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Original Research

Vascular Plants as Indicators of Organic Carbon Gradient in Subtropical Forested Soils

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Abstract

To assess the relations of vascular ground flora to soil organic carbon (SOC) contents, we carried out plant census and soil sampling in 3 forest stands, i.e. 1-year and 3-year eucalyptus planted forests and a secondary broadleaved forest in western Guangdong, China. Variability in species composition and diversity in the ground flora was tested using multi-response permutation procedures (MRPP), and potential species indicators were detected by indicator species analysis (ISA). Dominant species in the ground flora differentiated natural secondary forest from eucalyptus planted forests, and the SOC content of the 0~25 cm soil layer varied significantly across the 3 forest stands. Significant differences in SOC content were found among the 5 groupings of the ground flora, which was defined by two-way indicator species analysis (TWINSPAN). MRPP indicated that species composition and diversity of the ground flora varied significantly in response to a gradient of SOC content. ISA revealed that 19 species, including *Smilax glabra* (IV > 50), were indicative of soil environments with high SOC content (> 30 g·kg^{-1}), and only 1 species, i.e. *Inula cappa*, were indicative of low SOC content (< 20 g·kg^{-1}). This study suggested that indicator species of different SOC regimes can be detected using a simple community analysis technique, and such indicator plants can be used for soil nutrient and soil carbon pool dynamics monitoring and assessment.

Keywords: ground flora, soil organic carbon (SOC), indicator species

Introduction

Plant indication refers to the specific response of plants to a certain or integrated environmental gradient [1]. Plant adaptations to different habitats can be verified by their distribution and composition as well as morphological and physiological variations [2-4]. The quantification of plant-environment relations will help facilitate extraction of information on abiotic factors directly and save a lot of work in conducting measurements and chemical analyses [5, 6]. Plants as bio-indicators have been widely used in agriculture, forestry, and environmental monitoring, since plant indication can be easily detected by field observations of distribution or apparent morphological traits [7-9]. A number of studies have documented plant indicators in response to environmental gradients, especially those having a limiting effect on plant growth and distribution, such as soil acidity, soil moisture content, and nutrient availability [10, 11]. In contrast, few studies have documented plants indicative of soil organic carbon (SOC) gradient, since the effect of SOC on plants may be intermediate and can often be compensated for by other factors (such as soil porosity and nutrient status), though SOC plays an important role in improving soil fertility and physical properties.

SOC is important not only for its function in improving soil physical structure and nutrient availability, but also for its role in stabilizing the carbon cycle. A change in SOC content influences not only soil moisture and nutrient availability, thus affecting plant growth [10], but also emissions

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of carbon dioxide from the soil system to the atmosphere [11, 12]. The soil organic carbon pool is heavily dependent on vegetation and land use patterns [13, 14]. Unlike mosses and lichens, which have generally been well recognized as bio-indicators for soil, atmosphere, water, and environmental change [2, 15, 16], most vascular plants have quite extensive ecological magnitude. However, vascular ground vegetation (vascular plants less than 1 m in height, excluding tree seedlings) has proven to be a good candidate for indicating certain environmental gradients, because ground vascular plants of graminoids, forbs, shrubs, and ferns are not deeply rooted and will be heavily affected by the surface soil factors [5, 19].

As one of the most essential factors for the growth of ground vegetation, SOC was emphasized and widely studied recently, but less is known about the quantitative indication of vascular plants to SOC content. A widely used system for revealing plant indication is the Ellenberg indicator value, which assigns each plant species an indicator value (IV) for each environmental factor [6], whereas a new analytical technique, indicator species analysis (ISA), combines information of a species' relative abundance within plots and frequency of occurrence in samples within plots, and calculates an indicator value for each plant species tested for statistical significance using a randomization technique. In this study, we used ISA to determine the response of understory ground vegetation to SOC gradient, and eventually identify indicator plant species of different SOC regimes, which will have potential applications in monitoring soil nutrient and soil carbon pool dynamics.

Material and Methods

Study Area

This study was conducted at Sanchading Nature Reserve (23º24′~23º28′N, 111º59′~112º03′E) in south China's western Guangdong Province. The region is characterized by a southern subtropical monsoon climate and obvious wet and dry seasons, with an average annual temperature and precipitation of 21.5ºC and 1502.4 mm, respectively. Landform type of the study area is mainly low-middle mountains, with the highest point at 700 m a.s.l. and the lowest at 120 m a.s.l. The soils are comprised of lateritic red soil and mountain red soil.

Sampling Design and Plant Census

Three 1-ha plots were set up within the forest vegetation of the study area, representative of a 1-year-old *Eucalyptus urophylla* plantation, a 3-year-old *Eucalyptus urophylla* plantation, and a secondary broadleaved forest, which is dominated by *Schefflera octophylla*, *Lithocarpus corneus*, *Cyclobalanopsis chungii*, *Lithocarpus longanoides*, *Machilus chinensis*, *Lithocarpus glaber*, and *Symplocos adenophylla*. Each plot consisted of 100 contiguous quadrats (10 m×10 m). Vascular ground plants were censused using 5 sub-quadrats (2 m×2 m) placed in each of the 100-m^2 quadrats, 4 in the corners and 1 in the center. All plants rooted within the 5 sub-quadrats were recorded by species name, number of stems and percentage cover, and the data were pooled to represent a quadrat. Most of the species were identified on site except for those of uncertain identity, in which case voucher specimens were collected for further identification in the herbarium of the South China Agricultural University (CANT). Plant systematics follows Ye and Peng [20].

Soil Sampling and Determination

A pit was excavated in the centre of each quadrat to obtain a 30 cm×30 cm soil profile. Soil samples taken from the 0~25 cm soil layer were air-dried, ground, and sieved to determine soil organic carbon content using the dichromate oxidation (external heat applied) method [21]. Average soil organic carbon (SOC) content was further coded into 3 groups as follows: 1 stand for the low SOC content (< 20 g·kg^{-1}), 2 for the medium SOC content (20~30 g·kg^{-1}), and 3 for the high SOC content (> 30 g·kg^{-1}).

Importance Values and Diversity Index

Importance value (I.V.) of understory species was calculated according to the equation:

$$I.V.\ (\%) = RA_i + RC_i + RF_i$$

...where RF_i, RC_i, and RA_i stand for relative frequency (RF), relative coverage (RC) and relative abundance (RA) of the i-th species, respectively.

Species richness (S) and Shannon-Weiner diversity index (H') for understory ground vegetation were calculated using the following equation:

$$S = \text{number of species in specific plot}$$

$$H' = \sum P_i \ln P_i$$

...where P_i stands for the proportion of the number of the i-th species to the total number of species in the j-th plot.

Two-Way Indicator Species Analysis (TWINSPAN)

Understory data were classified using two-way indicator species analysis (TWINSPAN). This analysis detects one or several indicator species that are specifically good dividers to diagnostically divide the community into different clusters of ecological types or quadrats of similar vegetation [22]. The resulting clusters objectively reflect the major environmental gradient and diminish the effect of personal experience on the cluster results. Variability in SOC content among the TWINSPAN groupings was then assessed using the Kruskal-Wallis test.

Table 1. Dominant understory species of three forest stands.

Species	RA_i	RC_i	RF_i	*I.V.* (%)
1-year Eucalyptus plantation				
Dicranopteris dichotoma	30.59	25.39	3.49	59.46
Miscanthus sinensis	11.92	12.61	3.49	28.02
Rhus chinensis	5.33	8.85	3.35	17.52
Sapium discolor	5.55	7.92	3.49	16.97
Litsea cubeba	5.82	6.52	3.49	15.82
3-year Eucalyptus plantation				
Miscanthus sinensis	30.89	30.83	3.5	65.22
Dicranopteris dichotoma	20.88	19.08	3.36	43.33
Microstegium vagans	19.63	7.59	2.1	29.32
Rhus chinensis	2.61	5.93	3.5	12.04
Litsea cubeba	2.36	4.02	3.5	9.89
Secondary broadleaved forest				
Sinobambusa tootsik var. laeta	14.0	14.0	2.0	30.0
Adiantum flabellulatum	8.0	9.0	2.0	20.0
Rourea microphylla	6.0	6.0	2.0	15.0
Evodia lepta	5.0	5.0	2.0	12.0
Schefflera octophylla	4.0	4.0	2.0	10.0

RA_i – Relative abundance, RC_i – Relative cover, RF_i – Relative frequency, *I.V.* – Importance value, expressed as *I.V.* (%) = RA_i+ RC_i+ RF_i

Multi-Response Permutation Procedures (MRPP)

Heterogeneity in understory species composition in response to SOC gradient was tested using multi-response permutation procedures (MRPP) analysis. MRPP is distribution-free and non-parametric statistics that test the difference among two or more groups in a data matrix. This method has no requirement for homogeneity of variance, and consequently is widely applied in ecological research [23, 24]. MRPP output is composed of a test statistic (T), a measure to describe the separation between the groups, a P-value, and the chance-corrected within-group agreement (A), a descriptor of within-group homogeneity, and a measure of effect size. When all the items within groups are identical, then the observed $\delta = 0$ and $A = 1$ (the highest possible value for A). If within-group heterogeneity equals expectation by chance, then $A = 0$. Otherwise, if within-group agreement is less than that expected by chance, $A < 0$. Common values for A are below 0.1 in community ecology research [22]. A squared Euclidean coefficient was used to calculate the distance matrix.

Indicator Species Analysis (ISA)

The faithfulness of the understory species to a particular SOC gradient was detected using indicator species analysis (ISA), which is usually performed as a natural companion to MRPP. Two data matrices were used when performing indicator species analysis. The main matrix contains species composition data, while the secondary matrix has a categorical variable for soil organic carbon content (high, medium, and low). The analysis combines information of species relative abundance (RA) and relative frequency (RF) to a specific group, and the indicator values (IV) is calculated by the equation $IV = 100 \times (RA \times RF)$ for each species in each group. The indicator values range from 0 (no indication) to 100 (perfect indication) and was computed using the Dufrêne and Legendre method [25]. Detailed computation procedures for indicator species analysis can be found in the PC-ORD software documentation [22].

Multivariate analyses and the calculation of indicator values and diversity index were performed by PC-ORD 5.0 and the Kruskal-Wallis test was performed using Statistica 8.0.

Results and Discussion

Dominant Understory Species of Three Forest Stands

The five most dominant understory species of each forest stands are shown in Table 1. *Dicranopteris dichotoma* and *Miscanthus sinensis* showed obvious dominance in both 1-year and 3-year eucalyptus planted forests. These 2 pioneer plant species are able to occupy most of the understory resource niche, and consequently further influence the understory environment. However, in the natural secondary forest, some shade-tolerant plant species, including *Rourea microphylla* and *Adiantum flabellulatum*, had greater importance value, and the five most dominant understory species were different from those occurring in the 1-year or 3-year eucalyptus forest.

TWINSPAN-based Analysis of SOC Content

TWINSPAN was first proposed by Hill as a numerical method for community classification [26]. This method was employed to classify 75 plots of the Sanchading Nature Reserve into 5 groups among 3 division levels that corresponded to the 3 different forest stands. The 25 plots of natural secondary forest, in which species composition significantly differentiated from that of Eucalyptus planted forests, were separated in the D2 (division-2) level. The remaining 50 plots were further divided into 1-year and 3-year Eucalyptus planted forests according to their species composition.

Previous research on the effects of understory of Chinese fir plantation on soil productivity [27] showed that the forest understory provided various positive effects. Its participation in the forest nutrient cycling consequently increased the content of the nutrient elements and organic matter in key soil horizons, promoted nutrient availability, and ultimately contributed to the maintenance and recovery of soil fertility. Significant difference ($p<0.05$) of SOC content was detected among the 5 TWINSPAN groupings of sample units (Fig. 1), indicating that understory plant species composition had a specific effect on SOC content.

Species Diversity and Composition in Relation to SOC Content

In order to further investigate the effect of SOC regime on understory plant distribution, understory plant patterns were assessed for their differences on 3 levels of SOC content, i.e. low (< 20 g·kg^{-1}), medium (20~30 g·kg^{-1}), and high (> 30 g·kg^{-1}). No significant difference was found in the total number of understory individuals, but significant difference ($p < 0.05$) existed in species richness and Shannon-Wiener diversity index (Figs. 2 and 3). Both species richness and diversity index showed an increasing trend with the increase of SOC content. Significant difference was found between low and high, and between medium and high levels of SOC content, and there was no significant difference between low and medium levels of SOC content.

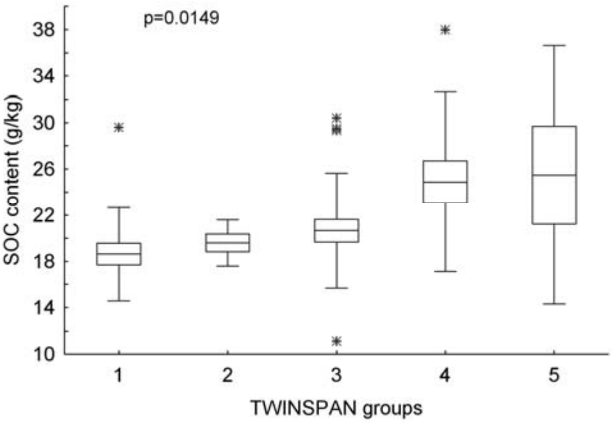

Fig. 1. Variation in SOC content among TWINSPAN groups.

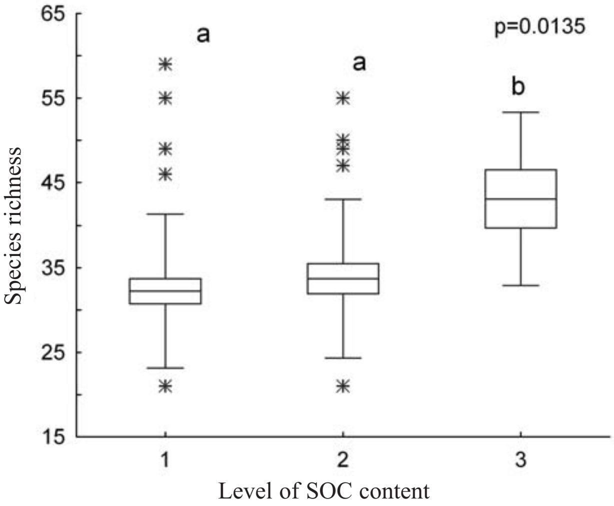

Fig. 2. Species richness in response to SOC gradient.

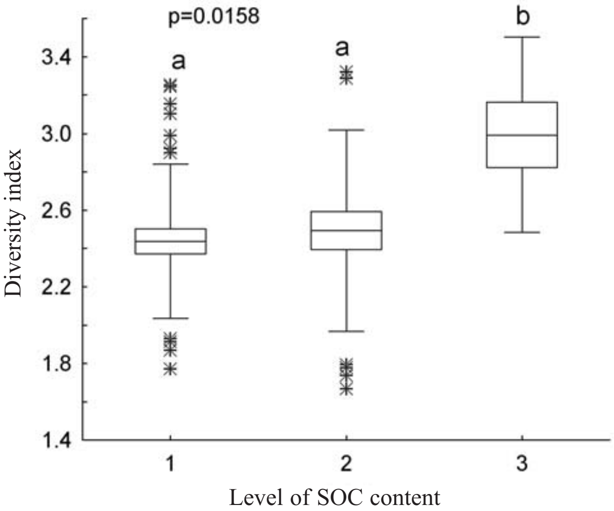

Fig. 3. Shannon-Wiener diversity index in response to SOC gradient.

Indicator Species Analysis

A highly significant difference (MRPP, $p<0.01$) was found in understory species composition across different SOC content levels (Table 2). The MRPP "effect size" (*A*) indicated moderate within-group homogeneity [26].

Indicator species analysis can detect ecological preference of certain plants for a particular environmental gradient. 20 understory plant species were notable indicators of SOC regime, with significant indicator values (*IV*) ≥ 25

Table 2. Multi-response permutation procedures for understory species composition across SOC content levels.

Variable	Observed delta	Expected delta	Variance	Skewness	*T*	*P*-value	*A*
SOC	0.6904	0.7133	<0.0001	-1.4402	-4.6101	0.0019	0.0321

Table 3. Indicator species of understory plants for different SOC regimes.

Species	Growth form	SOC content		Observed indicator value	*P*-value
		Group	Indicative meaning		
Smilax glabra	climber	3	High	54.9	0.002
Rourea microphylla	climber	3	High	49.9	0.005
Millettia dielsiana	climber	3	High	49.2	0.005
Ardisia elegans	shrub	3	High	47.1	0.001
Sinobambusa tootsik	graminoid	3	High	46.7	0.01
Adiantum flabellulatum	fern	3	High	43.7	0.031
Lophatherum gracile	graminoid	3	High	43.1	0.008
Maesa japonica	shrub	3	High	39.2	0.001
Evodia lepta	shrub	3	High	38.7	0.015
Lygodium flexuoum	fern	3	High	37.9	0.022
Glochidion eriocarpum	shrub	3	High	34	0.014
Tetracera asiatica	climber	3	High	31.3	0.048
Embelia laeta	climber	3	High	30.8	0.026
Alpinia chinensis	forb	3	High	30.5	0.012
Alpinia katsumadai	forb	3	High	29.6	0.007
Sarcandra glabra	forb	3	High	27.5	0.01
Ardisia crenata	shrub	3	High	26.6	0.011
Ardisia mamillata	forb	3	High	25.8	0.025
Glochidion wrightii	shrub	3	High	25.5	0.018
Inula cappa	forb	1	Low	29.6	0.04

(Table 3). 19 out of the 20 indicator species were indicative of high SOC content, including *Smilax glabra* (*IV* > 50). Only 1 understory species was indicative of low SOC content, i.e. *Inula cappa*. No indicator species was found indicative of medium SOC content. It was inferred that indicator species was indicative of a specific range of SOC content.

The indicator value in indicator species analysis combines information of species relative abundance and relative frequency, and is a strict standard for detecting the capacity of different species for indicating environmental conditions [26]. The ecological preference of certain species for soils with high or low organic carbon content may have underlying an biological mechanism for the indicating capacity [9, 19], irrespective of forest stands.

Conclusions

SOC content varied significantly across TWINSPAN groups, indicating patterns of understory ground flora were closely related to SOC regimes. Although no significant variation was found in the understory species abundance among different SOC content levels, species richness and diversity index, which were capable of comprehensively reflecting community structure, were significantly different under different SOC regimes. 19 understory species were detected as sensitive indicators for soils with high SOC content, and 1 species was indicative of low SOC content. The dominant understory plant species varied from stand to stand, but indicator species showed ecological preference for the SOC gradient, irrespective of stands. Plant indicators for SOC gradients, as detected in this study, can be used in soil fertility diagnosis and will provide important information for forest managers, especially in subtropical China, for silviculture and forest conservation.

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