



## Original Articles

## Evaluating indices of soil organic carbon stability. A case study for forest restoration projects near Beijing, China

Zeyu Zhang<sup>a,b</sup>, Tonggang Zha<sup>a,b,\*</sup>, Yang Yu<sup>a,b,\*</sup>, Xiaoxia Zhang<sup>c</sup>, Pete Smith<sup>d</sup>, Jesús Rodrigo-Comino<sup>e</sup><sup>a</sup> School of Soil and Water Conservation, Beijing Forestry University, Qinghua East Road 35, Beijing 100083, China<sup>b</sup> Jixian National Forest Ecosystem Research Network Station, CNERN, Beijing Forestry University, Beijing 100083, China<sup>c</sup> The Third Construction Co., Ltd. of China Construction First Group, Beijing 100161, China<sup>d</sup> Institute of Biological and Environmental Sciences, University of Aberdeen, 23 St Machar Drive, Aberdeen AB24 3UU, UK<sup>e</sup> Departamento de Análisis Geográfico Regional y Geografía Física, Facultad de Filosofía y Letras, Campus Universitario de Cartuja, Universidad de Granada, 18071 Granada, Spain

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## ABSTRACT

Afforestation of degraded lands close to mega-urban areas such as Beijing may help to restore some of the original soil carbon stocks and hold the potential for ameliorating the rate of increase in atmospheric CO<sub>2</sub>. However, the determinants of the stability of different soil carbon pools and the utility of indices of stability remain poorly characterized near these highly anthropogenic areas. In the current study, we compared metrics of soil organic carbon (SOC) stability taking into account different soil types and plantation forest combinations (Quartissamment soil-poplar plantation–QP, Eutrochrepts soil-Chinese pine plantation–ECP, Haplustepts soils-East-Liaoning oak plantation–HEO), in an experimental sub-humid area close to a mega-urban area (Beijing, China). We evaluated the following relative stability indices sequence: respired carbon from incubations (RI) for several incubation days to respire 5% of initial SOC (D), aggregate stability index (ASI), the ratio of SOC to total nitrogen (C: N), water-soluble carbon (WSC), particulate organic carbon (POC) and microbial biomass carbon (MBC). We examined the indices by three repeated measurements on soil samples from four soil layers (0–40 cm) in three soil-forest types in a forest area close to the peri-urban area of Beijing. Our results showed that there are inconsistencies among the six SOC stability indexes. The contribution rates of different indexes to the SOC in three plantations were different, for QP the highest contributor is WSC (54.73%), and for ECP and HEO the highest contributor is RI, contribution rates are 34.85% and 36.382%, respectively. Respired carbon from incubations registered the largest contribution rate to SOC (69.79%), and the correlation between RI and soil physical and chemical properties was the highest. We conclude that a combination of indices and knowledge of soil and vegetation types are needed for assessing SOC stability in restoration and reforestation projects close to mega-urban areas.

## 1. Introduction

Soils are key sources of the Earth's system as they can produce indispensable resources and goods and provide nutrients for natural and human ecosystems (Smith et al., 2020; Yu et al., 2017; Rodrigo-Comino et al., 2020). Some recent estimates confirm that the soil surface (in the top three meters of soil) could contain approximately 2344 Gt (1 gigaton = 1 billion tonnes) of organic carbon worldwide (Ma et al., 2021; Stockmann et al., 2013), which would be by far the largest carbon stock among the different terrestrial ecosystems (Falahaatkar et al., 2014; Li

et al., 2021; Tajik et al., 2020b; Xu et al., 2020; Zeraatpisheh et al., 2022). In natural and anthropogenic ecosystems, the main changes in organic carbon stores are derived from plant carbon and the decomposition of original organic carbon (Yan et al., 2020). In a context marked by potential climate change and rising atmospheric carbon dioxide concentrations coinciding with intensive land-use changes, forest ecosystems and their soils are a significant carbon pool that deserves protection (Angst et al., 2019; Assefa et al., 2020). Some authors estimate that forests account for 80% of the vegetation carbon pools on the Earth's surface and play a key role in the global carbon balance, therefore, net

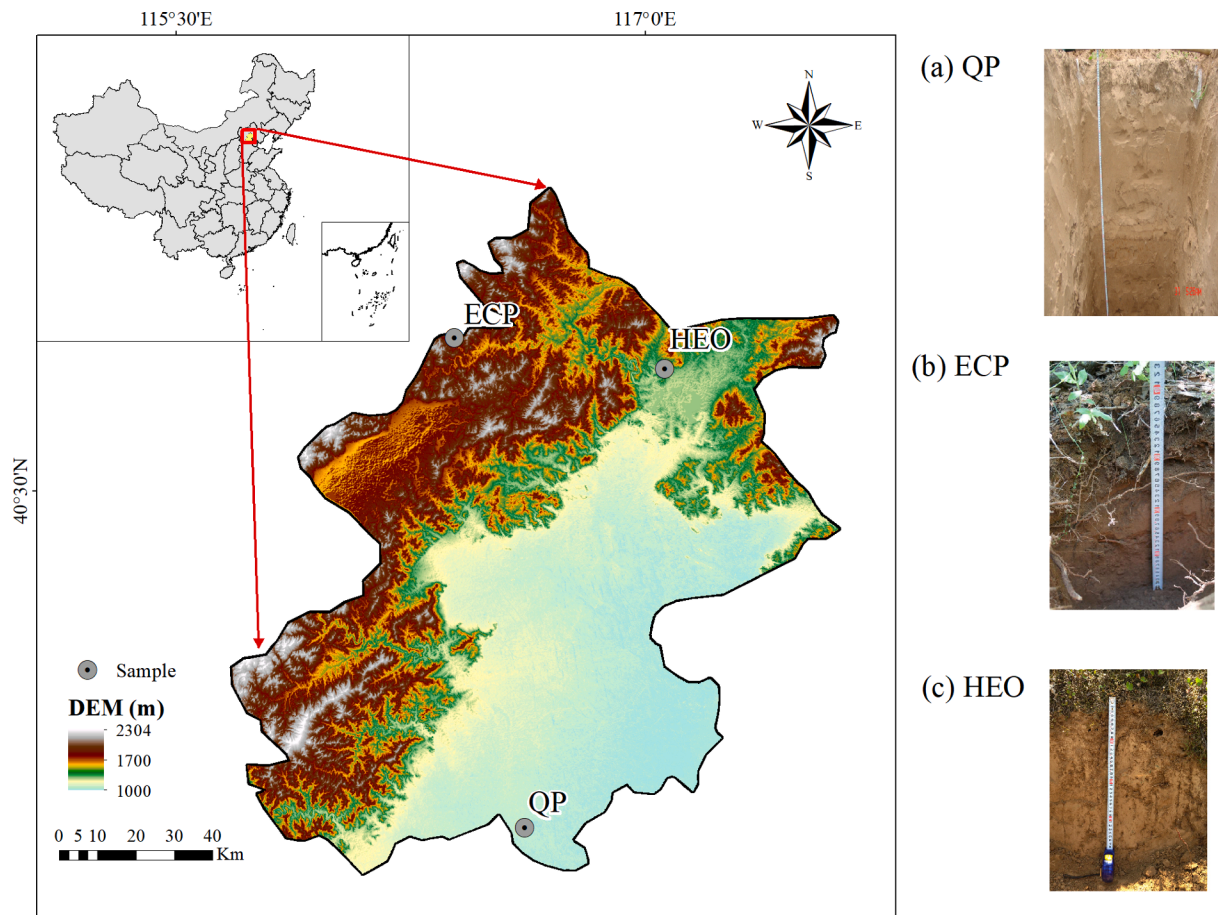
\* Corresponding authors at: School of Soil and Water Conservation, Beijing Forestry University, Qinghua East Road 35, Beijing 100083, China.  
E-mail addresses: [zhtg73@bjfu.edu.cn](mailto:zhtg73@bjfu.edu.cn) (T. Zha), [theodoreyy@gmail.com](mailto:theodoreyy@gmail.com) (Y. Yu).

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**Fig. 1.** Location map of three soil types-plantation forest combinations and soil profiles in Beijing of China. (a) QP- Quartissamment soil poplar (*Populus × eur-america* cv. “74/76”) plantation; (b) ECP- Eutrochrepts soil Chinese pine (*Pinus tabulaeformis*) plantation; (c) HEO - Haplustepts soil East Liaoning oak (*Quercus liaotungensis* Koidz) plantation.

carbon dioxide emissions could be reduced by conducting sustainable and well-planned afforestation actions (Cao et al., 2020; Segura et al., 2020). Afforestation is considered a strategy that is able to i) fix carbon dioxide; ii) prevent soil erosion; iii) restore soil properties (Soil organic carbon-SOC, Total nitrogen - TN, Available phosphorus - AP, Available potassium - AK, Cation exchange capacity - CEC, pH, Carbon to nitrogen ratio - C: N and Bulk density - BD); and, enhance forest biodiversity (Ayoubi et al., 2022; Segura et al., 2020; Yu et al., 2022).

Since the 1990s, exponential afforestation has been widely implemented in many countries (Yu et al., 2020a), increasing the global planted forest area by about  $1.05 \times 10^8$  ha (Hong et al., 2020). The influences of afforestation on soil has also been extensively studied and the indicators are varied. Vesterdal et al. (2012) simulated soil carbon dynamics using laboratory incubated mineral soil carbon turnover of 6 different tree species in Europe. Foote et al. (2015) used soil microbial biomass carbon (MBC), soil microbial biomass nitrogen (MBN), SOC, and TN as indicators to evaluate forest soil productivity in the United States, reflecting the storage and turnover of SOC and nitrogen. Deng and Shangguan, 2017 used SOC, TN, C: N, MBC, MBN, and other indicators to characterize the dynamic changes in soil carbon and nitrogen influenced by afforestation in terms of tree species, and soil depth and age. Angst et al. (2019) used heterotrophic respiration carbon, carbon in aggregate occluded particulate organic matter (POM-C), and mineral-related SOM to study the effects of different tree species on the stability of forest SOC in Poland. In China, Lin et al. (2018) used water-soluble carbon (WSC), MBC, and readily oxidizable carbon indicators to compare the changes in total SOC and unstable organic carbon storage after the natural forest was converted to a plantation forest. Soleimany

et al. (2021) use aggregate stability index (ASI) and POM-C and nitrogen in aggregate occluded particulate organic matter (POM-N) to characterize the positive impact of temperate afforestation in Iran. Chodak et al. (2022) compared SOC, AP, WSC, TN, MBC, and other indicators after afforestation and fire in Poland to obtain the limiting factors affecting soil restoration.

The indexes to measure the stability of SOC can be divided into the absolute stability index and the relative stability-one. Currently, the absolute stability of SOC is mostly measured by the isotope tracing method because it helps to compare the degree of change in SOC and nitrogen (Deng et al., 2016; Jia et al., 2012; Wang et al., 2019b). However, isotope labelling is suitable for small laboratory experiments but is difficult to apply in the field (Stockmann et al., 2013). At the same time, the isotope index of SOM stability is also conditioned by the uncertainty of many factors that affect the isotope ratio (Angst et al., 2019). The relative stability index currently is roughly studied considering soil MBC as the main component of soil activated carbon (Yu et al., 2020c) and ASI as a measure of soil aggregate organic carbon stability. Soil C: N is also one important factor to assess the relevance of SOC sequestration after vegetation restoration (Deng and Shangguan, 2017), which could be combined with the WSC because it impacts soil microbial communities and enzyme activities (Yang et al., 2017). Some authors also consider particulate organic carbon (POC) because it mainly affects the balance between soil carbon input from plant residues and soil carbon loss caused by microbial decomposition (Chen et al., 2020). Although, it is important to highlight that this measure should be combined with soil RI as an indicator of the degree of SOC mineralization (Coonan et al., 2019). Due to the differences in the definition and

**Table 1**  
Main characteristics and descriptors of the three-soil type-plantation forest combinations (2014<sup>†</sup>).

Sites <sup>‡</sup>		QP	ECP	HEO
Geographical and environmental conditions	Location	39°032'N, 116°015'E,	40°044'N, 116°065'E	40°035'N, 116°054'E
	Topography	Plain	Low mountain	Low mountain
	Inclination (°)	3	17	11
	Elevation (m a.s.l.)	30	730	310
Soil	Minimum depth (cm)	100	40	60
	Parent material	Alluvial deposit	Weathered granite	Weathered carbonate
	Soil taxonomy	Quartisamment	Eutrochrepts	Haplustepts
vegetation	Tree species	Poplar	Chinese pine	East-Liaoning oak
	Age (yr)	13	51	51
	Density (tree ha <sup>-1</sup> )	1,667	1,302	1,041
	Height (m) <sup>§</sup>	15.2 ± 1.2	11.6 ± 1.7	16.7 ± 1.6
	DBH (cm) <sup>§</sup> #	13.8 ± 1.1	12.3 ± 1.8	15.3 ± 2.3

<sup>†</sup> All values were determined in January 2014.

<sup>‡</sup> Abbreviation of the sites: QP- Quartisamment soil poplar (*Populus × euramericana* cv. "74/76") plantation, ECP- Eutrochrepts soil Chinese pine (*Pinus tabulaeformis*) plantation, HEO - Haplustepts soil East Liaoning oak (*Quercus liaotungensis* Koidz) plantation.

<sup>§</sup> values are mean ± SD.

composition of these indicators, or the reduced use of a few indicators because of economic reasons or availability of laboratory materials, some results could lead to some uncertainties (Angst et al., 2019).

It would be desirable to use a standardized indicator that could define SOM stability. In this research, considering SOC stability as an ecosystem attribute, the main goals of this research are to i) investigate the relationships of different stability indices within and across different so-called "soil type-plantation forest combinations" (STPFC-s); ii) to identify the best indicators of SOC stability (respired carbon from incubations -RI- or several incubation d to respired 5% of initial SOC, aggregate stability index -ASI-, the ratio of SOC to total nitrogen -C: N-, water-soluble carbon -WSC-, particulate organic carbon -POC- and microbial biomass carbon -MBC-) to define the best management practices that consider the requirements for soil C sequestration. The results of the study will provide the basis for the index selection of evaluating SOC stability in restoration and reforestation projects close to mega-urban areas. To achieve these objectives, we analyzed the landscape-level variability of these various relative stable indices (RSI-s) in three common STPFC-s close to a mega-urban region in the Beijing area, China (Jones, 2002). To date, there are few papers about this topic in China, a country experiencing fast urban sprawl resulting from the economic boom over the past three decades and expected in the coming years.

## 2. Materials and methods

### 2.1. Study area

The current study was conducted considering three typical STPFC-s close to the mega-urban area of Beijing, China (Fig. 1). The region, with a sub-humid continental monsoon climate (Wang et al., 2019a; Zhao and Wu, 2019), is surrounded by the Taihang Mountains on the west and the Yanshan Mountains on the north and east. The climate is temperate, with a mean annual temperature of 9°C and an average of 150 frost-free d per year. The mean annual precipitation is 600 mm, of which 70% falls between July and September. According to the soil survey conducted in 1990, about 75% of the study area (13,700 km<sup>2</sup>) fell in the mountains, and 24.7% in the flatland, the predominant soils in the mountainous region are eutrochrepts (9.5% of the area) and the haplustepts (65%) and along with the flatlands Quartisamment soil (Yang and Zhou, 2007).

There were three primary STPFC-s in this region, poplar (*Populus euramericana* cv. "74/76") on Quartisamment soil (QP) in flatlands, Chinese pine forests (*P. tabuliformis*) on Eutrochrepts soils (ECP) and East-Liaoning oaks (*Quercus liaotungensis* Koidz) on Haplustepts soil (HEO) in the low mountain area. The QP site in this study was a 12-year-old poplar plantation located in the Daxing district at the moment of the

investigation. The parent materials are alluvial deposits coming from the Yongding River nearby. The basement stratigraphy of QP is mainly composed of the Jixian System, Qingbaikouan System, Cambrian, and Ordovician systems. Soil depths average greater than 100 cm (Zha, 2007). The stand density was 1,176 trees per hectare, with a mean tree height of 15.2 ± 1.2 m and a mean tree of 13.8 ± 1.0 cm in 2010.

On the other hand, both the ECP and HEO lie in the Jiufeng National Forest Park (western mountain area) owned by the Beijing Forestry University. The plantations were established in the 1960s as part of the green belt for Beijing (Yang and Zhou, 2007). Rocks include granite, limestone, tuff, sandstone, and shale. The parent material of ECP is weathered granite and average soil depths reach 40 cm. The stand density was 1,302 trees per hectare, with a mean tree height of 11.6 ± 1.7 m and a mean tree of 12.3 ± 1.8 cm. In the HEO, soil depth averages 60 cm, and the parent material was weathered carbonate. The stand density was 1,041 trees per hectare, with a mean tree height of 16.7 ± 1.6 m and a mean tree of 15.3 ± 2.3 cm. Other key site characteristics are shown in Table 1.

### 2.2. Field survey

Three 400 m<sup>2</sup> sampling plots were established at each STPFC-s (20 m × 20 m for QP, 10 m × 40 m for ECP and HEO). Within each plot, soils were sampled at six locations, equidistant along the length of an S-shaped sampling line. Soil samples were taken from four different depths each 10 cm (0–10, 10–20, 20–30, and 30–40 cm) in all plots in May 2011. In addition, six-core samples (5 cm) were taken from each of the six points and mixed at the corresponding layers in one sample to form a bulk sample of about 1 kg. Samples were bagged and air-dried in the laboratory for 3–7 d. Then, they were sieved (2 mm) to remove large roots, stones, and the macro-fauna. Soil samples were also collected using the cutting ring method to determine soil bulk density (BD).

### 2.3. Laboratory analyses

#### 2.3.1. Soil property measurement

About half of each air-dried sample was ground to measure the soil's physical and chemical properties. The moisture content of undisturbed samples was determined by the oven-dry method (105°C for 24 h). Bulk Density, porosity, pH, total nitrogen (TN), and soil cation exchange capacity (CEC) were determined as outlined by Dewis & Freitas (1970). Particle size distribution was estimated by the pipette method. Soils were pretreated with 1 M NaOAc (sodium acetate) at pH 5.0 to remove carbonates and with NaOCl (sodium hypochlorite) at pH 9.5 to digest organic matter, followed by dispersion with a dilute Na-HMP (Sodium hexametaphosphate) solution (Soil Survey Staff, 2004). Following the dispersion procedure, samples were wet sieved at 20 μm, sands (N20 μm

**Table 2**

Soil physical and chemical properties in the three forest combinations. Data are means  $\pm$  SE (n = 3). The same upper case letters indicate no difference between forest combinations and the same lower case letters no difference between soil depth at P = 0.05.

SPC	Soil Layer	BD (g·cm <sup>-3</sup> )	Clay (%)	Sand (%)	Porosity (%)	SOC (g·kg <sup>-1</sup> )	TN (g·kg <sup>-1</sup> )	CEC (cmol <sub>(+)</sub> ·kg <sup>-1</sup> )	pH
QP	0–10 cm	1.38 $\pm$ 0.07Aa	0.02 $\pm$ 0.00Ca	0.92 $\pm$ 0.02Aa	0.48 $\pm$ 0.01Ca	2.63 $\pm$ 0.16Ca	0.17 $\pm$ 0.01Ca	3.22 $\pm$ 0.09Ba	8.01 $\pm$ 0.08Aa
	10–20 cm	1.43 $\pm$ 0.02Aab	0.03 $\pm$ 0.00Ca	0.88 $\pm$ 0.01Aa	0.47 $\pm$ 0.02Cab	1.33 $\pm$ 0.06Cb	0.07 $\pm$ 0.01Ca	2.82 $\pm$ 0.09Ba	7.99 $\pm$ 0.10Aa
	20–30 cm	1.40 $\pm$ 0.02Ab	0.02 $\pm$ 0.00Ca	0.83 $\pm$ 0.02Aa	0.47 $\pm$ 0.01Cab	0.76 $\pm$ 0.05Cb	0.07 $\pm$ 0.01Ca	3.16 $\pm$ 0.33Ba	8.14 $\pm$ 0.02Aa
	30–40 cm	1.41 $\pm$ 0.02Ab	0.02 $\pm$ 0.00Ca	0.82 $\pm$ 0.02Aa	0.48 $\pm$ 0.01Cb	0.75 $\pm$ 0.06Cb	0.07 $\pm$ 0.00Ca	3.31 $\pm$ 0.19Ba	8.11 $\pm$ 0.12Aa
ECP	0–10 cm	0.71 $\pm$ 0.07Ca	0.09 $\pm$ 0.01Ba	0.42 $\pm$ 0.07Ba	0.74 $\pm$ 0.02Aa	21.24 $\pm$ 0.74Aa	1.82 $\pm$ 0.09Aa	16.73 $\pm$ 0.64Aa	5.76 $\pm$ 0.10Ca
	10–20 cm	1.01 $\pm$ 0.08Cab	0.07 $\pm$ 0.00Ba	0.49 $\pm$ 0.04Ba	0.64 $\pm$ 0.02Aab	12.65 $\pm$ 0.34Ab	1.22 $\pm$ 0.09Aa	14.97 $\pm$ 1.00Aa	6.33 $\pm$ 0.31Ca
	20–30 cm	1.13 $\pm$ 0.07Cb	0.12 $\pm$ 0.01Ba	0.32 $\pm$ 0.02Ba	0.54 $\pm$ 0.01Aab	11.36 $\pm$ 0.95Ab	1.22 $\pm$ 0.1Aa	19.03 $\pm$ 2.83Aa	6.59 $\pm$ 0.10Ca
	30–40 cm	1.18 $\pm$ 0.07Cb	0.12 $\pm$ 0.02Ba	0.31 $\pm$ 0.02Ba	0.51 $\pm$ 0.02Ab	10.41 $\pm$ 0.69Ab	1.14 $\pm$ 0.04Aa	19.90 $\pm$ 1.04Aa	6.80 $\pm$ 0.10Ca
HEO	0–10 cm	1.10 $\pm$ 0.01Ba	0.09 $\pm$ 0.00Aa	0.29 $\pm$ 0.01Ca	0.52 $\pm$ 0.01Ba	18.98 $\pm$ 1.08Ba	1.08 $\pm$ 0.07Ba	19.27 $\pm$ 2.38Aa	6.89 $\pm$ 0.15Ba
	10–20 cm	1.16 $\pm$ 0.07Bab	0.14 $\pm$ 0.02Aa	0.20 $\pm$ 0.00Ca	0.58 $\pm$ 0.01Bab	9.86 $\pm$ 0.15Bb	1.03 $\pm$ 0.01Ba	16.00 $\pm$ 1.39Aa	7.03 $\pm$ 0.19Ba
	20–30 cm	1.24 $\pm$ 0.03Bb	0.16 $\pm$ 0.01Aa	0.19 $\pm$ 0.01Ca	0.55 $\pm$ 0.01Bab	6.71 $\pm$ 0.69Bb	0.43 $\pm$ 0.01Ba	15.40 $\pm$ 0.75Aa	7.06 $\pm$ 0.10Ba
	30–40 cm	1.28 $\pm$ 0.12Bb	0.17 $\pm$ 0.00Aa	0.21 $\pm$ 0.01Ca	0.53 $\pm$ 0.02Bb	5.36 $\pm$ 0.40Bb	0.32 $\pm$ 0.00Ba	16.57 $\pm$ 1.74Aa	7.01 $\pm$ 0.14Ba

fraction) collected, and oven-dried (determined by the POC). The remaining silt and clay fractions were collected in 1 l cylinders. Silt and clay were separated by sedimentation and an aliquot of clay was collected and oven-dried.

Total organic carbon (TOC) storage was calculated following the procedure of [Wairiu and Lal \(2003\)](#):

$$TOC = d \times \rho \times C \times 100 \quad (1)$$

where  $d$  is the depth of the soil layer (cm),  $\rho$  represents the BD (g cm<sup>-3</sup>),  $C$  means the carbon concentration (g C g<sup>-1</sup> soil), and 100 denotes the conversion factor to t C ha<sup>-1</sup>.

A part of each sample was air-dried and stored at 4°C, for the following analyses. Water-soluble organic carbon was assayed by stirring samples of soil with distilled water (soil: H<sub>2</sub>O = 1:20) for 24 h at room temperature. Then, the suspension was centrifuged at 10,000 rpm for 10 min, after filtration through a 0.4 mm fiberglass, after which the carbon content was determined by dichromate oxidation titration ([Ciavatta et al., 1991](#)).

Microbial biomass carbon was analyzed using the method described by [Voroney et al. \(1993\)](#). Two aliquots of 20 g of fresh soil were weighed into two 200 ml flasks and 40 ml of 0.5 M K<sub>2</sub>SO<sub>4</sub> (potassium sulfate) was poured into each flask. Then, 1 ml of CHCl<sub>3</sub> (chloroform) was added to one of the two mixtures. The flasks were stoppered and shaken at 200 rpm for 1 h. The filtrate was collected and bubbled with CO<sub>2</sub> free air for 30 s after filtration. Then, 8 ml of the filtrate was transferred to a 150 ml flask, and 0.075 g of HgO (mercury oxide), 2 ml of 0.2 M K<sub>2</sub>CrO<sub>7</sub> (potassium dichromate), 10 ml of concentrated H<sub>2</sub>SO<sub>4</sub> (sulfuric acid), and 5 ml of concentrated H<sub>3</sub>PO<sub>4</sub> (phosphoric acid) was also added into the flask. The mixture was digested at 250°C for 30 min, transferred into a 500 ml flask, and titrated with 0.017 M FeSO<sub>4</sub> (iron sulfate) using ferroin as an indicator. The test was replicated 3 times for each soil sample. MBC was calculated as follows:

$$MBC = (OCF - OCUF) / 0.18 \quad (2)$$

where  $OCF$  is the carbon content in fumigated solution and  $OCUF$  represents the carbon content unfumigated ([Voroney et al., 1993](#)).

Soil aggregate stability was determined by wet sieving according to [ONL 1072 \(2004\)](#). Briefly, soil aggregates with a diameter of 2000–1000  $\mu$ m were dipped in a sieve of 250  $\mu$ m. The mass of soil used in the experiment is 4 g (EW). The mass of stable aggregates after dipping ( $m_K$ ) and the mass of sand after chemical dispersion of the remaining aggregates ( $m_A$ ) were determined. Soil aggregate stability index was calculated as follows:

$$ASI = X/Y \quad (3)$$

where  $X$  is the number of soil aggregates retained on a 250  $\mu$ m sieve after treatment and shaking, and  $Y$  is the total amount of soil aggregates taken for aggregate analysis ([Aziz et al., 2013](#)).

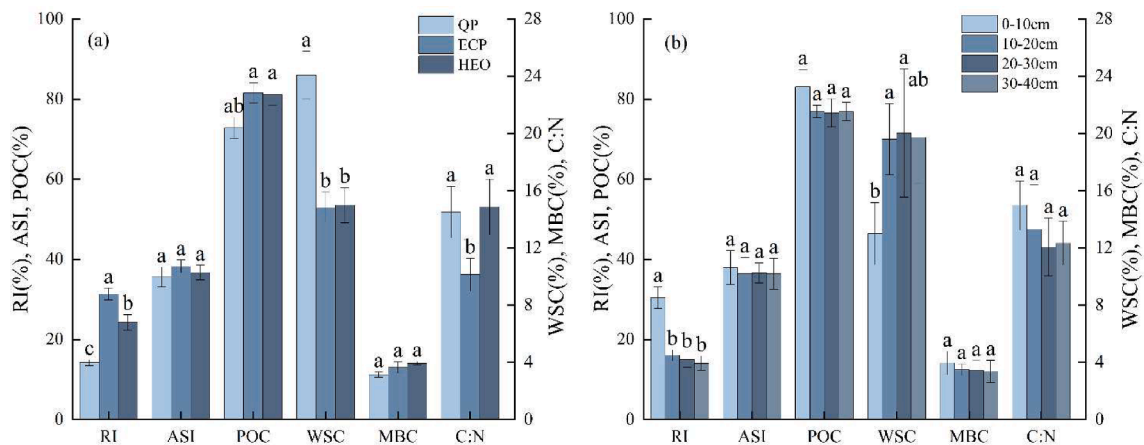
Soil organic carbon and fractional organic carbon contents were determined by dry combustion with an elemental analyzer (Elementar Vario EL III, Elementar, Germany) and expressed as organic carbon mass in the whole soil or fractions to the whole soil mass.

### 2.3.2. Incubation experiment

The incubation experiment was set up similar to [Haddix et al. \(2011\)](#) and [Plante et al. \(2011\)](#). Briefly, collected soil samples were air-dried, passed through a 2-mm sieve, and stored at room temperature until incubations began with natural BD. Four replicate subsamples from the 36 composite field samples were incubated at 25°C for 154 d, the same time duration as the Rh measurement. For each sample, 80 g of soil were rewetted and incubated at 60% water-filled pore space. Samples were placed in sealed canning jars fitted with septa, along with scintillation vials containing 20 ml of water to maintain humidity. Soils were pre-incubated for 7 d at 25°C before any measurement to allow the soil to equilibrate after wetting up ([Paul et al., 2006](#)). Headspace gas samples were analyzed for CO<sub>2</sub> concentration using a LICOR 820 infrared gas analyzer (IRGA) (LICOR Biosciences Lincoln, NE, USA). Jars were flushed with compressed tank air before CO<sub>2</sub> concentrations reached 5% to prevent CO<sub>2</sub> concentration from inhibiting microbial activity. The CO<sub>2</sub> measurements were taken daily during the first week of the incubation, weekly for the next 7 weeks, and then every 4 weeks for the other 7 weeks thereafter, generating a total of 21 sampling times over 154 d. Initial organic C concentrations of samples were determined by dry combustion as mentioned above. The number of incubation d to respire 5% of initial carbon (D) was determined by fitting cumulative respiration data to simple exponential equations and solving for the time given fixed amounts of CO<sub>2</sub> respired. The day is an indicator of the stability of SOC and cannot be compared to other indicators in terms of quantity. Relatively speaking, RI can more representatively express the stability of SOC in terms of a quantitative relationship. So, we selected the RI to TOC at 60 d of incubation as a measure of the stability of SOC.

### 2.4. Statistical analysis

Multiple comparisons of the sites and soil layers considering the above-mentioned soil properties and the difference of the RSI-s were conducted using the function of Tukey HSD in a “multcomp” package within R. We used the “relaimp” package in R to analyze the contribution rate ([Kabacof, 2011](#)). One-way ANOVA ( $P < 0.05$ ) was used to test the difference between the three soil samples in the same index. Also, the Spearman correlation coefficient ( $r$ ) was used to quantify the linear relationship between RSI-s and soil properties within and between sites. All statistical tests were conducted using R v. 3.6.3 (R Project for Statistical Computing, Vienna, Austria), and all drawings were completed by Origin 2021 (OriginLab, OriginPro 2021, USA).



**Fig. 2.** Relative Stability Indices (RSI-s) of Soil Organic Carbon in Different Soil type-plantation forest combination and different soil depths. QP - Quartissamment soil poplar (*Populus × euramericana* cv. "74/76") plantation, ECP - Eutrochrepts soil Chinese pine (*Pinus tabulaeformis*) plantation, HEO - Haplustepts soil-East Liaoning oak (*Quercus liaotungensis* Koidz) plantation, RI - Respired carbon from incubation, ASI - Aggregate stability index, POC - Particulate organic carbon, WSC - Water-soluble carbon, MBC - Microbial biomass carbon, C: N - ratio of carbon to nitrogen. The same lower case letters no difference at  $P = 0.05$ .

### 3. Results

#### 3.1. Soil chemical and physical properties

There are great differences in soil properties between different plantations (Table 2) and only BD, porosity, and SOC indicators were significantly different between different soil layers ( $P < 0.05$ ). It can be seen that QP, ECP, and HEO are alkaline soil, acid soil, and neutral soil, respectively. At the same time, sand accounted for the largest proportion of QP, sand, and BD was significantly higher than the other two types ( $P < 0.05$ ), and soil physical (clay, porosity) and soil chemical indices (SOC, TN, and CEC) were lower than the other two types ( $P < 0.05$ ). For ECP, porosity and soil nutrient content (SOC and TN) were significantly higher than the other two types, and BD was significantly lower than the other two types ( $P < 0.05$ ). For HEO, clay is significantly higher than the other two types ( $P < 0.05$ ). It can be seen that there are significant differences in soil physicochemical properties due to differences in topography, stand, age and soil type ( $P < 0.05$ ).

#### 3.2. Relative stability indices (RSI-s) of soil organic carbon within and across sites

Fig. 2a shows the different organic carbon stability indicators for the three different plantations, which manifested distinctly trends. It can be seen that POC accounts for the highest proportion of TOC, reaching more than 72% (the content is greater than  $30 \text{ g kg}^{-1}$ ), and MBC accounts for the smallest proportion of TOC, which is only 3% to 4% (content  $< 2.5 \text{ g kg}^{-1}$ ). POC, ASI, and MBC were not significantly different among the three different plantations with significant differences ( $P < 0.05$ ) in soil physicochemical properties (Table 2). WSC showed that QP was significantly higher than ECP and HEO ( $P < 0.05$ ), C: N showed that QP and HEO were significantly higher than ECP ( $P < 0.05$ ), and RI showed a trend of ECP  $>$  HEO  $>$  QP ( $P < 0.05$ ). It can be seen that the trends of these six indicators between the three different plantation combinations show specific inconsistencies. Comparing the stability indexes of organic carbon among soil depths (Fig. 2b), it can be seen that POC, ASI, MBC, and C: N showed no significant difference between different soil layers. RI showed that the topsoil (0–10 cm) was significantly larger than the deep soil in 10–40 cm ( $P < 0.05$ ). WSC showed that the topsoil (0–10 cm) was significantly different from deep soil in 10–40 cm ( $P < 0.05$ ). For these six indicators, there is no unified trend, and when comparing the stability of organic carbon considering different plantations and soil depths, consistent results cannot be

obtained. This leads to different results using different indicators. Even among the plantations with obvious differences in topography and soil physicochemical properties with different stand ages, some indicators were not significantly different. It can be seen that it is inaccurate to use these indicators to measure the changing trend of the plantation over time or terrain.

#### 3.3. Relative contribution of RSI-s to SOC in three different SPC

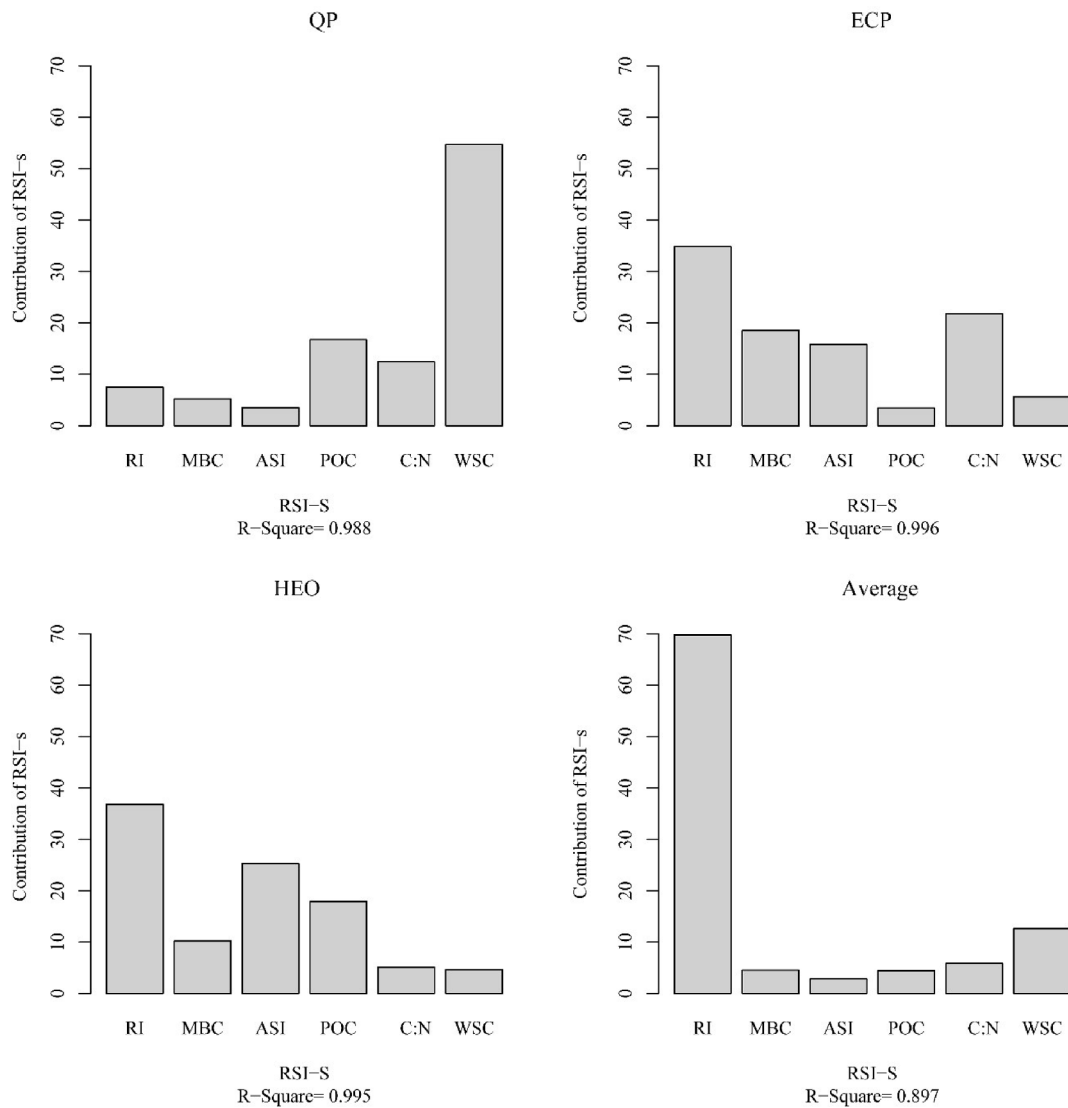
By comparing the contribution rate (Contribution rate refers to the degree to which RSI-s explain the variance ( $R^2$ ) in the RSI-s and TOC regression models) of RSI-s to SOC (Fig. 3), QP, ECP, and HEO showed differences, which further explains that the difference in organic carbon content in different soils and forests is caused by different organic carbon RSI-s.

In QP, WSC has the highest contribution rate to the SOC, reaching 54.73%. Other RSI-s (RI, MBC, ASI, POC, and C: N) have contributed  $< 20\%$  to the total content of SOC. The ASI has the smallest contribution rate, with only 3.53%. In the ECP, RI, MBC, ASI, and C: N have a higher contribution rate to the total SOC content. Among them, RI contributions was reaching 34.85%. The other three RSI-s contribute about 10% of the TOC in the soil. And the POC contribution rate is the lowest, at only 3.42%. The HEO, RI, ASI, and POC have a higher contribution rate to the SOC. Among them, RI has the highest contribution rate to the SOC, reaching 36.82%. The other three RSI-s contribute about 10% of the total SOC. However, the contribution of the RSI-s in three study areas to SOC content can be found. Among them, RI has the highest contribution rate to total SOC, reaching 69.79%, while the other RSI-s contribute about 10% to SOC. The SOC contribution is not significant. This shows that in terms of measuring SOC stability RSI-s, WSC indicators are more suitable for QP than other RSI-s. For the ECP, RI, MBC, ASI, and C: N, RSI-s are more suitable than other ones. For the HEO, it is more appropriate to use RI, ASI, and POC indicators as compared to other RSI-s. For RSI-s that measure the stability of total SOC, RI is more suitable than other indicators. It can be seen that different indicators should be used for different plantations.

## 4. Discussion

#### 4.1. Factors affecting the difference in RSI-s

We observed differences between different SOC indicators coinciding with other authors. For example, Zhu et al. (2014) also used SOC, DOC, MBC, SOC, soil C: N, Microbial C: N, and MWD (Soil aggregate

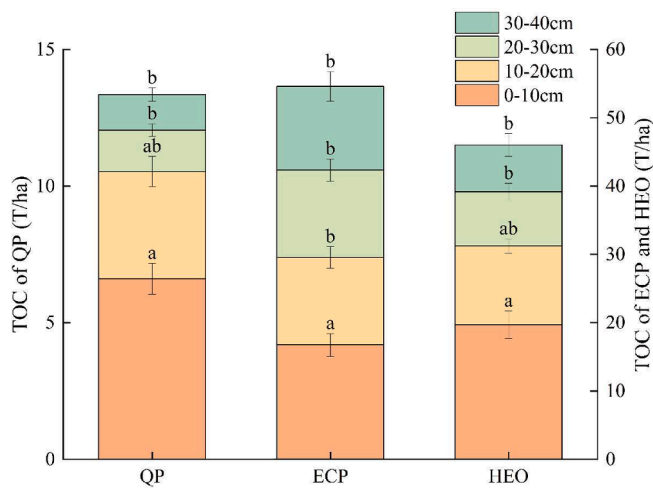


**Fig. 3.** Relative contribution of RSI-s to SOC in three different study areas. The contribution of RSI-s is determined by the relative weight of the contribution of each predictor variable (itself or in combination with other predictors) to the R square. QP- Quartisamment soil poplar (*Populus × euramericana* cv. “74/76”) plantation, ECP- Eutrochrepts soil Chinese pine (*Pinus tabulaeformis*) plantation, HEO - Haplustepts soil-East Liaoning oak (*Quercus liaotungensis* Koidz) plantation, RI- Respired carbon from incubation, ASI- Aggregate stability index, POC- Particulate organic carbon, WSC- Water-soluble carbon, MBC = -Microbial biomass carbon, C: N- ratio of carbon to nitrogen.

index). However, our results also highlighted that there was no complete consistency between these indicators, for example, the aggregate stability (Nie et al., 2018). Also, Cheng et al. (2015) selected TOC, aggregate C, and particulate organic matter-carbon (iPOM-C) concentration as indicators when studying the impact of different vegetation restoration years (3, 7, 15, 25, 36, and 56 years) on the Loess Plateau in China. They also confirmed that there was no complete consistency among these indicators. It is demonstrated that specific forests can show different impacts on soil microorganisms, which would have caused changes in SOC storage. Due to the different survival strategies of microbes at the surface and deep soil layers, it appears that more easily decomposed organic carbon has been accumulated in the deeper soil parts. This is the reason why both RI and MBC show higher values at the surface layer than the deep ones.

The difference in soil's physical and chemical properties was caused by the difference in tree species in stand age. The differences in soil physical and chemical properties and SOC stability index influence each other and form different trends. pH affects the stability of soil aggregates by influencing the forces between soil aggregates (Yu et al., 2020d). This

could explain why the ASI of the ECP is higher than the HEO and QP in the three plantation combinations. The ECP soils are acidic with the lowest pH, while the QP soils are alkaline and register the highest pH. As the clay content increases, the nitrate concentration in the soil also increases (Soenne et al., 2021; Li et al. 2022) which then affects the C: N ratio. This is consistent with our research. The clay content in the ECP and HEO is more than that in QP, so the TN content is quite different. In our study, it can be found that the nitrogen content of the topsoils is higher than that of the bottom ones (Cremer et al., 2016). Soil organic carbon concentration is significantly correlated with all unstable carbon components and enzyme activity (Tajik et al., 2012; Chen et al., 2016). Due to the differences in forest age between QP, ECP, and HEO, QP is relatively young in planting age and has relatively little impact on soil. The effects of litter in QP and enzymes in the soil produce a large amount of activated organic carbon (Tajik et al., 2020a) so that the content of WSC is relatively higher than that in the other two places. The levels of SOC and TN increase with the age of vegetation (Jia et al., 2012, Yu and Jia, 2014), which is consistent with our research. The content of SOC and TN in QP is significantly lower than that of ECP and HEO.



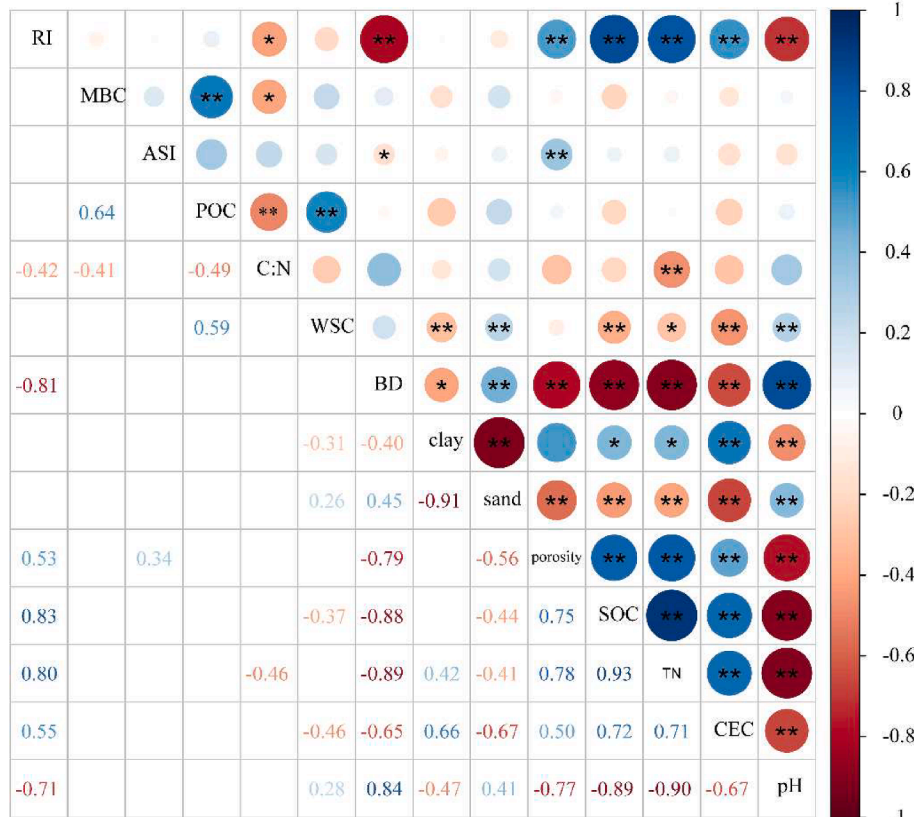
**Fig. 4.** Total soil organic carbon content in QP- Quartissamment soil poplar (*Populus × euramericana* cv. “74/76”) plantation, ECP- Eutrochrepts soil Chinese pine (*Pinus tabulaeformis*) plantation, HEO - Haplustepts soil East Liaoning oak (*Quercus liaotungensis* Koidz) plantation.

Our results show that as the depth of the soil layer deepens, the SOC content decreases (Fig. 4), which is also consistent with previous research results (e.g. Liu et al, 2021, Wang et al., 2017, Wordell-Dietrich et al., 2017, Wang et al., 2015, Jia et al., 2012). With the increase of vegetation restoration years, deep soil carbon storage showed an increasing trend (Yu et al., 2020b). As in our study, ECP and HEO have longer recovery years than QP, and the TOC content in deeper soils is relatively small (Ajami et al., 2016). At the same time, Yu et al., 2020b pointed out that the higher surface SOC content may be affected by the input of litter, and the vegetation type gradually weakens the contribution of SOC as the soil layer deepens.

#### 4.2. Correlation between various RSI-s

There is a significant correlation between the SOC index and the soil’s physical and chemical properties (Fig. 5). Mandal et al. (2020) also analyzed the correlation between SOC and soil physical and chemical properties showing that SOC has a significant negative correlation with pH, BD, and sand content, which is consistent with our research results. On the other hand, Baena et al., (2013) in South-eastern Spain studied thinning effects on soil microbial activity and biomass and also highlighted that soil microbial biomass had a significant correlation with soil physical and chemical properties, which differs from our investigation, which may be caused by the large difference in climate (Mediterranean climate) and soil type (sandy loam texture). By comparing the correlation between each index and the physical and chemical properties of the soil, we noted that RI, as the respired carbon from incubation to TOC at 60 d obtained the best linear correlation with each index. Among them, considering the Spearman correlation coefficient, RI showed a significant correlation with BD, sand, porosity, SOC, TN, CEC, and pH. Among the six indicators, RI was the most suitable one related to the soil stability of the three plantation combinations.

The correlations between different SOC stability indicators and soil physical and chemical properties were different (Fig. 5). Combining the contribution of different indicators to SOC (Fig. 3), it was observed that the organic carbon stability of different plantation combinations was affected by soil physical and chemical properties. For QP, WSC had the highest contribution to SOC change, and WSC was significantly correlated with clay, sand, SOC, TN, CEC, and pH ( $P < 0.05$ ), with the highest correlation with CEC (0.46). For ECP, RI was the highest contributor, where RI was significantly correlated with BD, sand, porosity, SOC, TN, CEC, and pH ( $P < 0.05$ ), with SOC being the most correlated (0.83). For HEO, RI, AIS, and POC were high contributors to SOC stability, where ASI was significantly correlated with BD and porosity ( $P < 0.05$ ), and MBC was weakly correlated with soil physical and chemical properties. At the same time, it can be seen from the comparison of the differences



**Fig. 5.** Spearman correlation among the soil organic carbon stability indicators and soil physical and chemical properties. Colors represent the direction of the correlation (blue = positive; red = negative). Different sizes of the circles are proportional to the R<sup>2</sup> value. P-values represented in black color are significant (\*P ≤ 0.05, \*\*P ≤ 0.01). The correlation coefficients are shown in the lower-left panel. The correlation coefficient of p-value ≤ 0.05 is displayed in the lower-left panel. RI- Respired carbon from incubation, ASI- Aggregate stability index, POC- Particulate organic carbon, WSC = Water-soluble carbon, MBC = Microbial biomass carbon, C: N- ratio of carbon to nitrogen.

of different indexes among different plantations and soil layers (Fig. 2) that RI, WSC, C: N had significant differences among different plantations ( $P < 0.05$ ), and RI and WSC had significant differences among different soil layers ( $P < 0.05$ ). As we described, different indexes showed distinct trends among the three plantations and four soil layers, and the dominant factors among the three plantations were also different. Therefore, a new index should be proposed as a constant and unified standard for evaluating the effects of plantations on SOC stability.

## 5. Conclusion

In this study, we analyzed six SOC stability indexes and influencing factors in the three different plantation combinations with different soil types. Among the six indicators, we found that different indicators should be selected for different forest stands, and combining three combinations of artificial forests, we found that different indicators presented inconsistent evaluation effects. In this study, we focused on six widely used SOC indicators. Meanwhile, respired carbon from incubation showed an inevitable loss of carbon content during the process of soil migration. In this way, *in situ* soil respiration carbon can be used to improve its accuracy. Our results have crucial implications for SOC stability assessment in theory and practice. It is important to select different indicators for different types of forest. Although RI performed well in measuring the stability of SOC in plantation forests, it required more time for incubation experiments during the measurement processes, while other indicators such as ASI were relatively simple and fast to obtain. Greater attention should also be directed toward the accuracy of the organic carbon assessment, *in-situ* observation should be implemented, aimed at better solutions for organic carbon stability evaluation in different plantations.

## CRedit authorship contribution statement

**Zeyu Zhang:** Conceptualization, Formal analysis, Writing – original draft, Visualization. **Tonggang Zha:** Conceptualization, Investigation, Resources, Writing – review & editing, Funding acquisition. **Yang Yu:** Conceptualization, Writing – review & editing. **Xiaoxia Zhang:** Writing – review & editing. **Pete Smith:** Writing – review & editing. **Jesús Rodrigo-Comino:** Writing – review & editing.

## Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

## Data availability

Data will be made available on request.

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