pooled and measured these ratios on a per tree basis. The disease index of a JRP tree was calculated according to the weighted average method of five grades, and then an average DSI per plot was obtained. The classification criteria for pine needle blight are shown in Table 1 (Zhang *et al.*, 2015). The 136 plots were divided into six stand types (ST) according to the disease severity index (DSI): ST-1 was uninfected (DSI = 0), while ST-2 ($0 < \text{DSI} \leq 20$), ST-3 ($20 < \text{DSI} \leq 40$), ST-4 ($40 < \text{DSI} \leq 60$), ST-5 ($60 < \text{DSI} \leq 80$) and ST-6 ($80 < \text{DSI} \leq 100$) had all been infected with pine needle blight albeit different in extent (Table 2).

According to the criteria above, the formula for a DSI can be described by using these equations:

$$\text{SARB}\% = \frac{\sum \text{DSL}}{\text{NL}} \times 100\% \quad (3)$$

SARB is the needle disease spot area ratio of each branch; DSL is the needle disease spot lengths of each branch; NL is needle lengths of each branch.

$$\text{DSIT} = \frac{\sum_{i=1}^{12} (\text{NDB} \times \text{CGV})}{\text{TB} \times \text{MRV}} \quad (4)$$

DSIT is the DSI of each tree; NDB is the number of diseased branches; TB is the total number of branches; i is the number of investigated branches, which was fixed at 12 per tree; CGV is the value of corresponding grade; **MRV is maximum representative value, which was always = 4 (Table 1).**

$$\text{DSIP} = \frac{\sum_{j=1}^{10} \text{DSIT}}{10} \quad (5)$$

DSIP is the DSI of each plot; j represents the number of investigated trees that is 10 per plot.

Table 1: The classification criteria for pine needle blight of JRP branch

| Grade of disease | Value corresponding grade | Basis of classification |
|------------------|---------------------------|--------------------------------------------------------------|
| I | 0 | Without any disease spots |
| II | 1 | The disease spot area is less than 25% of each branch |
| III | 2 | The disease spot area is between 25% and 50% of each branch |
| IV | 3 | The disease spot area is between 50% and 75% of each branch |
| V | 4 | The disease spot area is between 75% and 100% of each branch |

Table 2: Characteristics of six diseased stand types of JRP pure forest

| Stand type | DSI | Elevation (m) | Slope degree (°) | Canopy density |
|------------|--------------|---------------|------------------|----------------|
| ST-1 | 0.00 ± 0.00 | 381.5 ± 60.2 | 24.62 ± 4.50 | 0.77 ± 0.06 |
| ST-2 | 10.42 ± 2.23 | 270.0 ± 40.4 | 18.00 ± 3.71 | 0.65 ± 0.17 |
| ST-3 | 30.27 ± 4.54 | 250.2 ± 56.4 | 19.95 ± 9.65 | 0.72 ± 0.13 |
| ST-4 | 50.49 ± 5.48 | 209.1 ± 39.7 | 22.79 ± 1.87 | 0.75 ± 0.11 |
| ST-5 | 66.37 ± 4.75 | 218.7 ± 41.2 | 12.73 ± 2.47 | 0.79 ± 0.11 |
| ST-6 | 92.91 ± 6.29 | 173.0 ± 40.0 | 13.00 ± 4.26 | 0.88 ± 0.02 |

Notes: ST-1 is the control (no disease). DSI means the disease severity index. JRP is the Japanese red pine. Mean ± standard error. The same below

Soil Sampling and Processing

A composite soil sample was obtained from each plot by using the five-point sampling approach. Then, using the cutting ring method (ring = 100 cm³), soil samples were taken from 0–10 cm depth for physical analyses of soil bulk density (g cm⁻³), soil water content (%), full water capacity (%), capillary moisture capacity (%), capillary porosity (%), noncapillary porosity (%) and total porosity (%). For further details on these methods and measurements, refer to national forestry standards (LY/T 1215-1999) (Zhang *et al.*, 1999).

Soil samples were also taken from 0–20 cm depth in each plot and fully mixed to determine its chemical properties. Visible impurities in the soil samples, such as stones, animal and plant residues, as well as other litter, were removed manually. The soil particles were then air-dried and ground, so that the soil particles could pass through a sieve with a 2 mm diameter. The chemical variables of interest, which were tested by the key laboratory of the state forestry administration, consisted of soil organic matter, total nitrogen, total phosphorus, total potassium, available phosphorus, available potassium, available nitrogen, ferric ion, copper ion, zinc ion, and manganese ion and pH (Gao *et al.*, 2015b).

Data Analysis

Analysis of variance (ANOVA) followed by least significant difference (LSD) testing at the $P < 0.05$ level was used to compare means of soil physicochemical properties and the stand characteristics among the six JRP stand types. Data calculations and the statistical analyses were respectively performed using Microsoft Excel 2007 and SPSS v22.0.

Redundancy analysis (RDA) is a multivariable direct gradient analysis method that performs multiple regressions of many environmental variables and response data (Braak and Smilauer, 2012). To simplify the number of variables effectively, RDA carries out a series of decomposition and selection of eigenvalues. The relationships between species and environmental factors can be visualized on the same coordinate axis, so as to better analyze the effect of dominant factors on species factors (Juratfuentes and Adang, 2006).

The ordination of the JRP stand characteristics among the six stand types was conducted using CANOCO v5.0 (Braak and Smilauer, 2012). A detrended correspondence analysis (DCA) was used to test whether CCA or RDA was the best analytical method (Gao *et al.*, 2015b). In this study, 19 soil physical and chemical properties were considered as environmental predictor variables in the analysis of their effect on stand characteristics for each plot. On the basis of Monte Carlo permutation tests with $n = 499$ iterations, the forward selection procedure was used to select the environmental variables, in which those P -values < 0.05 were retained for the ordination of stand characteristics and used as the corresponding variables in the final analyses (HeděNec *et al.*, 2014).

Results

Changed Stand Characteristics in JRP Pure Forests

Table 3 summarized the mean changes of stand characteristics in the six stand types of JRP pure forests. The stand density in the blight-infected plots showed a significant upward trend relative to the healthy disease-free control plots (ST-1) and the effect of stand type was highly significant. Both the DBH and basal area at breast height of JRP pure forest increased significantly with DSI and both significantly differed among stand types. The mean stand volume in ST-1 was the highest, at ca. 78 m³·ha⁻¹, while those of ST-1, ST-2 and ST-3 were significantly higher than those of ST-4, ST-5 and ST-6. However, tree height and crown width were similar among the six stand types ($P > 0.05$).

Changed Soil Physicochemical Properties in JRP Pure Forests

Table 4 and 5 showed the changes in soil physical and chemical properties in the JRP pure forest stands with different blight infection grades. No remarkable differences were observed for the physical factors among the differently infested plots, except for soil bulk density and maximum moisture capacity. These results showed that the values of all factors apart from soil bulk density were higher in the healthy plots than in infected ones. Concerning the maximum moisture capacity in soil, the differences among stand types were significant ($P < 0.05$), while the differences in water content, capillary moisture capacity, capillary porosity and total capillary porosity were non-significant ($P > 0.05$).

Table 3: Stand characteristics of JRP pure forest in the different diseased stand types of JRP

| Stand type | Stand density (trees·ha ⁻¹) | Tree height (m) | Crown width (m) | DBH (cm) | Basal area at breast height (m ² ha ⁻¹) | Stand volume (m ³ ha ⁻¹) |
|------------|-----------------------------------------|-----------------|-----------------|----------------|----------------------------------------------------------------|-------------------------------------------------|
| ST-1 | 1878 ± 252 d | 7.05 ± 1.89 a | 2.72 ± 0.67 a | 13.89 ± 3.31 a | 17.67 ± 7.94 a | 77.58 ± 12.28 a |
| ST-2 | 1926 ± 475 cd | 5.53 ± 1.74 a | 2.65 ± 0.39 a | 11.89 ± 2.33 b | 16.66 ± 4.58 a | 59.68 ± 9.91 a |
| ST-3 | 1582 ± 365 d | 6.26 ± 1.98 a | 2.97 ± 1.00 a | 13.25 ± 3.81 a | 16.57 ± 8.89 a | 68.27 ± 10.07 a |
| ST-4 | 2351 ± 150 cd | 5.76 ± 1.87 a | 2.65 ± 1.05 a | 11.11 ± 2.88 b | 11.48 ± 5.76 b | 45.11 ± 8.63 b |
| ST-5 | 2872 ± 154 b | 5.82 ± 2.90 a | 2.50 ± 0.87 a | 11.10 ± 3.09 b | 11.53 ± 6.20 b | 45.09 ± 7.88 b |
| ST-6 | 3556 ± 362 a | 5.88 ± 1.13 a | 2.51 ± 0.61 a | 11.35 ± 2.03 b | 11.50 ± 4.19 b | 44.35 ± 8.09 b |
| F-value | 75.429** | 0.701 | 0.836 | 2.389* | 2.637* | 2.393* |
| P-value | < 0.001 | 0.624 | 0.526 | 0.042 | 0.027 | 0.041 |

Notes: * Significant difference among stand types ($P < 0.05$), **Highly significant difference among stand types ($P < 0.01$). Values within a column with different letters are significantly different. The same applies to the Tables below

Table 4: Water-related physical properties of soil in different stand types

| Stand type | Bulk density (g cm ⁻³) | Soil water content (%) | Maximum moisture capacity (%) | Capillary moisture capacity (%) | Capillary porosity (%) | Non-capillary porosity (%) | Total capillary porosity (%) |
|------------|------------------------------------|------------------------|-------------------------------|---------------------------------|------------------------|----------------------------|------------------------------|
| ST-1 | 1.08 ± 0.20 b | 9.60 ± 3.81 a | 22.25 ± 4.21 a | 15.07 ± 3.32 a | 15.07 ± 2.79 a | 7.89 ± 1.02 a | 24.03 ± 2.95 a |
| ST-2 | 1.21 ± 0.12 a | 9.25 ± 2.16 a | 19.69 ± 2.25 b | 12.40 ± 1.80 a | 12.65 ± 1.55 a | 8.74 ± 1.34 a | 23.70 ± 2.88 a |
| ST-3 | 1.14 ± 0.23 ab | 8.51 ± 2.97 a | 19.31 ± 3.12 ab | 12.70 ± 3.82 a | 14.29 ± 2.93 a | 7.72 ± 1.48 a | 23.01 ± 3.56 a |
| ST-4 | 1.19 ± 0.32 ab | 7.76 ± 2.73 a | 18.66 ± 2.55 ab | 12.18 ± 2.22 a | 14.45 ± 2.29 a | 7.24 ± 1.42 a | 21.70 ± 2.78 a |
| ST-5 | 1.16 ± 0.17 ab | 6.93 ± 1.44 a | 16.64 ± 3.07 a | 10.24 ± 3.09 a | 11.92 ± 2.71 a | 7.96 ± 1.11 a | 19.88 ± 2.57 a |
| ST-6 | 1.27 ± 0.15 a | 6.20 ± 0.43 a | 19.31 ± 2.97 ab | 11.73 ± 0.01 a | 14.48 ± 2.02 a | 10.24 ± 1.37 a | 25.72 ± 0.65 a |
| F-value | 2.85* | 1.474 | 3.633** | 1.480 | 1.282 | 1.047 | 1.291 |
| P-value | 0.020 | 0.209 | 0.005 | 0.206 | 0.281 | 0.396 | 0.276 |

Table 5: Chemical properties of soil in different stand types

| Stand type | Organic matter (g kg ⁻¹) | Total nitrogen (g kg ⁻¹) | Total phosphorus (g kg ⁻¹) | Total potassium (g kg ⁻¹) | Available nitrogen (mg kg ⁻¹) | Available phosphorus (mg kg ⁻¹) | Available potassium (mg kg ⁻¹) | Cupric ion (mg kg ⁻¹) | Zinc ion (g kg ⁻¹) | Ferric ion (g kg ⁻¹) | Manganese ion (g kg ⁻¹) | pH |
|------------|--------------------------------------|--------------------------------------|----------------------------------------|---------------------------------------|-------------------------------------------|---------------------------------------------|--------------------------------------------|-----------------------------------|--------------------------------|----------------------------------|-------------------------------------|---------------|
| ST-1 | 56.40 ± 11.82 a | 1.966 ± 0.883 a | 0.244 ± 0.033 a | 21.39 ± 3.85 b | 112.99 ± 12.21 a | 1.23 ± 0.22 a | 74.27 ± 9.29 a | 5.24 ± 0.56 a | 0.085 ± 0.001 a | 24.19 ± 4.94 a | 0.46 ± 0.08 a | 4.23 ± 0.26 a |
| ST-2 | 25.23 ± 2.33 b | 0.899 ± 0.012 b | 0.099 ± 0.010 a | 24.76 ± 2.88 a | 86.08 ± 8.51 a | 0.83 ± 0.14 a | 52.39 ± 8.65 a | 3.98 ± 0.45 a | 0.079 ± 0.003 a | 19.82 ± 1.15 a | 0.54 ± 0.11 a | 4.72 ± 0.13 a |
| ST-3 | 45.08 ± 8.42 a | 1.603 ± 0.021 a | 0.224 ± 0.014 a | 22.55 ± 4.15 b | 105.06 ± 10.86 a | 1.29 ± 0.19 a | 67.90 ± 10.95 a | 5.49 ± 0.47 a | 0.086 ± 0.001 a | 24.11 ± 4.34 a | 0.45 ± 0.08 a | 4.38 ± 0.24 a |
| ST-4 | 47.08 ± 8.17 a | 1.463 ± 0.154 a | 0.245 ± 0.019 a | 20.75 ± 4.61 b | 86.87 ± 8.23 a | 1.07 ± 0.12 a | 66.81 ± 6.38 a | 4.91 ± 0.51 a | 0.083 ± 0.003 a | 23.87 ± 4.89 a | 0.42 ± 0.03 a | 4.35 ± 0.25 a |
| ST-5 | 32.36 ± 6.23 b | 0.996 ± 0.157 b | 0.144 ± 0.069 a | 25.11 ± 2.52 a | 71.78 ± 10.41 a | 0.77 ± 0.06 a | 55.35 ± 7.02 a | 4.40 ± 0.59 a | 0.077 ± 0.006 a | 23.24 ± 4.65 a | 0.37 ± 0.04 a | 4.47 ± 0.24 a |
| ST-6 | 22.47 ± 4.80 b | 0.803 ± 0.133 b | 0.104 ± 0.025 a | 24.25 ± 3.23 a | 52.72 ± 9.74 a | 0.64 ± 0.08 a | 66.39 ± 6.05 a | 3.00 ± 0.12 a | 0.061 ± 0.004 a | 21.31 ± 2.88 a | 0.41 ± 0.07 a | 4.57 ± 0.11 a |
| F-value | 2.433* | 2.822* | 1.604 | 2.310* | 2.146 | 1.617 | 0.843 | 0.347 | 1.652 | 0.530 | 1.374 | 2.152 |
| P-value | 0.039 | 0.019 | 0.164 | 0.048 | 0.065 | 0.161 | 0.522 | 0.883 | 0.152 | 0.753 | 0.239 | 0.064 |

Compared with the healthy plots (ST-1), the values of soil bulk densities in the infected plots (ST 2-6) were higher, and the soil bulk densities showed an ascending trend as DSI increased. The value in ST-1 was significantly lower than that in ST-6 ($P < 0.05$; Table 4).

Table 5 showed that organic matter, total nitrogen and total potassium were significantly different among the six stand types ($P < 0.05$). The mean values of these three factors each showed a decreased trend, that is, they were highest in ST-1 yet lowest in ST-6. Soil total phosphorus, available phosphorus, available nitrogen, available potassium, cupric ion, zinc ion, ferric ion, manganese ion and pH were similar among the stand types ($P > 0.05$). The levels of available nitrogen, available potassium, cupric ion, and ferric ion were all slightly higher in healthy plots than in blight-infected plots.

Ordination of Stand Characteristics

Among the 19 physical and chemical soil variables, 12 were found to be significantly related ($P < 0.05$) with the ordination of stand characteristics, including available nitrogen, bulk density, pH, organic matter, total nitrogen, capillary water holding capacity, maximum moisture capacity, available phosphorus, ferric ion, available potassium, total phosphorus, and capillary porosity (Lambda-A, Table 7). The first two components of the RDA axes explained 43.33% of the variance in the relationship between stand characteristics and these 12 selected soil properties. In addition, the F -ratio was relatively high (4.6) and the P -value was low (0.002) (Table 6).

Forward selection of the 12 factors in the RDA ordinations indicated that JRP stand characteristics were

Table 6: Summary of statistical results by redundancy analysis

| Canonical axes | 1 | 2 | 3 | 4 | Total variance | F-ratio | P-value |
|-----------------------------------------------------------|--------|--------|--------|--------|----------------|---------|---------|
| Eigenvalues | 0.4148 | 0.0185 | 0.0159 | 0.0039 | 1 | 4.6 | 0.002 |
| Species-environment correlations | 0.7196 | 0.4356 | 0.4948 | 0.3261 | | | |
| Cumulative percentage of species variance (%) | 41.48 | 43.33 | 44.92 | 45.31 | | | |
| Cumulative percentage of species-environment variance (%) | 91.55 | 95.64 | 99.15 | 100.0 | | | |
| Sum of all eigenvalues | | | | | 1 | | |
| Sum of all canonical eigenvalues | | | | | 0.4531 | | |

Table 7: Marginal and conditional effects of soil variables on stand factors obtained from the summary of forward selection in RDA

| Variables | Lambda-A ¹ | | | Lambda-B ² | | |
|----------------------------------|-----------------------|---------|---------|-----------------------|---------|---------|
| | Contribution (%) | F-ratio | P-value | Contribution (%) | F-ratio | P-value |
| Available nitrogen | 47.3 | 23.8 | 0.002 | 47.3 | 23.8** | 0.002 |
| Bulk density | 33.5 | 15.5 | 0.002 | 13.3 | 7.8** | 0.002 |
| pH | 35.1 | 16.4 | 0.002 | 6.7 | 3.6** | 0.004 |
| Organic matter | 25.7 | 11.4 | 0.002 | 8.0 | 4.9* | 0.02 |
| Total nitrogen | 43.3 | 21.3 | 0.002 | 6.7 | 3.5* | 0.044 |
| Capillary water holding capacity | 21.8 | 9.4 | 0.006 | 2.6 | 1.6 | 0.188 |
| Maximum moisture capacity | 18.0 | 7.6 | 0.004 | 0.6 | 0.4 | 0.688 |
| Available phosphorus | 15.0 | 6.3 | 0.006 | 0.9 | 0.5 | 0.598 |
| Ferric ion | 14.1 | 5.8 | 0.008 | 3.4 | 2.1 | 0.116 |
| Available potassium | 13.8 | 5.7 | 0.016 | 1.0 | 0.6 | 0.558 |
| Capillary porosity | 8.3 | 3.5 | 0.05 | 0.8 | 0.5 | 0.624 |
| Total phosphorus | 9.3 | 3.8 | 0.04 | 0.2 | 0.1 | 0.944 |

¹Describes marginal effects, which show the variance explained when the variable is used as the only factor

²Describes conditional effects, which show the additional variance each variable explains when it is included in the model

³The Monte Carlo permutation with n = 499 iterations was performed at the 0.05 significance level

*Significant at $P < 0.05$, ** Significant at $P < 0.01$

primarily influenced by available nitrogen, bulk density, pH, organic matter and total nitrogen; their total combined contribution amounted to 91% (the contribution of Lambda-B, Table 7). However, the other seven factors did not contribute to that distribution of stand factors in a significant way (Lambda-B, Table 7). Despite the relatively high marginal effect, total phosphorus had a negligible conditional effect on JRP stand characteristics.

The RDA ordination biplot showed specific associations between stand characteristics and soil factors. Bulk density and pH were positively associated with stand density, crown cover and tree height, whereas they had a negative association with stand volume, basal area at breast height and DBH. Available nitrogen, organic matter, and total nitrogen were negatively related to stand density and crown cover, but positively related to DBH, basal area at breast height, and stand volume (Fig. 1).

Discussion

Basic traits of healthy *P. densiflora* pure forests, namely its stand characteristics and soil physicochemical properties, were different from forest stands infected with *P. funerea*. The stand characteristics of JRP pure forests varying in DSI caused by pine needle blight clearly were not alike in the Kunyushan Mountains. In dense plantations, the tree crowns may overlap and form a relatively closed environment (Zhou *et al.*, 2017). The pathogen responsible for pine needle blight

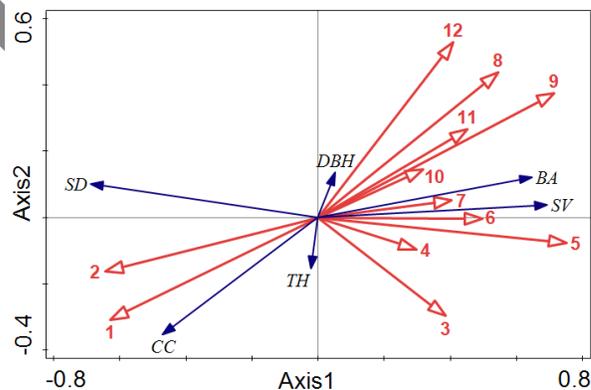


Fig. 1: First two canonical axes of the redundancy analysis (RDA) ordination biplot between the stand characteristics of pure Japanese red pine forest and dominant environmental variables. SD, stand density; TH, tree height; SV, stand volume; CC, crown cover; DBH, diameter at breast height; BA, basal area at breast height. 1. soil bulk density, 2. pH, 3. ferric ion, 4. capillary porosity, 5. available nitrogen, 6. capillary water holding capacity, 7. available potassium, 8. organic matter, 9. total nitrogen, 10. total phosphorus, 11. maximum moisture capacity, 12. available phosphorus

benefits from high temperatures and humidity (Kozłowska *et al.*, 2002; Cui *et al.*, 2015), so *P. funerea* likely breeds more quickly and spreads more easily in dense woodland.

The difference in size, thickness, and vigor of JRP trees ought to be discernible from their DBH, height and canopy (Mao *et al.*, 2014), so that mean DBH would reflect tree's accumulated growth and growing conditions when they are correlated positively (Wang *et al.*, 2006). Trees with greater DBH generally have stronger resistance ability to disease and other disturbances (Jiao *et al.*, 2010). Moreover, basal area and stand volume are important explanatory parameters for the health and value of forests. We found that ST-1 had the highest values of basal area at breast height and stand volume, confirming that it was indeed healthier and likely had a stronger ability to resist blight.

Cheng *et al.* (2010) had analyzed the physiochemical features of rhizosphere soil between a control (infected by tobacco bacterial wilt) and a treatment (biochar applied to control tobacco bacterial wilt), finding that the latter had lower bulk density, but higher porosity, pH, organic carbon, alkalized nitrogen, available phosphorus, and available potassium. The conclusion of our study on JRP forest and pine needle blight is consistent with that above, as there were clear differences in the soil physiochemical properties in JRP pure forests stand depending on their respective disease severity indexes.

The physicochemical properties of forest soils, especially of their surface horizons, are affected by the stand or directly by natural disturbances (Zhou *et al.*, 2008; Ilek *et al.*, 2015). Conversely, key soil properties, such as organic matter, total soil carbon, and nitrogen among others, can influence some foliar or root diseases (Mallett and Maynard, 2004; Rotenberg *et al.*, 2005). The result of our study showed that when compared to healthy plots (ST-1), bulk densities in the infected plots (ST-2 through ST-6) tended to decline, with the means significant different among the six stands. Bulk density affects the absorption of tree root systems and could be estimated from the organic matter content of soil because these variables often exhibit a negative correlation (Hossain *et al.*, 2015). We supposed that higher bulk density and lower organic matter content drove the JRP trees' potential decline, decreasing their disease resistance to *P. funerea*. However, the opposite trend occurred in terms of the soil maximum moisture capacity, which obviously was not the same among the six JRP stand types we investigated. The different water storage capacity of forest soil is probably due to different physical properties, such as bulk density and porosity (Ilek *et al.*, 2017), which affects the movement of both air and water through the soil layers and is an important indicator of soil aeration (Pagliai *et al.*, 2004; Hou *et al.*, 2015; Luna *et al.*, 2018). ST-1 had the lowest bulk density and the highest soil porosity, so its compaction was likely the lowest among stand types, which would explain it having the highest maximum moisture capacity (13.91%). However, the soil water contents were all low in this field study, which may be attributed to limited rainfall and high evapotranspiration from May to July in our study region (Cheng and Liu, 2014).

Table 5 shows the soil organic matter decreased gradually with a greater DSI. The bulk density of ST-6 was

the highest, while its soil organic matter was the lowest among JRP stand types. These results match well those of Wei *et al.* (2012), in that there was a significant negative correlation found between soil organic matter and bulk density. Along with the greater organic matter, total nitrogen was also higher, and they differed among six stands were significant. These results are supported by Gao *et al.* (2015b), who found that organic matter and total nitrogen were slightly lower in infected forest than in healthy forest. Soil organic matter plays a critical role in supplying nutrients to forest plants (Yan *et al.*, 2007), so trees with higher organic matter likely benefited from higher vigor and had a strong ability to generally resist disease. Our results also find support from Cheng *et al.* (2010) that they concluded total nitrogen content was positively related with organic matter content in Masson pine forest. Similar to the organic matter, nitrogen-deficiency stress strongly diminishes leaf expansion rate and leaf area duration, leading to poor growth and vigor of trees (Bu *et al.*, 2014). In general, we found that the content of total potassium in healthy JRP plots was lower than those in blight-infected plots. However, Zeng *et al.* (1985) reported a negative association of needle disease of slash pine and total potassium content in soil. Discrepancies between that study and ours may reflect changed soil properties with the stand conditions, host species investigated, and duration of observation period. The other soil chemical properties, namely total phosphorus, available nitrogen, available phosphorus, available potassium, pH and so on, in the JRP stands types seemed unaffected by DSI, perhaps because soil composition had some short-term stability (Gao *et al.*, 2015b).

The RDA ordination biplot is a useful approach, as it could show how different variables influenced the JRP stand characteristics positively or negatively. RDA not only simplifies the number of variables effectively, but also maintains the independent contribution of each variable to the environment, and it can describe the explanatory ability of specific indicators and the reliability of the ranking quantitatively (Braak and Smilauer, 2012; Brokaw, 2004). This study's results support the use of RDA to explore the relationship between soil properties and the stand characteristics, as it was able to capture the interaction among multiple variables intuitively, which helped us address our research questions.

Conclusion

This study reveals differences in the stand characteristics and soil properties among healthy JRP pure forests and those infected with *P. funerea*. Overall, when compared with the healthy plots, stand densities increased significantly in the infected plots, yet DBH, basal area at breast height, and stand volume were all diminished by pine blight disease. Soil bulk densities differed significantly among the stand types varying in disease severity, with healthy plots having the lowest mean value. Among the soil chemical properties, the mean values

of organic matter and total nitrogen were highest in healthy plots, while total potassium was highest in the infected ones. Other soil physicochemical properties didn't differ remarkably between the healthy and infected stands. In addition, RDA indicated that stand characteristics were strongly related to available nitrogen, bulk density, pH, organic matter and total nitrogen in soil. The results from this study will be used to guide forest management strategies to reduce pine needle blight disease in the Kunyushan Mountains and other similar regions.

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