Effects of Urbanization Intensity on Glomalin-Related Soil Protein in Nanchang, China: Influencing Factors and Implications for Greenspace Soil Improvement

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Abstract

Glomalin-related soil protein (GRSP) is a stable and persistent glycoprotein secreted by arbuscular mycorrhizal (AM) fungi that plays important roles in sequestering soil organic carbon (SOC) and soil quality improvement. Rapid urbanization has led to serious greenspace soil disturbances, resulting in soil degradation. However, few researches have examined the effects of urbanization on GRSP and its influencing factors. In this study, impervious surface area (ISA) was selected as an indicator of urbanization intensity. A total of 184 soil samples were collected from the 0-20 cm soil layer in the Nanchang greenspace, China (505 km^2). The GRSP content, soil properties, urban forest characteristics, and land-use configuration were determined and investigated. The results showed that total GRSP (TG) and easily extractable GRSP (EEG) averages were 2.38 and 0.57 $mg \cdot g^{-1}$, respectively. TG and EEG decreased by 16.22 % and 19.68 %, respectively, from low to heavy urbanization areas. Linear regression analysis revealed a negative correlation between SOC and GRSP/SOC. SOC decreased from 39.9 to 1.4 $mg \cdot g^{-1}$, while EEG/SOC and TG/SOC increased by about 17 % and 34 %, respectively, indicating the important contribution of GRSP to the SOC pool. Pearson and redundancy analysis showed that GRSP was positively correlated with soil SOC, P, N, vegetation richness, and tree height but negatively correlated with pH, bulk density, and impervious area. The partial least squares path model (PLS-PM) further showed that urbanization affected soil properties, forest characteristics, and land use factors leading to GRSP changes. This study revealed the effects and key influencing factors of urbanization on GRSP. In the future, urban greenspace soil improvement can be considered from the new perspective of enhancing GRSP soil content.

1. Introduction

In the context of global urbanization, the urban population has increased sharply, and the proportion of the global population living in urban settings has exceeded 55% (https://population.un.org/wup/). This has led to a rapid shift from natural and agricultural land to urban land, with significant impacts on ecosystem functions and services (Xie et al. 2019). Urban greenspace is a crucial part of the urban ecosystem and plays irreplaceable roles in enhancing the ecological environment, city beautification and safeguarding human health (Su et al. 2011). Soil is the basis of terrestrial ecosystem function and plays crucial roles in hydrological cycle regulation, plant nutrient supply, and waste disposal. (Daily et al. 1997). However, in the process of rapid urbanization, greenspace soil has been severely disturbed by humans, leading to a series of problems, such as soil degradation, soil compaction, and reduced soil fertility (Wang et al. 2019). Understanding the aboveground (vegetation composition and environmental factors), and belowground components (soil factors) of urban greenspace systems, and their interaction mechanisms is crucial for the effective protection and management of urban greenspace soil (Bauer et al. 2017; Wang et al. 2020b).

As an important soil component, soil microorganisms are major participants in the formation, transformation, and turnover of soil organic matter and play a vital role in soil ecosystems (Bardgett and Putten 2014). Arbuscular mycorrhizal (AM) fungi are one of the most important soil microorganisms, accounting for 30% of the total soil microorganism population (Olsson et al. 1999). It can form symbiotic relationships with nearly 90% of terrestrial plant roots and provide benefits to plant growth, in the form of nutrient transportation, providing water, and improving stress resistance (Nadeem et al. 2017; Smith and Read 2008). Glomalin-related soil protein (GRSP) is a very stable glycoprotein that is secreted into soil mainly by the degradation of AMF hyphae and spores (Driver et al. 2005), and it contributes to improved soil structure and enhanced fertility(Singh et al. 2020). As is outlined in Wu et al. (2014) GRSP can be divided into two fractions: total GRSP (TG, stable protein accumulated in soil) and easily extractable GRSP (EEG, freshly produced protein) using a citrate and autoclave extraction method. GRSP can be detected in various ecosystems, such as farmland, grassland, wetland, and forest, because of the widespread distribution of AMF. It is worth mentioning that GRSP is an important component of the soil organic carbon (SOC) pool, which can accumulate in soil for a long period because of its hydrophobic and non-degradable properties (Gao et al. 2019; Rillig et al. 2001; Wang et al. 2017a). Moreover, GRSP is effective in improving soil structure by bonding smaller soil particles to create larger ones, and increasing the stability of soil aggregates (Udayakumar et al. 2021). In addition, GRSP can fix heavy metals in soils through a combination of functional groups, such as carboxyl, hydroxyl, and carbonyl groups (Gujre et al. 2021). In particular, AMF can produce more GRSP to improve plant tolerance under stress conditions (Naheeda et al. 2021).

Currently, most GRSP-related studies have focused on forest, farmland, grassland, and wetland ecosystems (Banegas et al. 2020; Valerie and Ladislav 2019; Wang et al. 2018b), while few researches have focused on urban greenspace ecosystems. Moreover, most studies have focused on degraded soil ecosystems, using GRSP to indicate and remediate degraded soils, such as saline, grassland, desert,, and soils contaminated by mining (Li et al. 2021; Zhang et al. 2017c). In previous investigations, relevant members of our research group conducted a preliminary GRSP study in urban greenspaces in the black soil area of Northeast China. We found that the average GRSP content reached 6.19 $mg \cdot g^{-1}$, and urbanization significantly reduced the contribution of GRSP to the SOC pool (Wang et al. 2020a). Compared with other ecosystems (1.8-4.5 $mg \cdot g^{-1}$) (Banegas et al. 2020; Zhang et al. 2017c), the GRSP content of urban greenspace in Northeast China is relatively high, which may be closely related to the high fertility of black soil (Udayakumar et al. 2021). Wang et al. (2013) showed that GRSP plays a pivotal role in the organic matter composition of barren soils. Therefore, it is meaningful to select relatively poor soil conditions to carry out research on GRSP restoration of degraded soil.

Red soil is formed by subtropical bioclimatic conditions, which are mainly distributed in hilly areas, south of the Yangtze River in China. The parent material is Quaternary red clay with a high surface gravel content that is susceptible to erosion and soil degradation due to rainfall (Sun 2011). Nanchang is the capital of Jiangxi Province, China, with a high degree of urbanization and a typical red soil (Chen 2013). Due to strong disturbances from urban activities and urbanization processes, most of the red soil has become compacted, poorly structured, deficient in organic matter and nutrients, which has threatened the sustainable development of urban ecology in the region (Chen et al. 2014). This study considers the urban greenspace soil in Nanchang as research objectives and explores possible methods to improve soil quality from the perspective of GRSP. Consequently, the goals of this study are as follows: (1) to clarify the effect of urbanization intensity on the characteristics of GRSP content; (2) to explore the correlations between GRSP and environmental factors (soil properties, urban forest characteristics, and land uses); and (3) to propose future insights for enhancing the quality of degraded urban soils from the perspective of GRSP.

2. Materials And Methods

2.1 Study area

The study area is located in Nanchang City, Jiangxi Province, China (28°10'-29°11'N, 115°27'-116°35'E). Nanchang has a humid subtropical monsoon climate. The precipitation and annual average temperature are 1700 mm and 17°C, respectively. Hot and humid climatic conditions provide a suitable environment for the formation and development of red soil, which is the most prominent soil type in Nanchang (Chen 2013; ISRIC 2015). The resident population is about 5.6 million, and the urbanization rate of the resident population is 75.16%. The common tree species are *Cinnamomum camphora* (camphor), *Pinus* (pine) and *Cunninghamia lanceolata* (fir), among which camphor is the official city tree of Nanchang (SBON 2020).

2.2 Soil sampling

Soil samples (n=184) were collected in June 2020 in the built-up area of Nanchang (505 km^2). To reduce errors caused by plant species, each green space sample plot was placed where the official tree camphor was the dominant tree species, with a minimum interval of 1 km between sample plots. The sample plots were all 400 m^2 in size (Fig. 1). A 5-point sampling method was used for each plot, where five soil cores were collected from 0-20 cm (100 cm^3 cutting ring). Then five soil cores were completely mixed, and the fresh weight was recorded. Soil samples were air-dried to a constant weight in the laboratory. Plastics, coarse debris, and unwanted materials were removed before the experiment. All soil samples were sieved for further analysis.

2.3 Urbanization intensity identification

In our study, spectral mixture analysis was used to determine the impervious surface area (ISA) of Nanchang City (Zhang et al. 2017a). This study region was split into 100 m × 100 m grids. The impervious surface area (ISA) in the grid was used as the urbanization intensity index (Hutyra et al. 2011). Urbanization intensity based on the ISA value for each grid, including low urbanization areas (ISA<0.5), medium urbanization areas (0.5≤ISA<0.8), and heavy urbanization areas (ISA≥0.8) (Wang et al. 2020a).

2.4 Land uses classification and forest characterization

The GPS location for each sample plot was recorded and a circular buffer zone was established with a radius of 100 m. Google Earth images were used to extract different land uses classification data in the buffer zone, and then the percentage of road areas, building areas, greenspace areas and water areas in each buffer zone to the total area was calculated. The results of these calculations were used as land use factors and were divided into three categories: impervious (IM), vegetation (VE), and water (WA) (Zhang et al. 2017a).

During the field survey, information on plants in the 400 m^2 sample plots was recorded as forest characteristics, including woody species, number, woody height (WH), diameter at breast height (DBH), woody crown width (CS), herbaceous species and area. These data were used to calculate woody diversity within a sample plot and were expressed as the Shannon-Wiener diversity index (WSWI). In addition, tree density (TD), herbaceous cover (HC), and herb richness (HR) were also recorded.

2.5 Determination of GRSP

Total GRSP (TG) and easily extracted GRSP (EEG) extractions were performed as described by Wright and Upadhyaya (1998). For EEG, 0.5 g soil samples (particle size of 0.2 mm) were suspended in 4 mL of 20 $mmol \cdot L^{-1}$ sodium citrate (pH=7.0) and autoclaved for 30 min at 121°C. The supernatants were isolated by centrifugation at 4000 rpm for 6 min. TG was removed from 0.5 g (particle size of 0.2 mm) of the soil by adding 4 mL of 20 $mmol \cdot L^{-1}$ sodium citrate (pH=8.0) and autoclaving for 1 h at 121°C. The supernatants were isolated by centrifugation at 4000 rpm for 6 min. For TG, each sample was sequentially autoclaved for 30 min at 121°C until the typical reddish-brown color disappeared. Quantification was performed using the Bradford protein assay with bovine serum albumin as a reference standard. Moreover, the contribution of GRSP to SOC was quantified in terms of the GRSP to SOC ratio (Gujre et al. 2021).

2.6 Soil physicochemical properties determination

Soil pH was measured with a water-soil ratio (2.5:1) and measured by the pH meter (FE20, Mettler Toledo, Shanghai). Soil electrical conductivity (EC) was measured with an EC meter (DDS-307, Shanghai Precision Scientific Instruments Co., Ltd., Shanghai, China). Soil organic carbon (SOC) content was determined using the external heating potassium dichromate volumetric method. Soil total nitrogen (TN) was determined using the Kjeldahl method. Soil available phosphorus (AP) was extracted using $NaHCO_3$ and measured by the molybdenum blue method (UV–5550, Shanghai Metash Instruments Co., Ltd., Shanghai, China). Soil total phosphorus (TP) was determined using the NaOH fusion-molybdenum antimony colorimetric method. Soil total potassium (TK) was determined using NaOH melting-flame photometer. Nitrate nitrogen (NO_3^-) and Ammonium nitrogen (NH_4^+) were determined using phenol disulfonic acid colorimetry and indophenol blue colorimetry. Bulk density (BD) was calculated as the ratio of soil dry weight to soil volume (100 cm^3, cutting ring). Soil moisture content (MC) was measured using the 105°C drying method. Soil physical and chemical properties were determined using the method described by Bao (2000).

2.7 Statistical analysis

The Shapiro-Wilk test was used to determine if the data were normally distributed and non-normally distributed data were log transformed. Significant differences were analyzed using the Duncan's test in SPSS 22.0. The "corrplot" package in R was used for Pearson's correlation test, and Canoco5 was used for redundancy analysis (RDA). The experimental data were expressed as mean ± standard error.

Partial least squares path modeling (PLS-PM) was used to further identify potential pathways for the direct and indirect effects of urbanization on GRSP. The model was constructed by the SmartPLS 2.0 software. The PLS-PM model in this study contains three latent variables: soil factors (SOC, TN, TP, TK, AP, pH, EC, MC, BD, NH_4^+, NO_3^-), forest characteristics (WSWI, TD, WH, DBH, CS, HC, HR), and land uses (IM, VE, WA). The significance of the path coefficients in the model was determined by 1000 iterations of bootstrap resampling (Deng et al. 2018).

3. Results

3.1 The effect of urbanization intensity on GRSP content

GRSP contents of different urbanization intensities are shown in Figure 2. Average EEG and TG were 0.57 $mg \cdot g^{-1}$ and 2.38 $mg \cdot g^{-1}$, respectively. Both were the highest in the low urbanization areas, and were significantly higher relative to the heavily urbanized areas ($p<0.05$). EEG decreased from 0.62 to 0.5 $mg \cdot g^{-1}$, and TG decreased from 2.59 to 2.17 $mg \cdot g^{-1}$, a reduction of 19.68% and 16.22% from low to heavily urbanized areas, respectively.

3.2 Differences in soil factors, forest characteristics and land use factors under different urbanization intensities

Regarding soil physicochemical properties, SOC, TN, and MC were significantly higher in low urbanized areas than in other urbanized areas ($p<0.05$) (Table 1). In contrast, pH, EC, and BD were significantly lower in low urbanization areas than in heavily urbanized areas ($p<0.05$). However, TP, TK, AP, NH_4^+, and NO_3^- showed no significant differences among the three urbanization areas.

Table 1
Differences in soil factors, forest characteristics and land use factors under different urbanization intensities.

Factors	Urbanization intensity		
	Low	**Medium**	**Heavy**
Soil properties			
SOC ($mg \cdot g^{-1}$)	18.64±0.96 a	15.24±0.86 b	15.02±1.39 b
TN ($mg \cdot g^{-1}$)	0.98±0.04 a	0.84±0.03 b	0.82±0.04 b
TP ($mg \cdot g^{-1}$)	0.73±0.04 a	0.67±0.02 a	0.61±0.03 a
TK ($mg \cdot g^{-1}$)	9.75±0.31 a	9.67±0.37 a	9.65±0.52 a
AP ($mg \cdot g^{-1}$)	23.09±2.16 a	25.46±2.27 a	21.91±3.83 a
pH	6.76±0.09 b	7.09±0.09 ab	7.29±0.16 a
EC ($\mu S \cdot cm^{-1}$)	107.95±5.41 b	116.07±4.77 ab	126.66±6.94 a
MC (%)	21.47±0.76 a	18.84±0.80 b	18.14±0.88 b
NH_4^+ ($mg \cdot kg^{-1}$)	10.59±0.63 a	8.89±0.78 a	8.82±0.91 a
NO_3^- ($mg \cdot kg^{-1}$)	4.05±0.35 a	4.81±0.45 a	4.29±0.60 a
BD ($g \cdot cm^{-3}$)	1.31±0.01 b	1.35±0.01 ab	1.39±0.02 a
Forest characteristics			
TD ($Trees \cdot m^{-2}$)	0.09±0.01 b	0.10±0.01 b	0.13±0.01 a
WSWI	1.22±0.09 a	1.28±0.11 a	1.16±0.18 a
WH (m)	9.49±0.43 a	8.31±0.28 a	6.81±0.41 b
DBH (cm)	24.44±0.97 a	24.37±0.94 a	20.80±0.94 b
CS (m^2)	24.20±1.70 a	21.49±1.62 ab	16.80±2.42 b
HC (m^2)	98.77±13.07 a	70.49±10.74 a	73.71±22.48 a

Note: Different letters mean 5% significant differences. Abbreviations: Electrical conductivity (EC), Soil organic carbon (SOC), Total nitrogen (TN), Total phosphorus (TP), Available phosphorus (AP), Total potassium (TK), Ammonium nitrogen (NH_4^+), Nitrate nitrogen (NO_3^-), Bulk density (BD), Moisture content (MC), Woody Shannon Wiener diversity index (WSWI), Tree density (TD), Woody height (WH), Diameter at breast height (DBH), Woody crown size (CS), Herb coverage (HC), Herb richness (HR), Impervious (IM), Vegetation (VE), Water (WA).

Factors	Urbanization intensity		
	Low	**Medium**	**Heavy**
HR	2.16±0.24 a	1.85±0.22 ab	1.22±0.29 b
Land use			
VE (%)	69.73±1.97 a	36.28±1.02 b	10.34±1.89 c
IM (%)	26.26±1.54 c	63.48±1.03 b	89.57±1.90 a
WA (%)	4.01±1.64 a	0.24±0.12 a	0.09±0.09 a

Note: Different letters mean 5% significant differences. Abbreviations: Electrical conductivity (EC), Soil organic carbon (SOC), Total nitrogen (TN), Total phosphorus (TP), Available phosphorus (AP), Total potassium (TK), Ammonium nitrogen (NH_4^+), Nitrate nitrogen (NO_3^-), Bulk density (BD), Moisture content (MC), Woody Shannon Wiener diversity index (WSWI), Tree density (TD), Woody height (WH), Diameter at breast height (DBH), Woody crown size (CS), Herb coverage (HC), Herb richness (HR), Impervious (IM), Vegetation (VE), Water (WA).

In the case of forest characteristics (Table 1), TD increased by 31% from low to heavy urbanization areas, while WH, DBH, CS, and HR were significantly higher in low urbanization than heavy urbanization areas ($p<0.05$). WSWI and HC showed no differences among the three urban areas.

For the land use factors (Table 1), IM was lower in heavy urbanization areas than in other urbanization areas, while VE showed the opposite trend.

3.3 Contribution of GRSP to urban soil carbon pool

SOC content decreased significantly with increasing urbanization ($p<0.05$) (Table 1). Linear regression analysis showed a significant positive correlation between SOC and GRSP ($p<0.01$) (Fig. 3a, b). To further understand the contribution of GRSP to the SOC pool, the GRSP/SOC ratio and SOC were used for linear regression analysis, and were found to be negatively correlated. The mean values for TG/SOC and EEG/SOC were 15.89 and 3.94%, respectively. SOC decreased from 39.9 to 1.4 $mg \cdot g^{-1}$, the EEG/SOC ratio increased from 0.95 to 17.6% (Fig. 3c) and the TG/SOC ratio increased from 6.69 to 41.39% (Fig. 3d).

3.4 Pearson correlation analysis

Relationships between environmental variables and GRSP content were determined by Pearson correlation analysis (Fig. 4). SOC was positively correlated with TG and EEG ($p<0.01$) and the correlation coefficients were 0.67 and 0.53, respectively. In addition, TN, TP, WSWI, WH, HR, and VE were significantly positively correlated with EEG and TG ($p<0.05$), while TK, pH, BD, and IM showed significant negative correlations with EEG and TG ($p<0.01$). DBH and CS were positively correlated with TG levels. However, EC, MC, TD, HC, and WA were not significantly correlated with GRSP content. In general, more than 80% of soil factors were significantly correlated with GRSP, suggesting that soil factors play an important role in GRSP changes.

3.5 Redundancy analysis and variation partitioning analysis

We used GRSP (EEG, TG) contents as response variables and environmental factors as explanatory variables for RDA and variation partitioning analysis. The RDA results showed that the first axis explained 53.5% of the GRSP differences, the second axis explained 4.4% of the GRSP differences, and the cumulative explanation was 57.9% (Fig. 5). The main environmental factors related to the change in GRSP content were SOC, pH, CS, and TK, with explanatory degrees of 36.6%, 9.5%, 5.6%, and 3.3%, respectively. Most of the environmental variables, including CS, WSWI, VE, DBH, WH, HR, NO_3^-, SOC, TN, and TP were positively correlated with GRSP and negatively correlated with TK, BD, IM, and pH. In addition, EEG, TG, SOC, and TN contents were higher in low urbanization areas than in other areas, and pH was higher in heavy urbanization areas.

Variation partitioning analysis showed that soil properties explained the highest GRSP variation (64.4%), followed by forest characteristics (5.6%) (Fig. 6). In addition, the interaction among the three factors (d+e+f+g) explained 30.8% of the GRSP variation (Fig. 6d-g).

3.6 PLS-PM model analysis

The results of the PLS-PM model showed that different latent variables had different effects on GRSP (Fig. 7a). The direct effect of urbanization intensity on GRSP was negative but not significant (p>0.05), with a standardized path coefficient of -0.04 and a total indirect effect of -0.1, indicating that the indirect effect is more significant than the direct effect for GRSP. The indirect effect of urbanization intensity on GRSP was significant (p<0.05) through land use and forest characteristics, with an indirect effect factor of -0.05. It also indirectly affected GRSP through soil properties. Among the latent variables, soil properties had the greatest effect on GRSP, with a standardized path coefficient of 0.55. The factor loadings in each latent variable are shown in Figure 7b, where factors with loadings greater than 0.4 are substituted, and they explain most of the latent variables.

4. Discussion

4.1 Urbanization indirectly decreases GRSP

This study is the first to reveal the urban spatial distribution characteristics of GRSP in red soil in southern China. The average contents of EEG and TG in the 0-20 cm soil layer were 0.57 and 2.38 $mg \cdot g^{-1}$ (n=184), respectively. These values were lower than those in temperate forests, grasslands, and tropical rain forests, but higher than those in poorer ecosystems, such as farmland and deserts (Singh et al. 2013; Treseder and Turner 2007). Wang et al. (2020a) found that the average contents of EEG and TG were 0.56 and 6.19 $mg \cdot g^{-1}$ in black soil in Changchun, Northeast China. Comparatively, the average contents of EEG were similar, but TG was much higher in black soil than was found in this study. A possible explanation is that red soil is less fertile and has lower SOC and nutrients than other soil types, such as black soil (Wang et al. 2015b), resulting in a lower GRSP content. Climate may also play a role in regional variations of

GRSP soil content. Rillig et al. (2010) found that GRSP decomposes more rapidly at higher temperatures, and there is a large difference in climate between the southern and northern parts of China. The average annual temperature in the north is relatively low (4°C), whereas it is reaches 17°C in the south (Chen 2013). The relatively lower temperature environment may allow for a slower turnover rate of GRSP secreted by AMF in soil, which can be preserved longer and thus lead to a higher TG content. In addition, the magnitude of GRSP content is influenced by various factors, such as net primary productivity, vegetation type, and soil properties, etc. (Jiˇrí et al. 2020; Udayakumar et al. 2021). Additionally, GRSP is also affected by human activities, such as soil compaction, household waste, and other anthropogenic disturbances (Gujre et al. 2021). Therefore, the factors affecting the changes in GRSP characteristics in urban greenspaces are more complex than those in other ecosystems.

EEG and TG showed decreases of 19.68% and 16.22%, respectively, from low to heavy urbanization areas (Fig. 2). Interestingly, our analysis revealed that this negative impact was caused by indirect effects (Fig. 6a), consistent with the results of Wang et al. (2018c) in a study of a northern city. This may be due to the following reasons. First, GRSP is a sensitive indicator of the soil carbon pool, closely related to carbon, nitrogen, and other nutrients, and its content changes are closely related to soil quality (Jin et al. 2021). In this study, low urbanization areas had high SOC and TN contents (Table 1), resulting in a change in GRSP content. The decrease in soil nutrients caused by urbanization may be due to low vegetation cover in heavy urbanization areas and surface runoff during precipitation events, which can easily cause soil nutrient loss (Qin and Xu 2018). Second, serious anthropogenic disturbances can affect soil microbial activity, affecting GRSP secretion and storage (Udayakumar et al. 2021; Xu et al. 2017). Third, soil compaction and disturbance during urbanization reduced soil porosity and permeability, further reducing the AMF growth on plant roots (Entry et al. 2003), thus affecting the production and turnover of GRSP. Additionally, urbanization increased IM and reduced VE, thus triggering the heat island effect. Temperatures are higher in heavy urbanization areas than in low urbanization areas (Meehl and Tebaldi 2004), resulting in faster decomposition and less accumulation of GRSP in soils. Overall, urbanization has a multifaceted negative impact on GRSP. Generally, in terms of improving urban soil quality, the functional aspects of GRSP require more attention.

4.2 GRSP plays a 0 vital role in increasing SOC sequestration in heavily urbanized areas

GRSP, as the largest mycorrhizal carbon pool (Wang et al. 2018a), accounts for 27% of SOC, while soil humus accounts for only 8%. The carbon contribution of GRSP is 2-24 times greater than that of soil humus (Comis 2002). Pearson correlation analysis and RDA showed that both EEG and TG were positively correlated with SOC, indicating that GRSP can be used as an indicator of the soil carbon pool. Moreover, EEG/SOC and TG/SOC were 3.94 and 15.89% (n=184), respectively, suggesting that GRSP contributes more C to the urban red soil carbon pool. Similar values were also observed by Staunton et al. (2020), who reported an average of 9.7% for TG/SOC and 3.8% for EEG/SOC.

The SOC and GRSP levels were significantly lower in the heavy urbanization areas than in the low urbanization areas (Fig. 2, Table 1). Linear regression analysis showed a negative relationship between SOC and GRSP/SOC (EEG/SOC and TG/SOC) (Fig. 3). We found that the rate of SOC loss was much higher than the decomposition rate of GRSP. This also confirmed that GRSP is a component of inert carbon in the soil carbon pool (Wang et al. 2018b). Hence, increasing GRSP content can reduce soil carbon loss due to urbanization. In addition, GRSP can reduce the decomposition of soil organic matter by binding to soil aggregates and sequestering GRSP-C within aggregates (Liu et al. 2020). Therefore, GRSP has important roles in SOC sequestration, especially in areas with heavy urbanization.

4.3 Key factors affecting GRSP change in the urbanization process

Compared with natural ecosystems, urban ecosystems are dominated by humans. Climate change, animals and plants, and land use pattern are strongly disturbed by human in urban ecosystem (Meng et al. 2021). In contrast to natural ecosystems (Jin et al. 2021), we should consider how various urban environmental factors (forest characteristics, soil properties, and land use configuration) affect GRSP soil content.

Soil properties were the key factors affecting GRSP content during the process of urbanization. Pearson correlation analysis showed that more than 80% of the soil factors were significantly correlated with GRSP content. Variation partitioning analysis showed that soil factors could explain 64.4% of the GRSP differences. In particular, the PLS-PM model showed that soil factors were one of the key indirect factors influencing the decrease in GRSP. For instance, GRSP was significantly and positively correlated with TP, SOC, and TN, confirming that GRSP can be a crucial indicator of the dynamics of soil carbon sequestration and nutrient retention (Wang et al. 2017b). AMF mycelia promote plant growth and improve soil quality by increasing the uptake of soil nutrients in exchange for photosynthetic products from the host plant (Smith and Read 2008). This also creates an important pathway for the transfer of mycorrhizal carbon to and stored in the soil over time (Clemmensen et al. 2013), which can lead to improved soil quality. GRSP was positively correlated with NO_3^- and NH_4^+ concentrations (Fig. 4). GRSP also showed a strong correlation with NO_3^- and NH_4^+, which are the main forms of nitrogen uptake by plants (Zhang et al. 2015). Moreover, GRSP can enhance nitrogen mineralization, and hyphae bridges can also help plants absorb nitrogen from the soil (Terrer et al. 2016). Urbanization significantly increased soil pH from slightly acidic to neutral and then to weakly alkaline (Table 1). This may due to the fact that most urban greenspace soils contain construction backfill, which contains construction waste, cement, lime, and other alkaline substances (Gujre et al. 2021). Consistent with the results of Singh et al. (2016); Wang et al. (2015b), GRSP was negatively correlated with BD and pH. Slightly acidic or neutral soils are favorable for GRSP accumulation, mainly because the optimal habitat for AMF is in slightly acidic soils, whereas alkaline soils are not favorable for spore production and therefore GRSP secretion (Bonfim et al. 2016; Wang et al. 2015a). Previous studies have shown that AMF is an aerophile fungi, and soil with better permeability is more suitable for AMF spores production and increases colonization of plant roots (Sun et al. 2011). In the present study, BD was significantly lower in low urbanization areas than in heavily urbanized areas. Since BD can indicate soil compaction, this result suggests that the reduction in GRSP caused by urbanization may be due to soil hardening and human disturbance, which reduced soil microbial activity and GRSP secretion.

The influence of forest characteristics on GRSP content should not be neglected. Unlike natural systems, urban vegetation is subject to more human modifications and disturbances, such as transplanting and pruning. Urbanization intensity could indirectly influence GRSP changes through forest characteristics (Fig. 7a). In particular, the interaction between forest characteristics and soil factors could explain 24.1% of the GRSP differences (Fig. 6). Correlation analysis and RDA also indicated that WSWI, WH, HR, DBH, and CS were positively correlated with GRSP content. In addition, the PLS-PM model selected these five factors as sufficient indicators of forest characteristics (Fig. 7b), which indicates that these factors had a significant impact on GRSP soil content. WSWI is a measure of woody diversity, which indicates that high vegetation diversity can increase GRSP soil content. This was consistent with the findings of Sousa et al. (2013), who found that the composite planting method can increase GRSP content compared to the traditional monoculture pattern. WH, DBH, and CS are tree growth parameters. Generally, the large and dense root systems of large woody plants facilitate the formation of symbioses between AMF and plant roots, thereby increasing AMF biomass and GRSP content (Gujre et al. 2021; Nautiyal et al. 2019). Trees with large canopies have large shade areas, which not only reduces the negative effects of high temperature and precipitation on SOC sequestration (Zhang et al. 2021), but also increases surface litter, which increases the soil organic mulch, and thus facilitates AMF colonization and GRSP production (Udayakumar et al. 2021). In general, there is a relatively strong correlation between GRSP content and plant biomass (Liu et al. 2020). Interestingly, we found that TD was not correlated with GRSP, and TD increased linearly from low urbanization areas to heavy urbanization areas. This result can be attributed to that most of the heavy urbanization areas are newly built urban areas with small and single tree species, resulting in higher TD. Low urbanization areas are not overdeveloped and built up, and some old trees are preserved, so the WH and DBH of the areas with low urbanization are higher. This phenomenon is consistent with the results of Zhang et al. (2017b).

Land use factors indirectly affected GRSP changes by influencing forest characteristics and soil factors (Fig. 7). The Pearson correlation analysis and RDA together indicated that GRSP content was closely related to the percentage of VE. There are some negative effects on GRSP accumulation in with increasing urbanization because increased impervious surface (e.g. buildings and roads) and a decrease in vegetative cover. Therefore, in the context of rapid urbanization, it is necessary to manage and protect urban greenspace soils by improving the GRSP content.

4.4 Implications and suggestions

China has experienced a rapid and continuous urbanization process over the last few decades and the urban population continues to increase, with an average annual growth rate of 0.16 billion (Guan et al. 2018). Resource-driven population growth in cities threatens limited natural resources by improper disposal of waste, resulting in soil and water pollution (Deus et al. 2020). Arbuscular mycorrhizal fungi (AMF) are ubiquitous soil microbial communities (Smith and Read 2008; Stürmer et al. 2018). GRSP secreted by AMF is a key indicator for maintaining plant growth and improving soil quality (Singh et al. 2020). Therefore, to increase urban soil GRSP content, we put forward the following suggestions and their implications.

Soil factors had the greatest impact on GRSP, and some soil nutrients were positively correlated with GRSP. Due to the development and utilization of land in the process of urbanization, the consumption of soil organic matter also decreased. In this study, nutrients, such as SOC, N, and P, were significantly higher in low urbanization areas than in heavily urbanized areas. Therefore, in heavy urbanization areas, appropriate fertilization can be used in greenspace management to promote plant growth and improve basic soil nutrients. In addition, GRSP was negatively correlated with pH and BD, especially in areas with heavy urbanization. The dumping of construction and domestic waste should be minimized to keep the soil pH slightly acidic, which will be beneficial to AMF growth and sporulation. Urban greenspaces in China are public spaces managed by local governments, and their soil compaction will continue to increase due to human trampling, crushing, etc. To increase soil permeability in heavy urbanization areas, greenspace soil should be loosened regularly to reduce BD. These soil management measures can increase the accumulation of GRSP and contribute to the improvement of soil quality.

Greenspace vegetation plays an increasingly important role in urban areas, greatly contributing to ecosystems and biodiversity (Kendal et al. 2020), and it is essential that greenspaces are properly managed to protect floristic health. The percentage of vegetation should be increased in areas with heavy urbanization, which can lead to an increase in GRSP content. In addition, GRSP is positively correlated with WSWI, WH, DBH, etc. The existing larger trees should be protected and managed, try not to transplant the original tree. Tree species in new urban development areas should be diversified under the premise of a beautification to effectively increase soil GRSP. In addition, the effects of tree species composition impact the organisms living in the soil. How to effectively select different plant ratios to maximize their functions requires further study. In addition, we should adopt a near-natural forest management pattern to manage urban greenspaces and minimize human interference. For example, reducing soil compaction and retaining forest litter can effectively increase the sequestration of GRSP and soil carbon. Overall, we can consider improving the soil quality of greenspace from the perspective of enhancing GRSP content to maintain the sustainable development of urban ecosystems in the future.

5. Conclusions

To the best of our knowledge, this study is the first report on the spatial distribution characteristics of GRSP in the red soils of southern China in an urban setting. We used ISA as an indicator of urbanization intensity to explore changes in GRSP. The increase in urbanization intensity can indirectly cause a decrease in GRSP content through changes in forest characteristics, land use configuration, and soil factors. Moreover, urbanization causes a rapid decrease in SOC content. GRSP, as an inert carbon component, plays a crucial role in stabilizing soil carbon sequestration. Among the different environmental factors, soil factors (SOC, TN, BD, pH) and forest characteristics (WSWI, WH, HR, DBH, and CS) play key roles in influencing the changes in GRSP content. Future management measures should include reducing human trampling, avoiding garbage dumping, increasing greenspace area, improving vegetation diversity, and protecting existing trees to effectively improve urban soil quality by increasing GRSP content.

Declarations

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Conflicts of interest No potential conflict of interest was reported by the authors.

Author contributions Wei Liu and Qiong Wang conceptualized the study, and Taotao Jin collected, processed, and identified samples with help from Ming Zhao, Yu Wang, Tianyu Luo, and Hang Fu. Qiong Wang and Taotao Jin primarily interpreted the data with contributions from Yao Fu and Yulin Dong. Taotao Jin, Qiong Wang, and Wei Liu wrote the manuscript, and all authors were involved in the review, revision and final approval of the manuscript.

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Figures

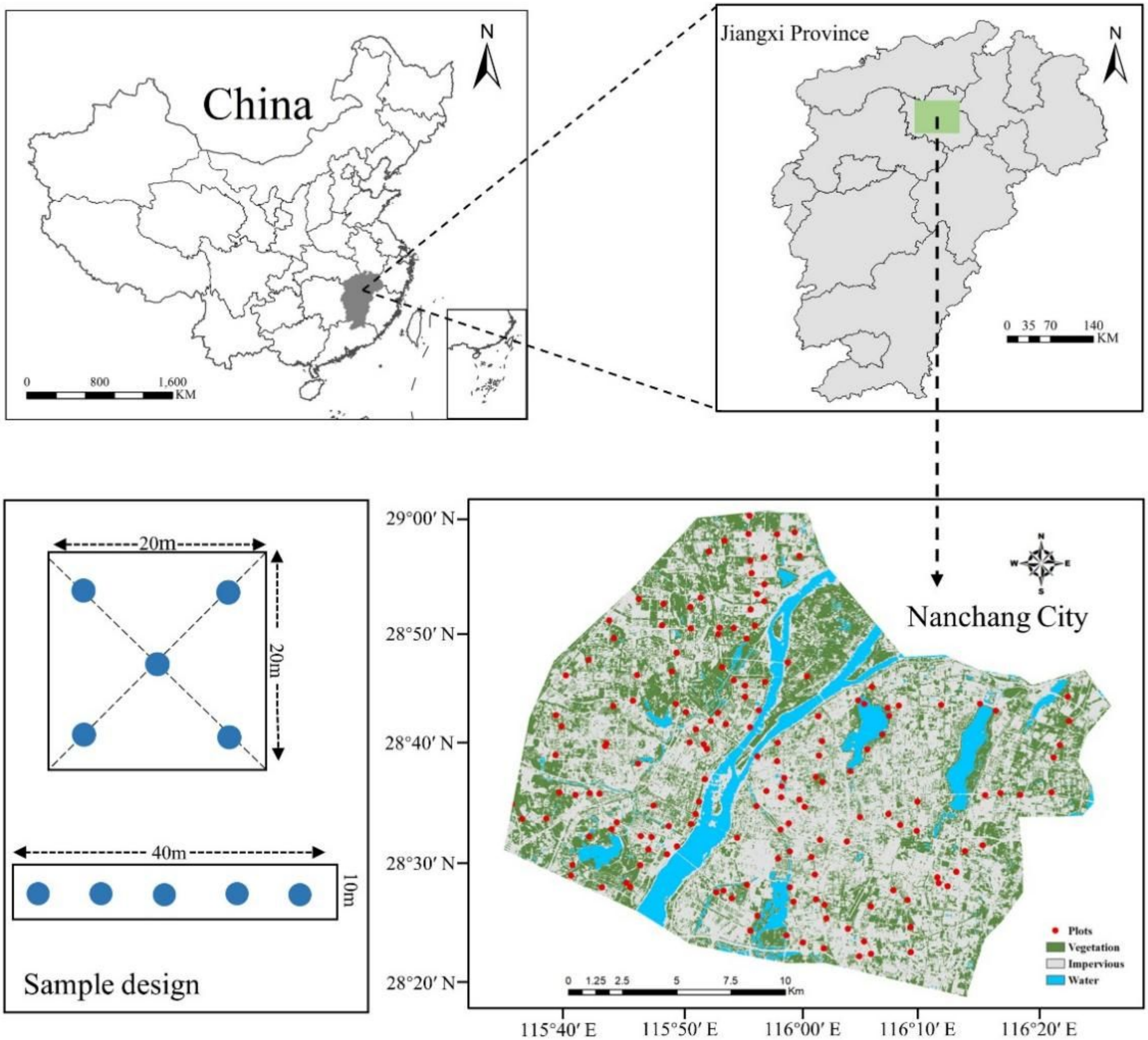

Figure 1

Map of the study area and sampling locations.

Note: Different letters mean 5 % significant differences. Error bars are standard errors (SE).

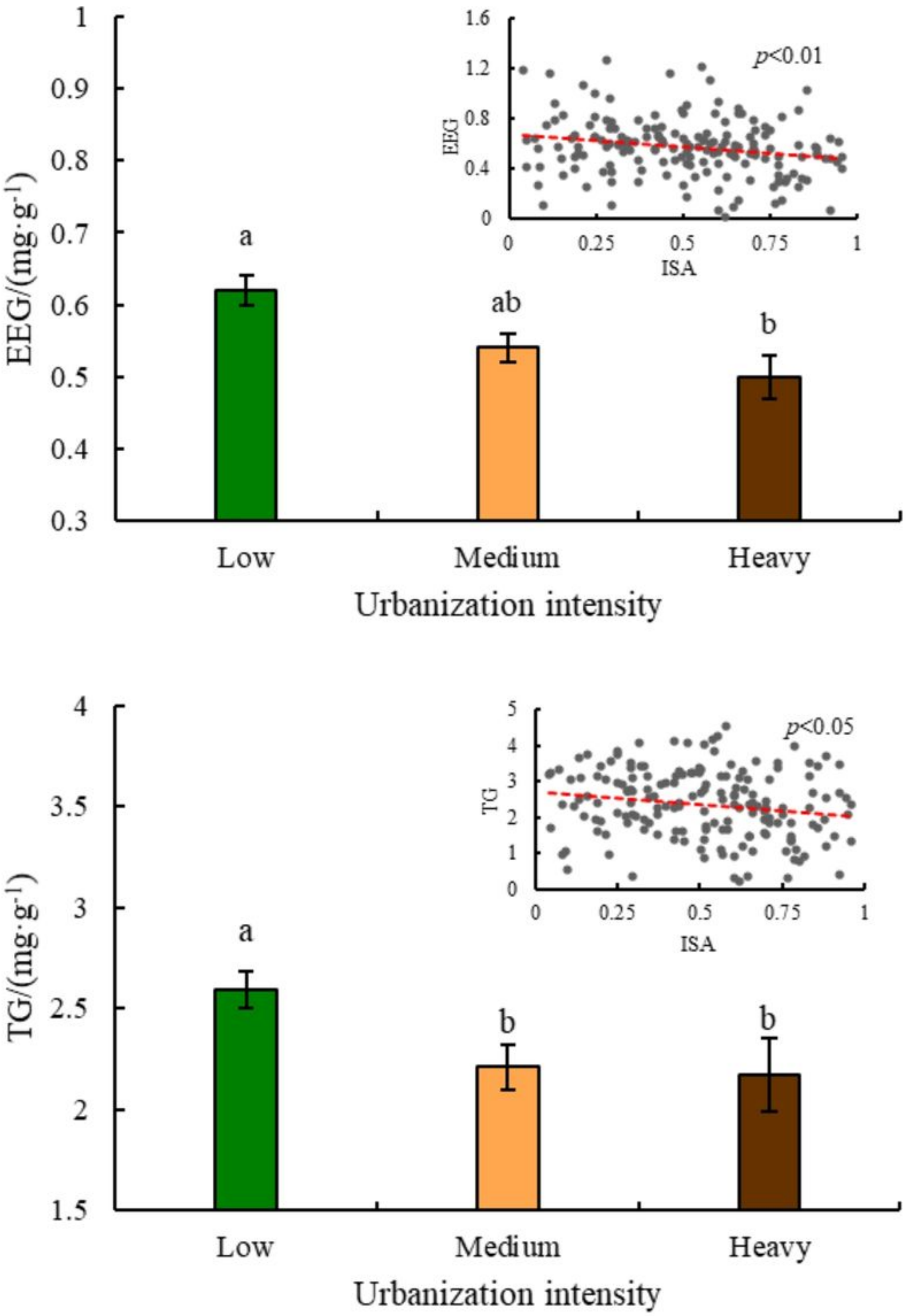

Figure 2

Effects of urbanization intensity on GRSP characteristics.

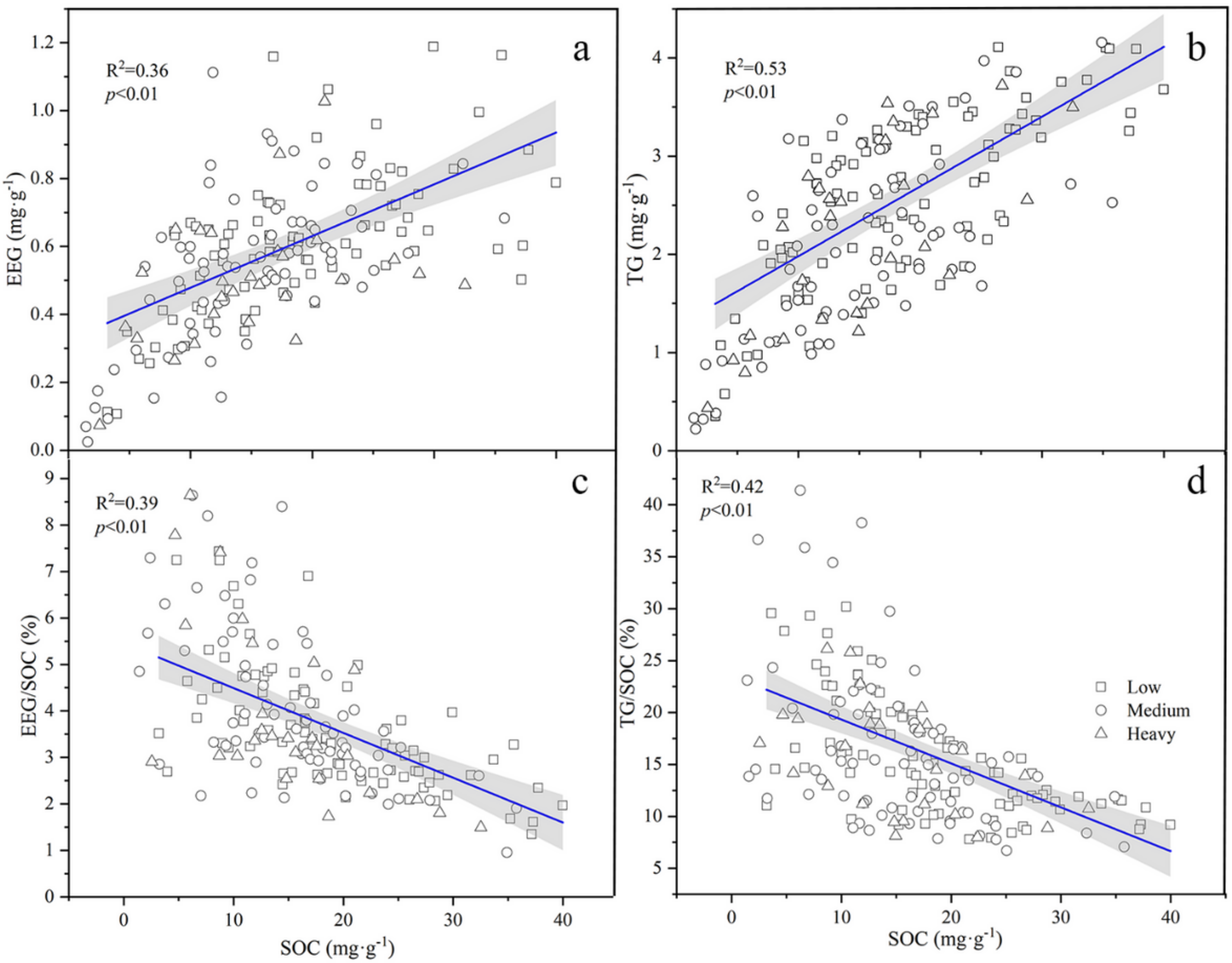

Figure 3

Relationships between SOC and GRSP (GRSP/SOC).

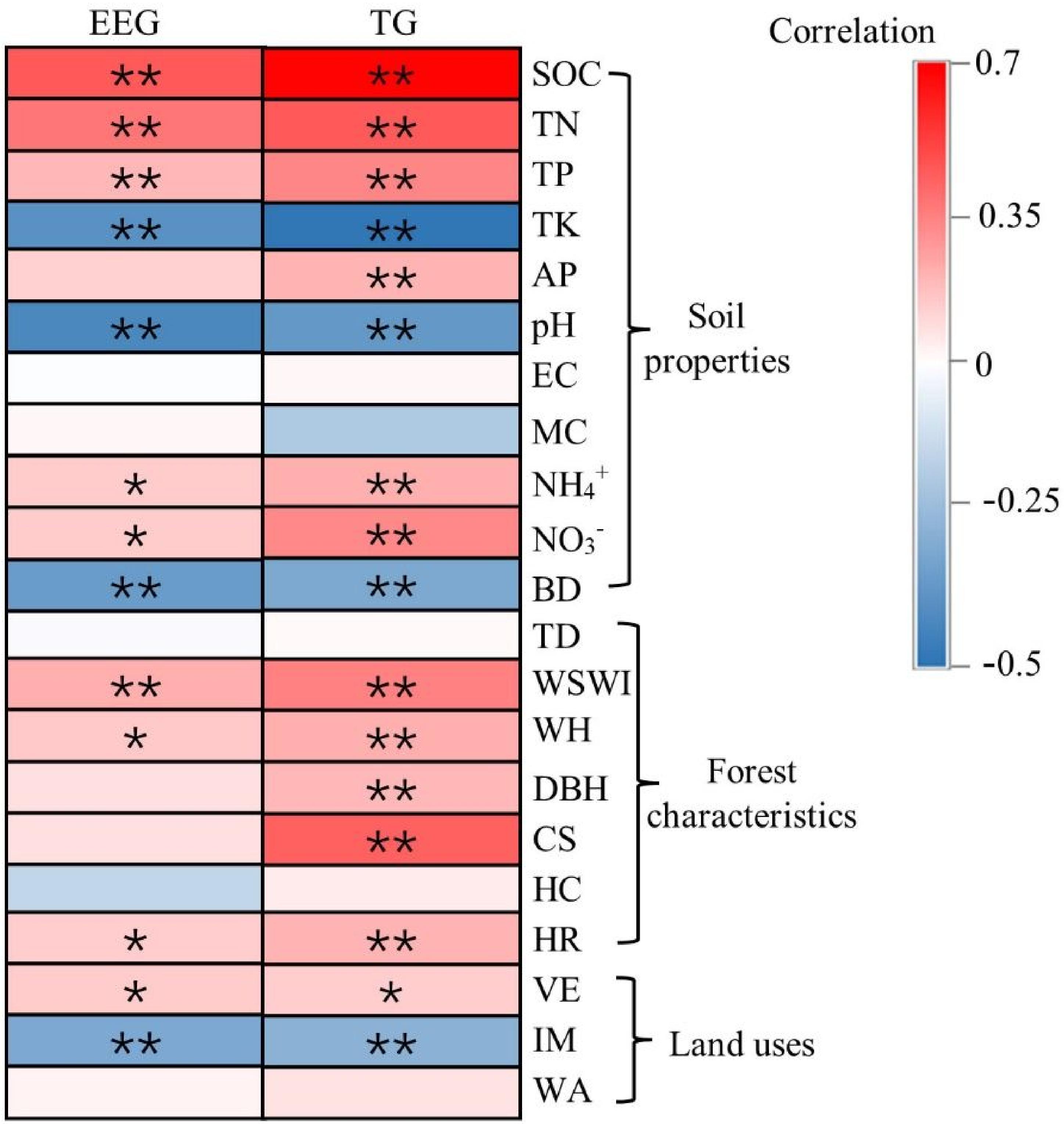

Figure 4

Pearson correlation analysis between GRSP and environmental factors.

Note: "*" mean significant at p<0.05 level. "**" mean significant at p<0.01 level. Abbreviations: Electrical conductivity (EC), Soil organic carbon (SOC), Total nitrogen (TN), Total phosphorus (TP), Available phosphorus (AP), Total potassium (TK), Ammonium nitrogen (NH_4^+), Nitrate nitrogen (NO_3^-), Bulk density

(BD), Moisture content (MC), Woody Shannon Wiener diversity index (WSWI), Tree density (TD), Woody height (WH), Diameter at breast height (DBH), Woody crown size (CS), Herb coverage (HC), Herb richness (HR), Impervious (IM), Vegetation (VE), Water (WA).

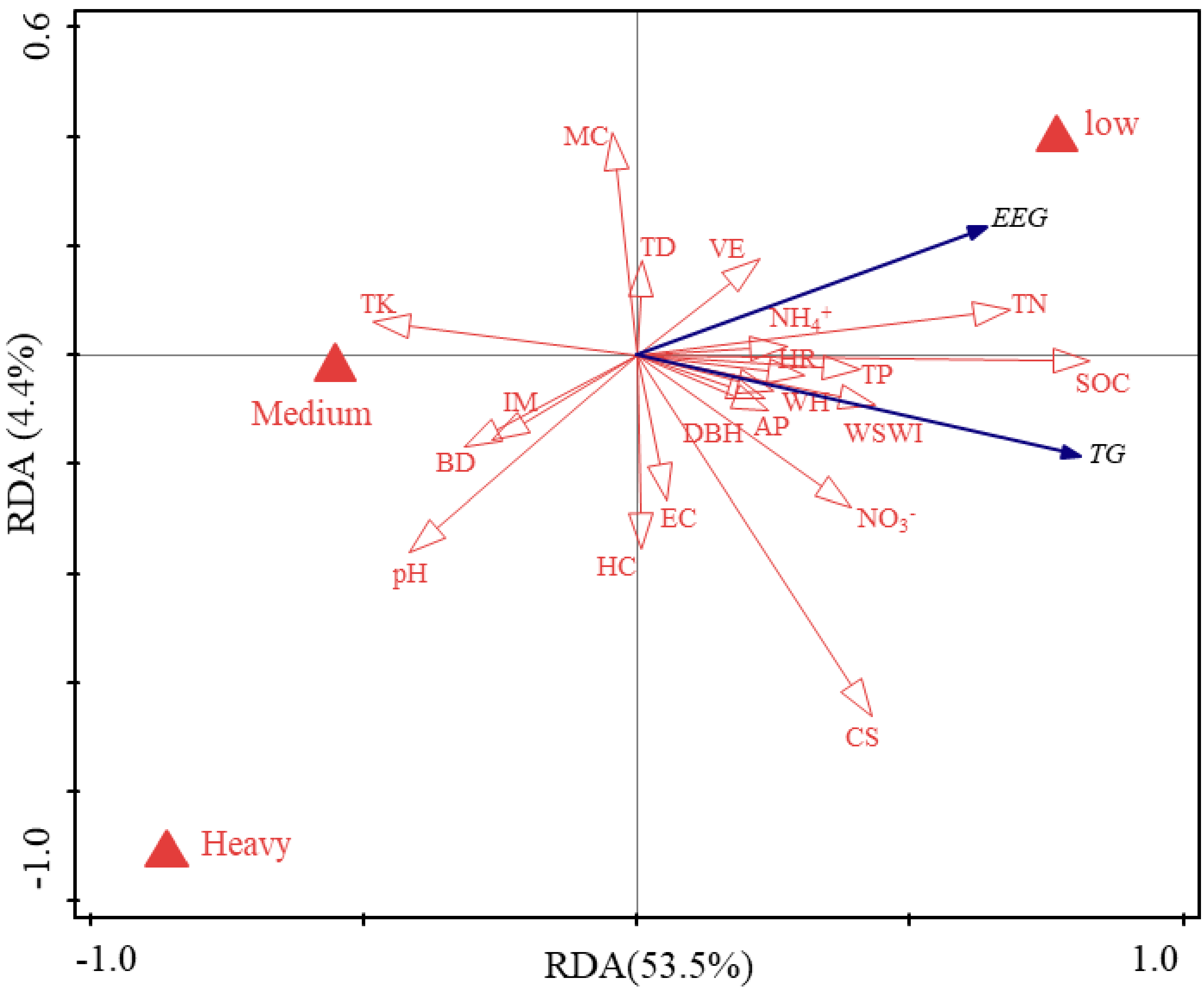

Figure 5

Redundancy analysis of GRSP and environmental factors.

Note: Red lines and arrows point to explanatory variables; blue lines and arrows point to response variables. Abbreviations: Electrical conductivity (EC), Soil organic carbon (SOC), Total nitrogen (TN), Total phosphorus (TP), Available phosphorus (AP), Total potassium (TK), Ammonium nitrogen (NH_4^+), Nitrate nitrogen (NO_3^-), Bulk density (BD), Moisture content (MC), Woody Shannon Wiener diversity index (WSWI), Tree density (TD), Woody height (WH), Diameter at breast height (DBH), Woody crown size (CS), Herb coverage (HC), Herb richness (HR), Impervious (IM), Vegetation (VE), Water (WA).

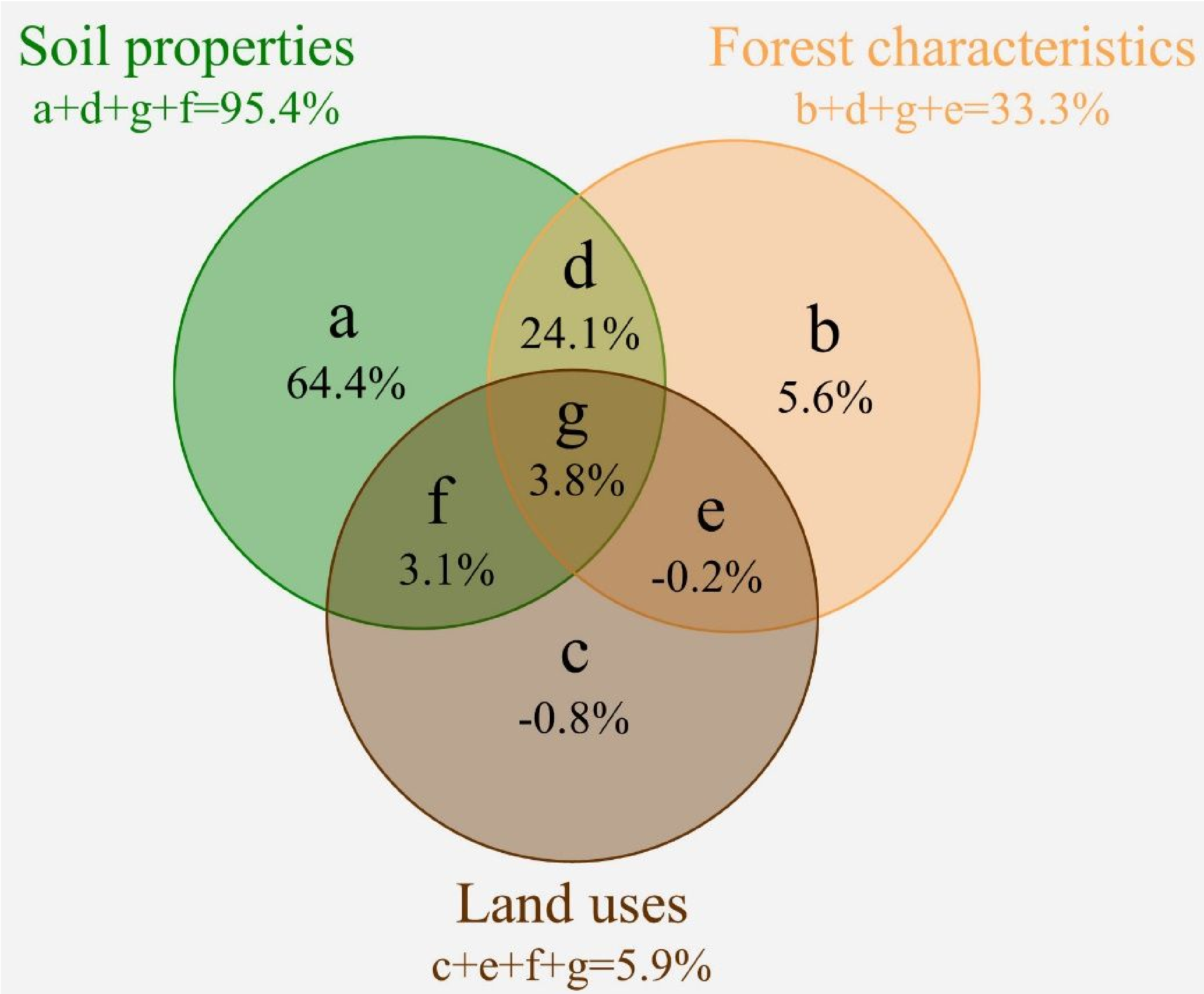

Figure 6

Variation partitioning analysis results in RDA analysis of “Var-part-3groups-Conditional-effects-tested”.

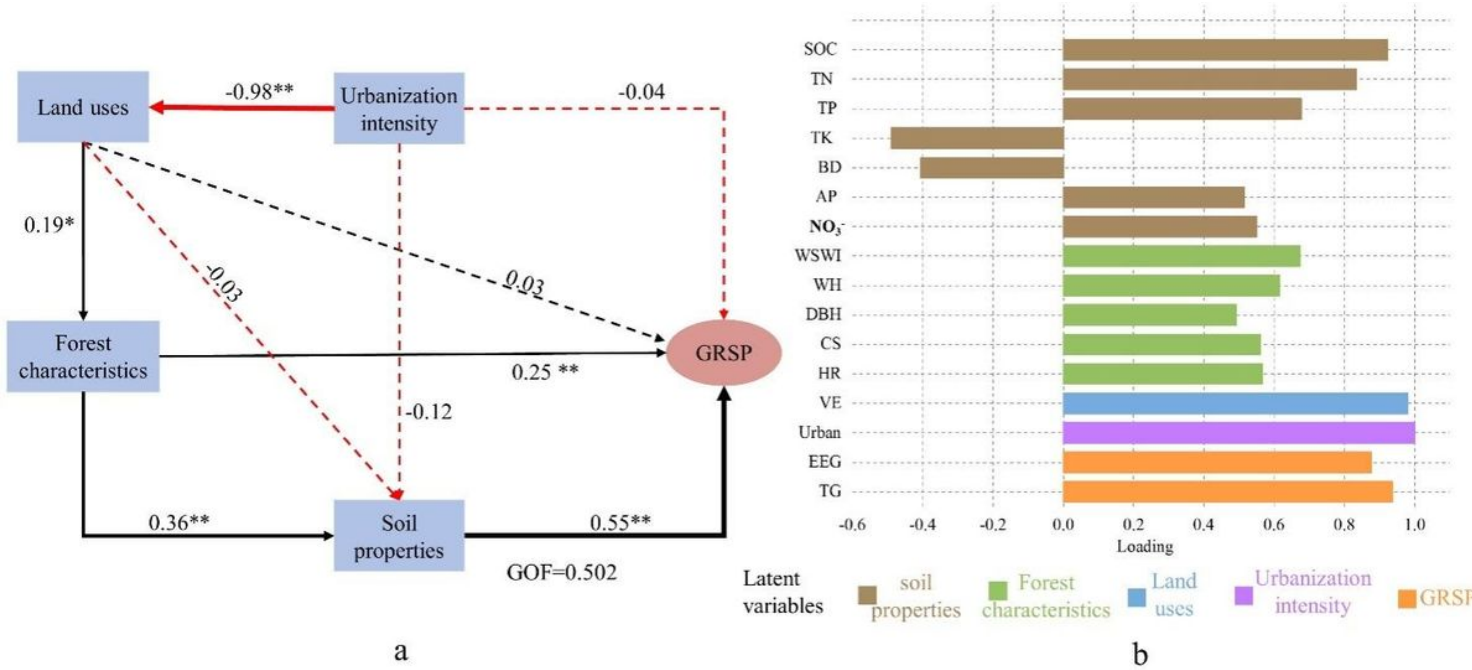

Figure 7

PLS-PM model analysis.

Note: The asterisk, *, indicates that the path coefficients are significant at the 95 % confidence level through 1000 bootstrap resampling; **, indicates that the path coefficients are significant at the 99 %. Abbreviations: Soil organic carbon (SOC), Total nitrogen (TN), Total phosphorus (TP), Available phosphorus (AP), Total potassium (TK), Nitrate nitrogen (NO_3^-), Bulk density (BD), Woody Shannon Wiener diversity index (WSWI), Woody height (WH), Diameter at breast height (DBH), Woody crown size (CS), Herb richness (HR), Vegetation (VE).