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Protection mechanisms and influencing factors of soil organic carbon pools in the *Larix gmelinii* forests

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A R T I C L E I N F O

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A B S T R A C T

The stability and dynamics of soil organic carbon (SOC) pools in forest soils are crucial for assessing and predicting the effectiveness of forest carbon sequestration. Various factors such as soil depth, nutrients availability, crustal elements, pH, and stand characteristics (tree type, tree age) influence the stability of SOC pools. To better understand SOC stability of cold temperate forest soil, soil samples were collected from 0–10 cm, 10–20 cm, 20–40 cm, and 40–60 cm depths in a *Larix gmelinii* forest. Then, the distribution characteristics of SOC pools under different protection mechanisms and their influencing factors were examined. Our results showed that the unprotected SOC pool was the highest, while the physical–chemical protected SOC pool was the lowest. Total organic carbon (TOC) had a linear positive relationship with unprotected and physical-protected SOC pools, and a logarithmic positive relationship with physical–chemical protected and chemical-protected SOC pools. Soil depth was the predominant factor affecting the SOC pools of each protection mechanism. The physical–chemical protected SOC pool significantly differed from forest types and forest ages, and the chemical-protected SOC pool was associated with the synergistic effects of forest age and type. Furthermore, soil moisture and acidity, and the crustal elements may play prominent role in the variation of SOC pools in the *Larix gmelinii* forests. Our findings suggested that the silt- and clay-associated SOC pools in the *Larix gmelinii* forest soils had approached or achieved saturation, while more organic C was fixed in the relatively active unprotected SOC pool. This unprotected pool was more sensitive to ecosystem change, indicating its importance in assessing forest carbon sequestration. Our study provided insights into the active and dynamic SOC pools in forest soils and their responses to different environmental factors. These findings can be utilized to accurately assess the carbon sink potential of cold temperate forest soils and inform the management of forest ecosystems.

1. Introduction

The carbon (C) pool of forest soils is the largest in terrestrial ecosystems, with significant impact on global C balance owing to their storage capacity and role as a source or sink of atmospheric CO_2 (Schlesinger, 1990). Consequently, the study of forest soil C pools has garnered increasingly attention in recent years across temperate, subtropical, tropical, and other regions (Wang et al., 2016; Xing et al., 2017; Qin et al., 2012). SOC is a highly complex collection of chemicals that are derived from plant, animal, and microbial residues at different stages of degradation (Christensen, 1992; Post and Kwon, 2000). It is composed of C banks that vary greatly in terms of stability and turnover cycle (Liao et al., 2011; Liu et al., 2007). SOC exhibits differential response to environmental changes depending on its sources, degradation stages, and fractions, suggesting diverse environmental behavior and stability. Therefore, SOC storage capacity and its ecological service function differ (Doetterl et al., 2015; von Lützow et al., 2007). Quantifying the storage capacity of SOC pools is of fundamental importance in the field of SOC dynamics (Yuan et al., 2018). Previous studies have revealed that the total organic C (TOC) always has the characteristic of lag in response to environmental changes, despite its high background value in the soil (Song et al., 2015). However, the labile SOC pools in TOC demonstrate sensitivity to such changes, highlighting the need to better understand the ecological and environmental implications of various SOC pools, fractionations or morphologies in forest soils (Poeplau et al., 2017; Zimmermann et al., 2007; Zhang, 2007; Zhang et al., 2014a), including their contents, spatial patterns, and annual dynamics of SOC pools (Gu et al., 2013; Wang et al., 2016;

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Mu et al., 2020; Tian and Man, 2016).

Soil C stabilization has been found to be associated with various physicochemical properties of soil, including the presence of mineral surfaces capable of adsorbing organic materials (thus providing a protective capacity), the chemical nature of the soil mineral fraction, the presence of cations and the nutrients, and the soil matrix architecture (Stewart et al., 2008; Baldock and Skjemstad, 2000). The general extraction methods for SOC pools include chemical, physical, and stability fractionations. The chemical approach is typically based on the solubility and oxidizability of soil organic matter in various extractants. However, this method is not always reliable since the original structure of soil organic matter may be damaged during the extraction process using chemical extractants, which can lead to misunderstanding results when attempting to explain dynamic changes in the SOC pool. According to stability, SOC is often categorized into three distinct pools; namely, active organic C, slow-acting organic C, and inert organic C. However, a long incubation period and stringent limiting factors in this method poses a challenge in accurately reflecting biochemical mechanism of SOC. Alternatively, the physical fractionation method operates on the basis of SOC density or soil particulate size, which allows the maintenance of soil undisturbed status with minimal damage. This method is widely used to capture SOC dynamics, soil fertility status, and their response to human management measures. Recently, the physical–chemical combined fractionation method proposed by Six et al. (2002), Six et al. (1998) and Stewart et al. (2008) has become the predominant in the study of SOC pool fractionation. This technique has the capability of effectively isolating SOC pools based on corresponding physical, chemical, and biochemical protection mechanisms, which has been widely applied in farmland ecosystem research (Li et al., 2014; Yang, 2018).

Generally, SOC contents follow a pattern of temperate forest > subtropical forest > tropical forest, with an exponentially increase observed in correspondence with latitude (Wang et al., 2016). It was reported that SOC density and storage were highest in the northern hemisphere, particularly at higher latitudes, making it a potential hotspot for biosphere CO_2 flux (Scharlemann et al., 2014). The Greater Khingan Mountains represent a unique habitat as it hosts the sole cold temperate coniferous forest in China, rendering it susceptible to global climate modifications. Thus, it is essential to conduct further research into the organic C stability of the cold temperate forest soils. As the prevalent tree species in the Greater Khingan Mountains, the environmental and ecological significance of SOC have garnered significant scholarly interests in the ecosystem of *Larix gmelinii* forests (Zu et al., 2009; Wang et al., 2011; Wei et al., 2021). These findings illustrated that the variations in SOC levels closely associated with the growth of *Larix gmelinii*, especially the vertical distribution pattern (Wang et al., 2013a; Wang et al., 2013b); the thinning management can enhance the sequestration and stability of SOC in the *Larix gmelinii* plantations (Zhou et al., 2019; Wang et al., 2013a; Wang et al., 2013b). Comparatively, the C sequestration abilities by *Larix gmelinii* were higher than other tree species in Northeastern China (Wang et al., 2017a; Wang et al., 2017b). These findings hold significant implications for managing forest ecosystems to promote C storage and mitigate climate change. In summary, there are various factors influencing the SOC pools in forests soils such as tree type, tree age, soil depth, and their interactions, apart from soil physicochemical properties. As the zonal vegetation in the Greater Khingan Mountains, *Larix gmelinii* plays an irreplaceable role in C sinks in cold temperate forests. In this paper, we assume soil C sequestration of *Larix gmelinii* forest in different protecting mechanism varied with soil depth, forest types, and tree growth, while such changes are strongly associated with soil N, P, and crustal elements. The detail objectives of this paper are the following: (1) analyze the characteristics of SOC pools under different protection mechanisms; (2) examine the synergistic effects of forest type, forest age, and soil depth on SOC pools; and (3) identify the key soil physicochemical properties influencing SOC pools in the *Larix gmelinii* forests. This work is beneficial to improve the understanding of the active and dynamic SOC pools in forest soils, which can provide scientific basis for accurate assessment of C sinks in cold temperate forest soils, and the management of forest ecosystems in Greater Khingan Mountains.

2. Materials and methods

2.1. Study area

The research site, located in the *Larix gmelinii* virgin forest of China's northern Greater Khingan Mountains (50°54′–50°57′N, 121°28′–121°32′E, Fig. 1), is characterized by a cold temperate humid climate with an annual average temperature of −5.4 ℃ and precipitation of 450–550 mm. This region, situated an altitude of 800–1000 m, is a low mountainous area with Alfisols (brown coniferous forest soils) and an abundance of swampy wetlands, as well as continuous permafrost. *Larix gmelinii* is the dominant species accompanied by *Betula platyphylla*, *Populus davidiana,* and other trees. *Rhododendron simsii, Ledum palustre*, *Betula fruticosa*, *Vaccinium vitis-idaea*, *Convallaria majalis*, *Maianthemum bifolium*, *Pyrola incarnata*, and *Deyeuxia langsdorffii* are the main species of understory plants. In accordance with the wide ecological niche of *Larix gmelinii*, and the dominant species of shrubs or grasses beneath the forest are contingent upon diverse site characteristics. Therefore, in the relevant researches of *Larix gmelinii* forest, the forest types are usually classified according to the dominant species of understory plants. Among them, the grass-*Larix gmelinii* forest, *Ledum-Larix gmelinii* forest and *Rhododendron-Larix gmelinii* forest are widely distributed in the study area.

2.2. Plot setting and soil sampling

In July 2017, 28 sites of 30 m × 30 m were established based on forest types and forest ages (young, middle-aged, near-mature, mature and over-mature). In terms of the forest types, there are 12 sites of grass-*Larix gmelinii* forests (3 for each age group), 8 sites of *Ledum-Larix gmelinii* forests (2 for each age group), and 8 sites of *Rhododendron-Larix gmelinii* forests (2 for each age group). Before sampling, the topographic information and vegetation status were recorded at each site. Three distinct dig sites were selected per location, and soil samples were carefully collected from layers positioned at 0–10 cm, 10–20 cm, 20–40 cm, and 40–60 cm. Three samples from identical depths were combined to rminimize soil heterogeneity. After being transported to the laboratory, the soil samples were air-dried and sieved for further analysis.

2.3. Laboratory analysis

2.3.1. Measurement of soil physiochemical properties

The oven drying method was used to determine the soil water content (SWC) and bulk density (BD). Soil pH was determined using a pH meter and a soil/water mass ratio of 1:5 (w/v). TOC content was measured using the potassium dichromate oxidation spectrophotometric method (HJ 615-2011). The total phosphorus (TP) was determined using the spectrophotometry method (Pierzynski, 2009). The inorganic phosphorus (IP) was determined by the SMT method (Ruban et al., 2001), and the organic phosphorus (OP) was calculated by subtracting IP from TP. The universal extract-colorimetric method (NY/T 1849-2010) was applied for the determination of ammonium nitrogen (NH_4^+-N), rapidly-available potassium (AK), and available phosphorus (AP). The crustal elements in the soils were determined using an XRF analyzer (BRUKER S8 TIGER SERIES 2, Germany) and expressed as their corresponding oxides including Na_2O, MgO, Al_2O_3, K_2O, CaO, and Fe_2O_3.

2.3.2. Soil fractionation and the associated C

Separation of the various SOC pools was accomplished by a combined fractionation technique (Stewart et al., 2008; Stewart et al., 2009;

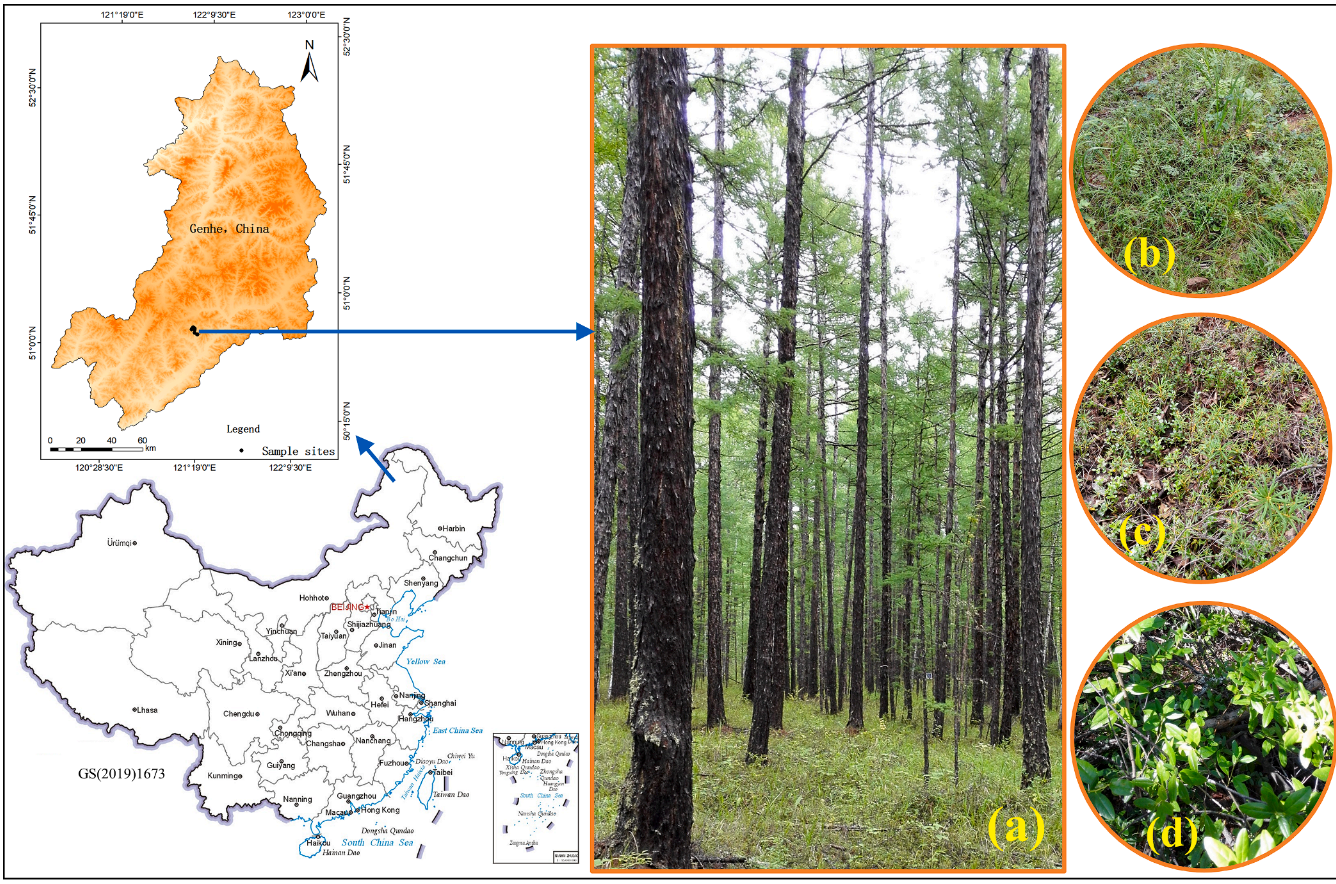

Fig. 1. The distribution map of sampling sites and pictures of field sampling sites. (a) *Larix gmelinii* forest; Understory plants: (b) Grass; (c) *Ledum palustre*; (d) *Rhododendron simsii*.

Six et al., 2002) (Fig. 2). Firstly, using an aggregate separator, the soil samples were dispersed and physically separated into three size fractions: >250 μm unprotected coarse particulate organic matter (cPOM), 53–250 μm microaggregate (μagg), and <53 μm easily dispersed silt and clay fraction (dSilt and dClay). Secondly, heavy liquid flotation and dispersion were applied for further fractionation of the microaggregate fractions isolated from the first step, by which four fractions were obtained, i.e., the fine unprotected POM (LF), the microaggregate-protected POM (>53 μm in size, iPOM), the microaggregate-derived silt- and clay-sized fractions (μSilt and μClay). Finally, seven soil fractions were successfully isolated: cPOM, LF, iPOM, dSilt, dClay, μSilt, and μClay. All soil fractions were dried at 60 °C and weighed for further determination of C contents.

The C contents of the seven soil fractions was determined by the TOC Analyzer (SHIMADZU TOC-V CPH, Japan). The attributes of soil

Table 1
Soil fractionation and the associated C pools.

SOC pool	Isolated soil fraction	Particulate size (μm)
Unprotected POM (uPOM)	cPOM	>250
	LF	0.45
Physical-protected POM (pPOM)	iPOM	53–250
Physical-chemical protected POM (pcPOM)	μSilt	2–53
	μClay	<2
Chemical-protected POM (CPOM)	dSilt	2–53
	dClay	<2

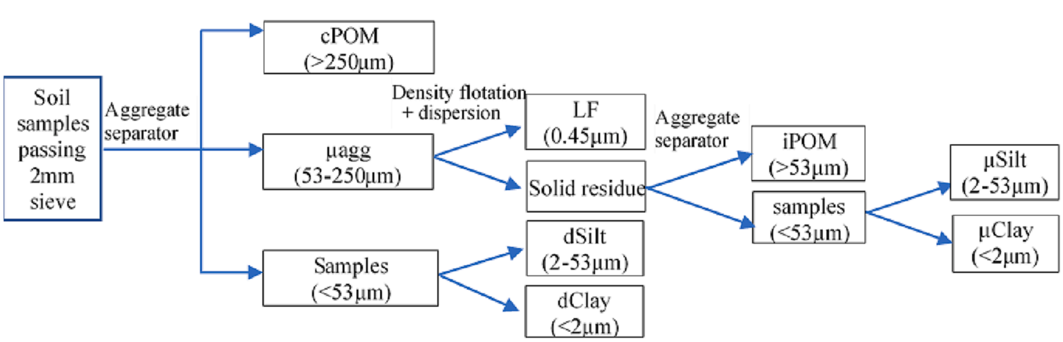

Fig. 2. Flow chart of soil fractionation. Soil fractions abbreviations: cPOM – Unprotected Coarse Particulate Organic Matter; μagg – 53–250 μm microaggregate; dSilt – <53 μm Easily Dispersed Silt; dClay – <53 μm Easily Dispersed Clay; LF – Fine Unprotected Particulate Organic Matter; iPOM – Microaggregate-protected Particulate Organic Matter; μSilt – Microaggregate-derived Silt; μClay – Microaggregate-derived Clay.

fractions were listed in Table 1. In detail, the SOC pool of unprotected POM (uPOM) consists of two fractions, cPOM and LF, which exhibit high biological activity influenced largely by soil organic matter inputs (Conant et al., 2004). The fraction of iPOM is a type of physical-protected organic C (pPOM) that accumulates in microaggregates and is active in the soil (Lal, 2004). The chemical-protected organic C (CPOM) pool includes dSilt and dClay fractions, whereas the physical–chemical protected organic C (pcPOM) pool consists of μSilt and μClay fractions. The SOC pools of CPOM and pcPOM are inert fractions, which is conducive to long-term preservation (Dou et al., 2011), and can also be used to predict soil C saturation (Zhang et al., 2014a).

See Fig. 2 for soil fractions abbreviations.

2.4. Data calculation and statistical analysis

The storage capacity of each SOC pool was calculated using the following formulas:

$$S_f = \sum_{i=1}^{n} (D_i \times B_i \times W_i/10) \tag{1}$$

where S_f is the storage capacity of each SOC pool (t hm^{-2}), D_i is the soil depth of the i^{th} layer (cm), B_i is the soil bulk density of the i^{th} layer (g cm^{-3}), W_i is the C content for each SOC pool of the i^{th} layer (g kg^{-1}).

An ANOVA was employed to test the differences between soil fractions in different soil layers. The general linear model (GLM) and Duncan multiple comparison were used to analyze the effects of forest type, forest age, soil depth, and their interactions on SOC pools protected by different mechanisms. The relationships between SOC pools and soil physicochemical properties were investigated using Pearson Correlation Analysis and stepwise regression. The dependent variables were transformed by natural logarithm to meet the statistical analysis requirements. All statistical analysis was completed using IBM SPSS 22.0 and Excel 2016.

3. Results

3.1. Multiple analysis of variance (MANOVA)

The MANOVA results revealed significant effects of forest age, forest type, and soil depth on the SOC pools of uPOM, pPOM, pcPOM, and CPOM in the *Larix gmelinii* forests (Table 2). In particular, soil depth played a crucial role in regulation SOC pools, showcasing significant effects on the pools of uPOM, pPOM, and CPOM ($P < 0.001$), thereby rendering it the most significant factor. Relatively, weak effects of forest type and age were observed on the SOC pools of uPOM, pPOM, pcPOM, and CPOM based on the sample size in this work. A significant co-effect of forest type and age were found on the pool of CPOM ($P < 0.05$). The order of the factors influencing the SOC pools was: soil depth > forest age × forest type > soil depth × forest age > soil depth × forest type × forest age > forest type > forest age > soil depth × forest type.

The Duncan multiple comparison results were showed in Table 3. Regarding soil profile, the SOC pools of uPOM, pPOM, and CPOM shared the same trends, with higher contents in the upper soils (0–10 and 10–20 cm) than in the subsoils (20–40 and 40–60 cm); while no significant discrepancies were observed between the layer of 20–40 cm and 40–60 cm. In terms of forest types, the SOC pool of pcPOM in the grass-*Larix gmelinii* forest was notably greater than that in the *Rhododendron-Larix gmelinii* forest. Additionally, the pcPOM pool in young forests was significantly lower compared to other age groups.

3.2. Physicochemical properties and SOC fractions

Table 4 shows the statistics of the soil physicochemical properties from the *Larix gmelinii* forest. There were some variances in different indicators among samples in general. SWC varied from 5.0 % to 50.61 % with an average of 17.93 %, presenting a decreasing trend with the soil depth. In contrast, BD ranged from 0.66 g·cm^{-3} to 1.39 g·cm^{-3} with an average of 1.07 g·cm^{-3}, presenting an increasing trend with the soil depth. The soils of *Larix gmelinii* forest were acidic and the mean pH was 5.59 with a range of 4.01–6.34. For nutrients, NH_4^+-N ranged from 3.15 mg kg^{-1} to 59.18 mg kg^{-1} with the mean of 17.94 mg kg^{-1}, AK ranged from 6.88 mg kg^{-1}to 347.88 mg kg^{-1} with the mean of 89.05 mg kg^{-1},

Table 2
Effects of forest type, forest age, and soil depth on SOC pools using a GLM.

Variance source		*SS*	*f*	*MS*	*F*	*P*
Age	uPOM	4.527	3	1.509	2.358	0.085
	pPOM	0.234	3	0.078	0.093	0.963
	pcPOM	4.224	3	1.408	2.717	0.056
	CPOM	0.341	3	0.114	0.439	0.726
Type	uPOM	3.998	2	1.999	3.123	0.054
	pPOM	4.818	2	2.409	2.878	0.067
	pcPOM	1.559	2	0.780	1.504	0.234
	CPOM	0.282	2	0.141	0.545	0.584
Depth	uPOM	109.388	3	36.463	56.973	0.000
	pPOM	41.884	3	13.961	16.682	0.000
	pcPOM	3.702	3	1.234	2.381	0.083
	CPOM	7.638	3	2.546	9.824	0.000
Age × type	uPOM	3.971	6	0.662	1.034	0.417
	pPOM	5.054	6	0.842	1.007	0.434
	pcPOM	5.771	6	0.962	1.856	0.111
	CPOM	4.708	6	0.785	3.028	0.015
Age × depth	uPOM	4.948	9	0.550	0.859	0.568
	pPOM	7.078	9	0.786	0.940	0.502
	pcPOM	4.398	9	0.489	0.943	0.499
	CPOM	1.393	9	0.155	0.597	0.792
Type × depth	uPOM	2.363	6	0.394	0.615	0.717
	pPOM	3.885	6	0.648	0.774	0.595
	pcPOM	1.856	6	0.309	0.597	0.731
	CPOM	0.321	6	0.054	0.207	0.973
Age × type × depth	uPOM	5.455	15	0.364	0.568	0.883
	pPOM	3.787	15	0.252	0.302	0.993
	pcPOM	4.038	15	0.269	0.520	0.916
	CPOM	1.425	15	0.095	0.367	0.981

Explanation: *SS – Square Sum; f* – Freedom; *MS* – Mean Square. See Table 1 for SOC pools abbreviations.

Table 3
The differences in SOC pools between different soil depths, forest types, and forest ages using the Duncan multiple comparison (g kg^{-1}).

Factor	Group	uPOM	pPOM	pcPOM	CPOM
Depth	0–10 cm	116.53 ± 56.41^{a}	7.00 ± 7.93^{a}	1.05 ± 0.41^{ab}	10.38 ± 2.99^{a}
	10–20 cm	17.56 ± 15.28^{b}	1.96 ± 1.50^{b}	1.28 ± 0.72^{b}	7.38 ± 3.80^{b}
	20–40 cm	5.57 ± 5.57^{c}	0.96 ± 1.14^{c}	1.01 ± 1.27^{a}	4.83 ± 3.05^{c}
	40–60 cm	4.35 ± 3.01^{c}	0.97 ± 0.95^{c}	0.98 ± 1.14^{a}	4.53 ± 3.22^{c}
Type	Grass-*Larix gmelinii* forest	28.61 ± 45.26^{a}	2.54 ± 5.55^{a}	1.32 ± 1.18^{a}	6.90 ± 4.16^{a}
	Ledum-Larix gmelinii forest	52.72 ± 72.51^{a}	3.67 ± 5.54^{a}	1.02 ± 0.69^{ab}	7.56 ± 3.73^{a}
	Rhododendron-Larix gmelinii forest	35.33 ± 51.18^{a}	2.45 ± 2.18^{a}	0.75 ± 0.44^{b}	6.22 ± 4.01^{a}
Age	Young forest	48.56 ± 74.53^{a}	2.60 ± 3.42^{a}	0.59 ± 0.36^{a}	6.45 ± 3.70^{a}
	Middle-aged forest	38.45 ± 45.73^{a}	2.71 ± 2.81^{a}	1.15 ± 0.69^{b}	7.39 ± 3.75^{a}
	Near-mature forest	20.76 ± 34.01^{a}	1.78 ± 1.92^{a}	1.31 ± 1.33^{b}	6.46 ± 3.96^{a}
	Mature and over-mature forest	48.68 ± 72.32^{a}	4.53 ± 8.89^{a}	1.03 ± 0.73^{b}	7.15 ± 4.70^{a}

Explanation: The data in the table are the Mean ± SD of SOC pools. The lowercase letters of a, b, c represents the significance of differences in SOC pools between different soil depths, forest types or forest ages. See Table 1 for SOC pools abbreviations.

Table 4
Summary of soil physicochemical properties.

Indexes	Min-max	Mean ± SD.
SWC (%, n = 83)	5.0–50.61	17.93 ± 11.40
BD ($g \cdot cm^{-3}$, n = 56)	0.66–1.39	1.07 ± 0.14
pH (n = 89)	4.01–6.34	5.59 ± 0.42
NH_4^+-N (mg kg^{-1}, n = 92)	3.15–59.18	17.94 ± 9.82
AK (mg kg^{-1}, n = 93)	6.88–347.88	89.05 ± 63.01
AP (mg kg^{-1}, n = 79)	3.89–71.04	17.85 ± 12.68
OP (mg kg^{-1}, n = 96)	95.88–1338.75	432.31 ± 224.68
Na_2O (%, n = 85)	0.21–3.17	1.80 ± 0.61
MgO (%, n = 85)	0.37–1.95	1.30 ± 0.37
Al_2O_3 (%, n = 85)	6.27–18.98	13.77 ± 1.96
K_2O (%, n = 85)	0.80–3.03	2.33 ± 0.34
CaO (%, n = 85)	0.71–3.33	1.25 ± 0.52
Fe_2O_3 (%, n = 85)	2.70–8.50	5.44 ± 1.09

Explanation: SWC – Soil Water Content; BD – Bulk Density; AK – Rapidly-available Potassium; AP – Available Phosphorus; OP – Organic Phosphorus.

AP ranged from 3.89 mg kg^{-1} to 71.04 mg kg^{-1} with the mean of 17.85 mg kg^{-1}, OP ranged from 95.88 mg kg^{-1} to 1338.75 mg kg^{-1} with the mean of 432.314 mg kg^{-1}. The crustal elements in the soils were measured using XRF and showed as their corresponding oxides (Table 4), in which Na_2O varied from 0.21 % to 3.17 %, MgO varied from 0.37 % to 1.95 %, Al_2O_3 varied from 6.27 % to 18.98 %, K_2O varied from 0.80 % to 3.03 %, CaO varied from 0.71 % to 3.33 %, and Fe_2O_3 varied from 0.70 % to 8.50 %. On the whole, the pH had the smallest variation, followed by BD and crustal elements, while the changes in nutrients were noticeable. These results highlighted the significance of considering soil heterogeneity.

There seven soil fractions were isolated as cPOM, iPOM, LF, dSilt, dClay, μSilt, and μClay according to the combined fractionation technique, and a decreasing trend of the corresponding SOC contents were observed in these soil fractions as the particulate size got smaller i.e., cPOM (34.51 g kg^{-1}) > dSilt (6.47 g kg^{-1}) > iPOM (2.81 g kg^{-1}) > LF (2.43 g kg^{-1}) > μSilt (0.89 g kg^{-1}) > dClay (0.43 g kg^{-1}) > μClay (>0.20 g kg^{-1}). Based on the protection mechanisms, the seven isolated soil fractions were grouped into four SOC pools (Table 1), following uPOM (36.94 g kg^{-1}) > CPOM (6.90 g kg^{-1}) > pPOM (2.81 g kg^{-1}) > pcPOM (1.09 g kg^{-1}), in which the uPOM pool was the highest (56.39 %) and the pcPOM pool was the lowest (5.57 %) in the *Larix gmelinii* forests (Fig. 3).

Further, the C storage of uPOM, pPOM, pcPOM, and CPOM pools were 155.95, 11.35, 3.90, and 28.12 t hm^{-2} in the 0–40 cm soil layers, respectively, from the *Larix gmelinii* forest (Fig. 4), accounting for 71.38 %, 5.19 %, 1.79 %, and 12.87 % of the soil TOC storage. The uPOM pool was the largest in SOC.

3.3. SOC pools in soil profiles

The SOC pools of uPOM, pPOM, and CPOM decreased gradually with soil depth and their contents in the top layers (0–10 cm) were significantly larger than those of bottom layers (Fig. 5). The SOC pools under the four protection mechanisms were in the following order: uPOM > CPOM > pPOM > pcPOM in the 0–10 cm and 10–20 cm layers. Generally, the SOC pools of uPOM and CPOM dominated in the whole soil profiles. It was notable that these two SOC pools showed clearly opposite trends with the soil depth and reached equal from 20–40 cm to bottom.

3.4. Correlations and stepwise regression analysis

We observed a noteworthy positive correlation between TOC and various SOC pools. Specifically, the TOC linearly related to the uPOM and pPOM pools ($P < 0.01$), while logarithmically related to the pcPOM

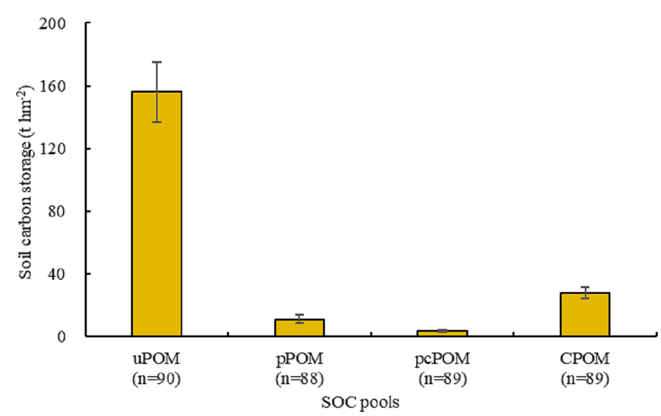

Fig. 4. SOC storages of different protection mechanisms in the *Larix gmelinii* forest (Mean ± SD). See Table 1 for SOC pools abbreviations.

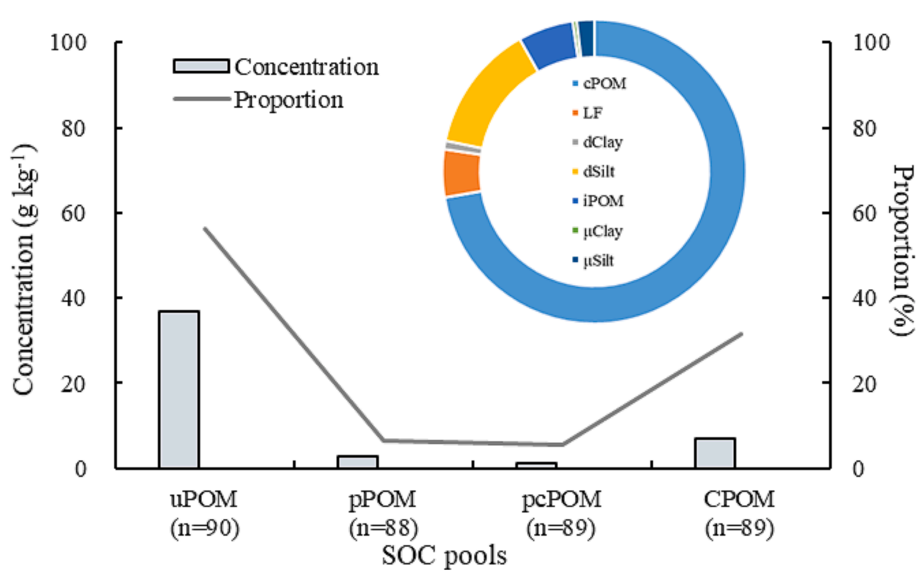

Fig. 3. Contents and proportions of SOC pools, and proportions of soi fractions. The contents of SOC pools are displayed as rectangles. The proportions of SOC pools are displayed as a solid line. The proportions of soil fractions are displayed as a ring. See Fig. 2 for soil fractions abbreviations. See Table 1 for SOC pools abbreviations.

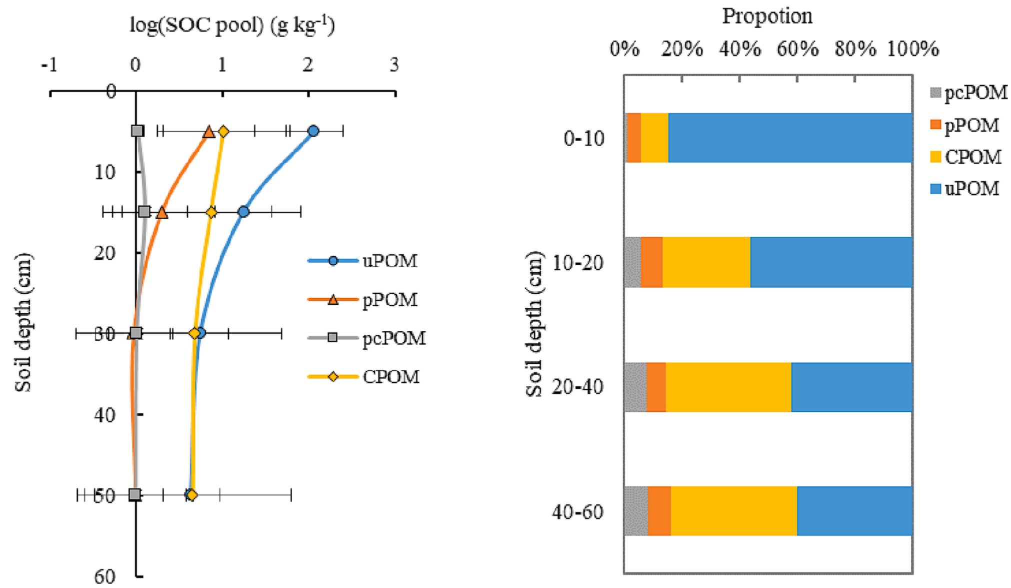

Fig. 5. Profile characteristics of SOC pools. The profile contents of SOC pools are displayed as solid lines. The profile proportions of SOC pools are displayed as rectangles. See Table 1 for SOC pools abbreviations.

($P < 0.05$) and CPOM pools ($P < 0.01$, Fig. 6). Significantly positive and negative correlations of the SOC pools were also found with SWC ($P < 0.05$) and BD ($P < 0.01$), respectively (Table 5). Soil pH significantly negatively correlated with the SOC pools of uPOM, pPOM, and CPOM ($P < 0.01$). The SOC pools of uPOM, pPOM, and CPOM significantly negatively correlated with the crustal elements (Na_2O, MgO, Al_2O_3, and K_2O) ($P < 0.01$ or $P < 0.05$) and also significantly positively correlated with NH_4^+-N, AK, OP, and CaO ($P < 0.01$ or $P < 0.05$); While the pcPOM pool significantly positive correlated with NH_4^+-N, MgO, Al_2O_3, and CaO in soils from the *Larix gmelinii* forests ($P < 0.01$ or $P < 0.05$). These results indicated that the changes in the SOC pools under four protection mechanisms inherently associated with the soil physicochemical properties in *Larix gmelinii* forests.

Further, to quantitatively assess the influence of single and multiple factors on each SOC pool, the stepwise regression analysis was performed with forest type, forest age, soil depth, and soil physicochemical properties as the independent variables and the pools of uPOM, pPOM, pcPOM, and CPOM as the dependent variables. The adjusted R^2 of the regression equation was used to evaluate the relative importance of factors on the SOC pools. As shown in Table 6, soil depth could independently explain 42.3 % of the variance in the SOC pool of uPOM, followed by pH and CaO with cumulative loading of 72.3 % for the variation of the uPOM pool. Soil depth could also independently explain 30.4 % of the variance in the pPOM pool, followed by SWC, Na_2O, pH,

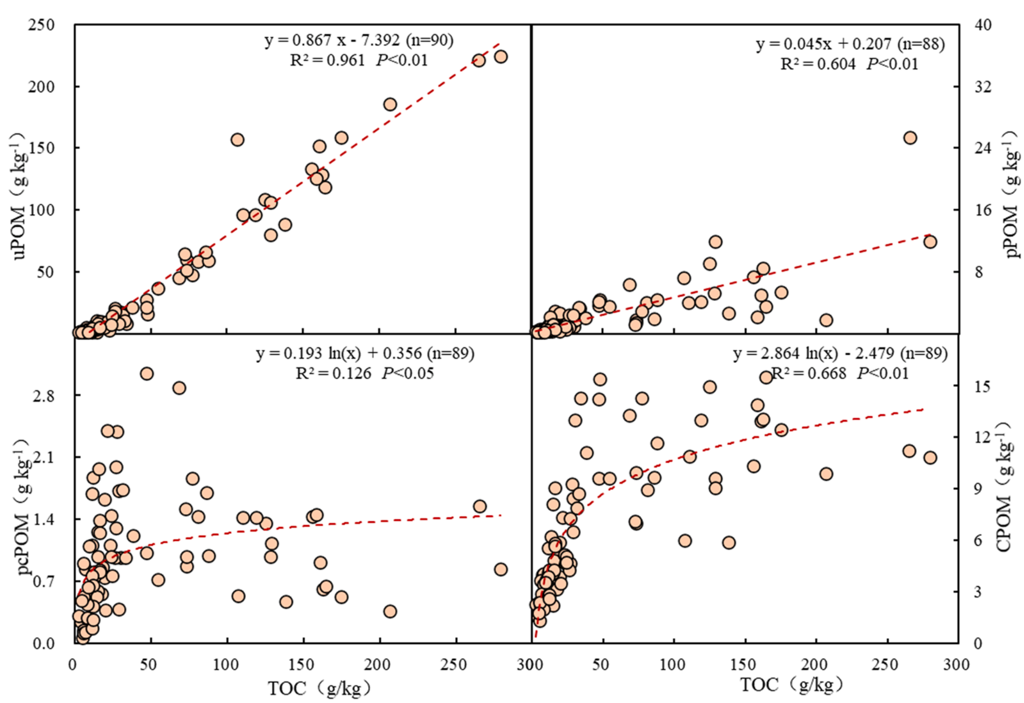

Fig. 6. Relationships between soil TOC and SOC pools (uPOM, pPOM, pcPOM, and CPOM). The trendlines are displayed as dash lines. See Table 1 for SOC pools abbreviations.

Table 5
Correlation coefficients between SOC pools and soil physicochemical properties.

	uPOM	pPOM	pcPOM	CPOM
SWC (n = 73)	0.738**	0.528**	0.244*	0.564**
BD (n = 50)	−0.400**	−0.486**	−0.440**	−0.516**
pH (n = 79)	−0.587**	−0.348**	0.001	−0.411**
NH_4^+-N (n = 82)	0.296**	0.171	0.229*	0.382**
AK (n = 83)	0.436**	0.317**	0.000	0.348**
AP (n = 72)	0.091	0.152	−0.119	0.050
OP (n = 87)	0.792**	0.532**	0.172	0.555**
Na_2O (n = 75)	−0.687**	−0.590**	−0.221	−0.664**
MgO (n = 75)	−0.667**	−0.435**	0.246*	−0.417**
Al_2O_3 (n = 75)	−0.709**	−0.415**	0.325**	−0.375**
K_2O (n = 75)	−0.537**	−0.318**	0.055	−0.263*
CaO (n = 75)	0.501**	0.386**	0.244*	0.496**
Fe_2O_3 (n = 75)	−0.280*	−0.198	0.144	−0.259*

Explanation: *$P < 0.05$, **$P < 0.01$. See Table 4 for abbreviations of soil physicochemical properties. See Table 1 for SOC pools abbreviations.

Table 6
Stepwise regression results (SOC pools of four protection mechanisms as the dependent variables and forest type, forest age, soil depth, and soil physicochemical properties as the independent variables).

SOC pool	Variable	Adjusted R^2	F	P
uPOM	Soil depth	0.423	26.685	<0.001
	Soil depth, pH	0.607	28.001	<0.001
	Soil depth, pH, CaO	0.723	31.392	<0.001
pPOM	Soil depth	0.304	15,836	<0.001
	Soil depth, SWC	0.480	16.689	<0.001
	Soil depth, SWC, Na_2O	0.585	16.961	<0.001
	Soil depth, SWC, Na_2O, pH	0.626	15.525	<0.001
	Soil depth, SWC, Na_2O, pH, K_2O	0.666	14.539	<0.001
pcPOM	BD	0.404	24.689	<0.001
	BD, CaO	0.480	17.157	<0.001
	BD, CaO, OP	0.566	16.202	<0.001
CPOM	Na_2O	0.410	25.316	<0.001
	Na_2O, SWC	0.588	26.017	<0.001
	Na_2O, SWC, Soil depth	0.658	23.431	<0.001

See Table 4 for abbreviations of soil physicochemical properties. See Table 1 for SOC pools abbreviations.

and K_2O with the cumulative loading of 66.6 % for the variation in the pPOM pool. The pools pcPOM is mainly archived in microaggregates particulates (<53 μm), BD could independently explain 40.4 % of the variance in the pcPOM pool, followed by CaO and OP with the cumulative loading of 56.6 % for the variation of the pcPOM pool. Na_2O could independently explain 41.0 % of the variance in the CPOM pool, followed by SWC and soil depth with the cumulative loading of 65.8 % for the variation in the CPOM pool. The main source of organic matter in forest soil is dead leaves and their decomposition, resulting in an overwhelming trend of SOC in the topsoil. Thus, soil depth was extracted as the dominant factor of the SOC pools of uPOM, pPOM, and CPOM. It showed that the crustal or petrogenetic elements (Ca, Na, K, et al.) may play more prominent role in the variation of SOC pools achieved in fine soil particulates.

4. Discussions

Previous attempts to quantify SOC sequestration capacity mainly focused on clay adsorption protection, neglecting the effects of soil structure protection and biochemical protection (Stewart et al., 2008). Soils' ability to preserve C by clay adsorption is limited and dependent not only on the chemical connection of clay, but also on aggregate protection and biochemical resistance (Baldock and Skjemstad, 2000). The stability of SOC is a result of chemical interactions with clay particulates, physical protection in microaggregates, and biochemical mechanisms of organic compounds (Six et al., 2002). Theoretically, the accumulation of the four SOC pools, including free activity, physical protection, chemical protection, and biochemical protection C pools, could lead to SOC pool saturation. Generally, the fractions of uPOM and pPOM decompose quickly, with a short turnover time, and are sensitive to environment changes, which can be used as a precursor for the changes of the soil C pool. The inert fractions of CPOM and pcPOM exhibit slow response to changes in surrounding conditions and decomposition, thereby promoting long-term preservation (Dou et al., 2011). These fractions can serve as an indicator to forecast soil C saturation (Zhang et al., 2014a) and are essential in predicting long-term C storage in soil.

4.1. Characteristics of SOC pools

In soils from the *Larix gmelinii* forest, the SOC pools followed the order of uPOM > CPOM > pPOM > pcPOM. Due to the abundant organic inputs (especially dead leaves and branches) and less human interference, more active organic C (uPOM, mainly > 250 μm) has been accumulated in the forest soil. Therefore, the uPOM pool was the largest, accounting for >50 % of TOC in soils. Similar as previous study (Chung et al., 2010), the chemical protection mechanism sequestered more organic C than the physical protection mechanism, highlighting the distinctive role of physical protection mechanism. The silt-associated C pool under the physical–chemical protection and chemical protection mechanism were greater than clay-associated C pool, consistent with the findings of Li et al. (2014) and Stewart et al. (2009). Theoretically, physical–chemical protection and chemical protection mechanisms inherently related to soil texture, especially the relative proportion of silt and clay. Meanwhile, the stability and C adsorption capacity of silt and clay differed (Stewart et al., 2008).

The dominant SOC pools in forest soils are unstable and physically stable C pools (Xu et al., 2018). The unstable C pool (uPOM pool), composed of the pools of cPOM and LF, is primarily formed by semi-decomposed plant residues and has not been stabilized on soil minerals by physical adsorption or chemical binding. As labile SOC, cPOM and LF are the key substances of the biochemical processes in soils, providing energy and nutrients for soil biological activities (Zhang et al., 2013). Natural ecosystems, such as forests, typically contribute more C to the soil through litter, and their C inputs in the soil profile are usually greater than those of farming systems. Furthermore, the litter of *Larix gmelinii* decomposes slowly due to more difficult-to-decompose substances such as lignin, tannin, resin, and wax (Grünzweig et al., 2003). Meanwhile, due to the dense coverage of the withered needles on the soil surface, which reduces the oxygenysis by air, it is more conducive to the preservation of SOC. The pool of CPOM composes of clay- (dClay) and silt-associated C(dSilt) pools, in which its stability mechanism depends on its chemical composition and the turnover period ranges from tens to hundreds of years (Torn et al., 2013). The protection mechanism is significant for soil C sequestration. Hassink and Whitmore (1997) reported that the amount of organic C sequestered by silt and clay particulates had a saturation value dependent on the specific surface area of the particulates. The silt- and clay-associated C pools had approached or reached saturation in the *Larix gmelinii* forest soils (Fig. 6), indicating that more organic C will be enriched in larger aggregates and fixed in the relatively active uPOM pool, suggesting a more sensitive response to ecosystem change.

4.2. Synergistic effects on SOC pools

Previous studies on *Larix gmelinii* plantation revealed the significant effects of soil depth, vegetation species, and tree age-group on the SOC pools (Wang et al., 2014; Wang et al., 2017). Our study was conducted in the primeval forest region of the northern Greater Khingan Mountains, known for its vigorous vegetation growth, dense litter layer, and absence of human intervention. In addition, SOC significantly accumulated in the top soils (Wang et al., 2021). This may be attributed to the predominate root system of *Larix gmelinii* being concentrated in the topsoil, resulting in the preservation of humus generated from decomposition in this layer. More labile organic C accumulation was consistently observed in forest soils, attributed to the plentiful inputs of organic matter, such as litters, coupled with the absence of anthropogenic activities. Therefore, compared with forest age and type, the effect of soil depth on the SOC pools was more significant. This was reflected in the results of MANOVA and stepwise regression analysis (Table 2, Table 3, and Table 6). Consistent with our hypothesis, the soil depth presented a significant single-factor influence; the growth of trees and the composition of understory vegetation both have an impact on the SOC pools; while synergistic effects were observed on soil C sequestration of *Larix gmelinii* forest in different protection mechanisms, attributed to multiple factors such as forest age × forest type, soil depth × forest type, soil depth × forest age, and soil depth × forest type × forest age. It is important to note that these factors are interrelated and influence each other. Improving the growth of trees and promoting the growth of diverse understory vegetation can enhance the accumulation and stabilization of SOC.

4.3. SOC pools associated with soil physicochemical properties

As a whole, the SOC pools significantly correlated with TOC in the soils from the *Larix gmelinii* forest. It was noteworthy that the uPOM pool, accounting for approximately half of TOC, increased linearly with TOC in the soils (Fig. 6), suggesting that the retention capacity of this pool was still not saturated and had the potential to sequester and stabilize organic C under current conditions. The input–output balance of C is critical to the uPOM pool under the unprotection mechanism. Plant residues are often included in the uPOM pool during plants converting to SOC, resulting in significant effects on the magnitude and stability of the uPOM pool (Xu et al., 2018). Therefore, due to the co-effects of soil temperature, humidity, and biodegradability of substance, the relationship between uPOM pool and TOC was not the same in different conditions.

It was reported that the pPOM pool had the maximum level of C sequestration (Stewart et al., 2008; Hai et al., 2010), associating with soil type, texture and moisture, and management measures. Additionally, the organic C content in iPOM soil fraction (pPOM pool) linearly correlated with TOC contents in soils, but not TOC saturation (Yang, 2018; Chung et al., 2010). A significantly linear positive correlation between the pPOM pool and TOC was also observed in soils from the *Larix gmelinii* forests (Fig. 6). Stewart et al. (2008) found that the relationship between the pcPOM pool and TOC followed both linear and saturation models. Thus, the significant logarithmic relationship observed between the pcPOM pool and TOC in our study area indicated the increasing saturation with increased TOC.

Like the findings of Yang (2018) and Stewart et al. (2008), as the second largest SOC pool, the increasing trend of the CPOM pool with TOC was also observed in the *Larix gmelinii* forests, in which the logarithmic relationship suggested that the CPOM pool may be approaching or have achieved its maximum capacity at some sites, agreeing with previous studies on the saturation of the CPOM pool (Sleutel et al., 2006; Stewart et al., 2007; Gulde et al., 2008). Organic C can be adsorbed by clay and silt in soils and converted to nonlabile mineral-bound organic C; however, due to the limited binding ability, the adsorption rate of clay and silt will decrease and the clay- and silt-associated C pools quickly peak with the increase of SOC (Xu et al., 2018). As a result, in the *Larix gmelinii* forests, SOC was largely retained in the uPOM pool, causing more effective nutrient release and transformation from these coarse particulates (>250 μm). A large accumulation of organic matter will lead to an increase in the labile pool of uPOM rather than the stable pools of CPOM and pcPOM. As a result, it is possible to achieve effective SOC fixation through reasonable human interventions, in turn the improvement of C sinks and the reduction of C emissions (Xu et al., 2018).

There were some differences in the relationship between the SOC pools and soil physicochemical properties in the *Larix gmelinii* forests. The four SOC pools significantly positively correlated with SWC and negatively correlated with BD. The SOC pools of uPOM, pPOM, and CPOM significantly negatively correlated with pH. In the *Larix gmelinii* forests, soil moisture and acidity can influence the formation and stability of soil aggregates, in turn the SOC pools (Wang et al., 2021). The pcPOM pool significantly positively correlated with the NH_4^+-N; While the other three SOC pools significantly positively correlated with OP, AK, and NH_4^+-N, similar as Jiang (2016)'s findings. The pool of uPOM is mainly composed of fresh and incompletely decomposed plant residues. Nitrogen, phosphorus, and potassium fertilizer can significantly accelerate the growth of plants and their roots, resulting in an increased input of plant residues to the soils and a consequent increase of the uPOM pool in soils (Yang, 2018; Tian et al., 2017). As the nutrient levels increase, the microbial activities will accelerate the SOC formation and provide more cementitious substances for soil aggregation, enhancing the micro aggregation of soil to a certain degree (Tian et al., 2017). On the other hand, the turnover rate of microaggregates is slower than macroaggregates, which aids in the physical protection of organic C (Zhang et al., 2014b). Hence, more SOC may be preserved in the pPOM pool (Yang, 2018) since the pPOM pool is the microaggregate-enclosed stable C. Stewart et al. (2009) found that the response of free clay and silt to C addition was faster than that of clay and silt protected by microaggregates. We observed a better trend of soil nutrients associated with the CPOM pool than the pcPOM pool, indicating the different response of free silt and clay to the soil environment compared to microaggregate-protected silt and clay, as a result of their different protection mechanisms on C pools. Similarly, Yang (2018) also observed that microaggregate-protected silt and clay, as well as free silt and clay, responded to long-term fertilization in various ways, with an enhanced effect on the pcPOM pool for a long time.

5. Conclusion

Due to the abundant organic inputs, such as dead leaves and branches, and the lack of human interference, the active organic C was mainly the uPOM pool (mainly > 250 μm) in soils from the *Larix gmelinii* forest, accounting for >50 % of TOC. Comparatively, the effect of soil depth on the SOC pools of each protection mechanism was more significant than that of forest age and type. The pool of pcPOM significantly differed from forest types and ages, and the pool of CPOM was associated with the synergistic effects of forest age and type, and soil organic matter inputs. The pool of CPOM and pcPOM may be approaching or had achieved their maximum capacities; while the retention capacities of the uPOM and pPOM pool were still not saturated and had the potential to sequester and stabilize organic C under current conditions of the *Larix gmelinii* forests. In the *Larix gmelinii* forests, soil moisture and acidity can influence the formation and stability of soil aggregates, which in turn can affect the SOC pools. Additionally, the crustal elements (Ca, Na, K, et al.) may play more prominent role in the variation of SOC pools achieved in fine soil particulates. As a whole, soil depth, forest types, and tree growth presented both single-factor effects and multi-factor synergistic effects on soil C sequestration of *Larix gmelinii* forest in different protecting mechanisms, while such changes were strongly associated with soil N, P, and crustal elements. Therefore, it is possible to achieve effective SOC fixation through reasonable human interventions, in turn the improvement of C sinks and the reduction of C emissions.

CRediT authorship contribution statement

Bing Wang: Conceptualization, Data curation, Formal analysis, Investigation, Methodology, Data curation, Writing – original draft, Writing – review & editing, Funding acquisition. **Shuai Hao:** Investigation, Data curation. **Qiuliang Zhang:** Funding acquisition, 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

The data that has been used is confidential.

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