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Effects of Freeze–Thaw Cycles on Soil Aggregate Stability and Organic Carbon Distribution Under Different Land Uses

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Abstract

Soil aggregates are critical determinants of soil erosion resistance and nutrient retention capacity, while freeze–thaw cycles (FTCs) induce the structural reorganization of soil aggregates, thereby altering soil stability and influencing soil organic carbon (SOC) sequestration. This study was located in the Minjia River Basin in the typical seasonal freeze–thaw areas of the Loess Plateau and aimed to quantify the effects of FTCs on soil aggregate stability and SOC content under different land use types. Farmland, grassland, and forestland with more than 20 years of usage in the region were selected, and a 0–20 cm soil layer was subjected to seven FTCs (−8 °C to 20 °C), followed by wet and dry sieving classification, focusing on soil aggregate distribution, aggregate stability, mean weight diameter (MWD), geometric mean diameter (GMD), aggregate particle fractal dimension (APD), and SOC content of the aggregate. The results showed that soil aggregates in all land use types were dominated by macroaggregates (>2 mm), with the proportion in forestland (61–63%) > grassland (54–58%) > farmland (38–51%). FTCs enhanced aggregate stability across all land use types, especially in farmland. Concurrently, FTCs reduced the SOC content in all aggregate size fractions, with reduction rates ranging from farmland (9.00–21%) to grassland (4–26%) to forestland (5–31%). Notably, FTCs significantly increased the contribution of 2–5 mm water-stable (WS) aggregates to SOC sequestration, with increment rates of 86% (farmland), 80% (grassland), and 86% (forestland). Furthermore, FTCs altered the correlation between SOC content and aggregate stability. Specifically, the positive correlations of SOC with MWD and GMD were strengthened in aggregates < 0.5 mm but weakened in aggregates > 0.5 mm. These findings advance our understanding of the coupled mechanisms underlying soil erosion and carbon cycling across land uses under freeze–thaw, providing a theoretical basis for ecosystem restoration and optimized soil carbon management in cold regions.

Keywords: soil aggregates; soil organic carbon; freeze–thaw; land use

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1. Introduction

Soil aggregates constitute a pivotal component of soil structure, formed by soil particles bound together through organic matter (such as fungal hyphae and polysaccharides) and inorganic substances, and their stability exerts a profound influence on the preservation of soil biological functions and resistance to erosion [1]. The composition and stability of soil aggregates exhibit a strong correlation with soil organic carbon (SOC) content, as SOC—predominantly enriched within aggregate interiors—serves as a critical binding agent in soil and is intricately associated with aggregate formation. In turn, SOC is encapsulated within

the pores of soil aggregates, isolating it from decomposer microorganisms and extracellular enzymes, thereby facilitating the long-term sequestration of carbon in soil systems [2]. Freeze–thaw cycles (FTCs) induce soil structural degradation and modifications in soil properties via the freezing and thawing of soil matrices [3]. During freezing, soil water turns into ice crystals, a process accompanied by volumetric expansion. This expansion generates cryostatic pressure, pushing soil particles apart and disrupting aggregate bonds. Upon thawing, the ice melts, leading to a loss of structural support and potentially increasing soil erosion intensity [4]. Notably, FTCs can modulate aggregate stability through structural rearrangements of soil, leading to shifts in the aggregation capacity of soil particles [5] and disturbances in the system dynamics of SOC encapsulated within aggregates through redistribution [6,7].

FTCs can disrupt soil macroaggregates (>2 mm) and promote microaggregate (<0.25 mm) formation, thereby facilitating the release of SOC [8]. However, inconsistencies exist in the literature regarding FTC-induced changes in aggregate size distribution. Wang et al. [9] demonstrated that FTCs increased the abundance of aggregates >1 mm while reducing those <1 mm, whereas Yao et al. [10] reported that FTCs diminished aggregate water stability by increasing the content of particles <0.25 mm and decreasing aggregates >2 mm. Li et al. [11] further revealed that larger aggregates (>5 mm, 3–5 mm, and 1–3 mm) exhibited decreased stability with declining freezing temperatures and increased FTC frequency, whereas smaller aggregates (0.5–1 mm and 0.25–0.5 mm) became more stable under such conditions. Beyond aggregate quantity dynamics, FTCs influence SOC cycling through multiple pathways. Koponen et al. [12] highlighted that low-temperature (-17.3 to 4.1 °C) FTCs disrupt leaf litter decomposition, enhance root mortality, and lyse microbial cells, thereby releasing labile organic matter, stimulating surviving microorganisms, and accelerating mineralization which may deplete native SOC stocks, leading to reduced accumulation or even net losses. Notably, while the initial disruptive phase of FTCs exert a more pronounced impact on SOC loss, other studies suggest that frequent FTCs may enhance the physical protection of SOC by aggregates, with this effect being particularly evident in high-SOC aggregates [13]. Chistensen et al. [14] conducted laboratory FTC simulations and found that variable freezing temperature regimes impaired aggregate stability indicators MWD and GMD, yet SOC content increased with escalating FTC frequency—this contradiction underscores the complexity of FTC-mediated SOC dynamics, necessitating further investigation into context-dependent mechanisms. Inconsistencies exist in the literature regarding the direction and magnitude of these effects, with findings that are highly dependent on soil type, experimental treatments, and crucially, land use context.

Furthermore, different land use types (e.g., farmland, forestland, grassland, etc.) additionally regulate the magnitude of soil disturbance induced by FTCs via variations in vegetation cover, agricultural practices, and organic matter inputs. For instance, forestland and grassland soils may exhibit enhanced aggregate resistance to FTCs owing to their elevated organic matter content and root exudates [15], whereas soil aggregates in long-term tilled farmland soils may be more prone to disaggregation and the release of sequestered SOC during FTCs, attributable to frequent mechanical disturbances and diminished organic matter inputs [16]. Existing studies have reported inconsistent results, which are highly dependent on soil type and experimental treatments; moreover, these studies often focus on a single land use type, leading to a paucity of research on the interactive effects of FTCs and land use on soil aggregates and SOC. Furthermore, current research has predominantly concentrated on the frozen soil region characterized by prolonged freeze–thaw periods (e.g., the black soil region of Northeast China), with relatively few studies conducted in regions with shorter freeze–thaw durations.

Accordingly, this study selected three land use types in the Minjia River Basin of Shaanxi Province as the study sites. This basin is typical of a seasonal freeze–thaw area and contains farmland, grassland, and forestland. Employing freeze–thaw cycle (FTC) simulation experiments integrated with dry–wet sieving techniques, this research aimed to (1) quantify the land use type changes in SOC content and aggregate size distribution induced by FTCs; (2) evaluate the differential response of aggregate stability indices (MWD, GMD, APD) to FTCs across farmland, grassland, and forestland; (3) determine how FTCs alter the relationship between SOC and aggregate stability, and identify the aggregate fraction that becomes the dominant SOC pool post FTC. Ultimately, the findings of this study seek to provide scientific insights for soil carbon management, conservation, and sustainable utilization in freeze–thaw-affected regions.

2. Materials and Methods

2.1. Sample Collection

The study region is located within the Minjia River Basin, Shaanxi Province, with a total area of 28.19 km². Geographically, the basin lies within the transition zone between the northern warm temperate zone and the mild temperate zone, characterized by a frost-free period of 194 days and a mean annual temperature of 10.3 °C. The mean annual rainfall is 890.4 mm and the average absolute altitude is 1240 m. The basin exhibits a diverse array of soil types, with skeletal soils, mountain brown loam, and silt–sand soils predominating. Land use is dominated by forestland and agricultural land, with forestland coverage exceeding 78% and vegetation coverage exceeding 89%. The geographical location of the study area and the spatial distribution of sampling sites are illustrated in Figure 1.

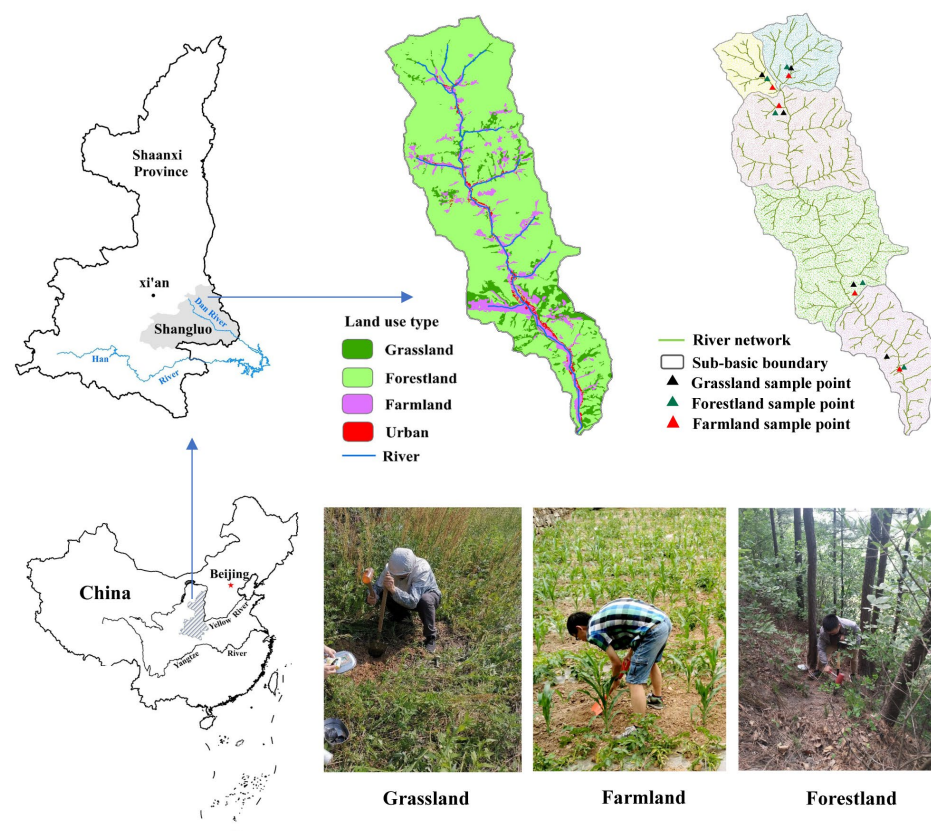


Figure 1. Location of the study area and spatial distribution of sampling points.

Soil sampling was conducted in the Minjia River Basin. A stratified sampling design was adopted, wherein five sub-basins were selected across upstream, middle, and down-

stream reaches, representing gradients in land use structure. Within each sub-basin, three land use types (farmland, forestland, grassland) were established as experimental plots, resulting in 15 plots. Plot characteristics, including altitude, slope gradient, vegetation cover, and anthropogenic disturbances (e.g., fertilization regimes, tillage practices), are summarized in Table 1. At each plot, six subsamples were randomly collected using a systematic random sampling method. Soil profiles were excavated with a shovel, and in situ soil samples (0–20 cm depth) were collected using aluminum containers to preserve aggregate structure. Additionally, undisturbed soil cores were collected using a core sampler (5 cm diameter $\times$ 5 cm height) from the same profiles to determine bulk density and gravimetric moisture content. Prior to laboratory analysis, bulk soil samples were transported to the laboratory, gently broken into aggregates (<1 cm diameter) to maintain soil structure, manually cleared of visible plant residues and stones, and air-dried in a cool, well-ventilated environment. A total of 90 bulk soil samples and 15 core samples were collected. All analytical parameters were determined in triplicate, with results reported as the mean of three replicate measurements.

Table 1. Basic characteristics of sample plots in the study area.

Sub-Basin	Land Use Types	Altitude (m)	Vegetation Characteristics
1	Farmland	1389.00	Corn and soybeans, with plowing, no fertilizers
	Grassland	1407.00	Daisies, lavender, and ginger
	Forestland	1405.00	Pine trees and walnut trees
2	Farmland	1261.00	Potatoes and corn, with plowing, no fertilizers
	Grassland	1339.00	Daisies and lavender
	Forestland	1338.00	Pine trees and walnut trees
3	Farmland	1263.00	Potatoes and corn, with plowing and fertilizers
	Grassland	1269.00	Water mugwort
	Forestland	1263.00	Persimmon trees and walnut trees
4	Farmland	1172.00	Potatoes, corn, and scallions, with plowing and fertilizers
	Grassland	1172.63	Ginger and lavender
	Forestland	1072.63	Persimmon trees and pine trees
5	Farmland	1015.65	Potatoes, corn, and ginger, with plowing and fertilizers
	Grassland	1088.26	Daisies and lavender
	Forestland	1172.63	Persimmon trees and walnut trees

2.2. Experimental Design

Prior to freeze–thaw treatments, soil aggregate samples were equilibrated at a controlled ambient temperature (20 °C) for 24 h to achieve a stable moisture content (15% moisture content, based on the average of long-term monitoring results) and hermetically sealed with plastic film to minimize evaporative moisture loss. Climatic data from the study region (December 2019–June 2020) indicated a mean temperature of 3.5 °C, with extreme values ranging from −8.4 °C to 25.6 °C; freeze–thaw events typically occurred within the temperature range of −5 °C to −10 °C. Based on preliminary trials, a 12 h freeze–thaw cycle was determined to ensure the complete phase transition of soil moisture. According

to the fact that seven FTCs are the critical number for changes in soil physicochemical properties [17–19], experimental treatments consisted of seven consecutive FTCs, with three replicate samples established per treatment. The freezing temperature was set at -8°C , the thawing temperature at 20°C , and the freeze/thaw duration was 12 h. Subsequent to freeze–thaw treatments, samples were air-dried under laboratory conditions and subjected to dry–wet sieving analysis to determine aggregate size distribution and associated SOC content in non-water-stable and water-stable aggregates.

2.3. Analytical Methodology

Soil aggregates were classified into six size fractions, >5 mm, 2–5 mm, 1–2 mm, 0.5–1 mm, 0.25–0.5 mm, and <0.25 mm, using a combination of dry sieving and wet sieving methods as described below. Dry sieve method: A nested sieve assembly was prepared with mesh sizes of 5 mm, 2 mm, 1 mm, 0.5 mm, and 0.25 mm, stacked in descending order. Air-dried soil samples were uniformly distributed on the top sieve, and the sieve stack was manually agitated for 5 min to facilitate size separation. Aggregates retained on each sieve were sequentially collected from top to bottom, manually cleared of visible plant debris and stones, and weighed to determine the mass distribution of each size fraction. Wet sieve method: For wet sieving, a vibrating sieve apparatus was used with the same nested sieve sequence. A 500 g subsample of air-dried soil was carefully transferred into a 1 L beaker, and deionized water was slowly added to submerge the soil aggregates, allowing them to equilibrate by soaking for 10 min. The sieve stack was then immersed in a water bath, and the apparatus was activated at a vibration frequency of 30 cycles per minute for 5 min to separate water-stable aggregates. Aggregates retained on each sieve were rinsed gently to remove fines, transferred to pre-weighed aluminum dishes, and oven-dried at 40°C to constant weight. The mass of each water-stable aggregate fraction was recorded after cooling to room temperature.

Soil bulk density (SBD) was determined using the core sampler method (5 cm diameter $\times$ 5 cm height). Soil particle size distribution was analyzed with a laser diffraction particle size analyzer (Mastersizer 2000, Malvern Panalytical, Malvern, UK). SOC concentration was measured via the potassium dichromate oxidation method with external heating by the Bisutti method (2004) [20]. Soil total phosphorus (STP) concentration was determined using an automated discrete chemical analyzer (CleverChem 200, DeChem-Tech, Hamburg, Germany) by the Bowman method (1988) [21]. Soil total nitrogen (STN) content was analyzed with a Foss 8400 automatic Kjeldahl nitrogen analyzer by the Bremner method (1960) [22]. Soil physico-chemical properties of the study area are summarized in Table 2.

Table 2. Soil physico-chemical properties of the study area.

Land Use	Clay (<0.002 mm)/%	Silt (0.002–0.05 mm)/%	Sand (0.05–2 mm)/%	SBD/ (g cm^{-3})	SOC/ (g kg^{-1})	STN/ (g kg^{-1})	STP/ (g kg^{-1})
Farmland	0.36	72.07	27.57	1.28 ± 0.11	15.37 ± 3.61	1.95 ± 0.70	0.81 ± 0.33
Grassland	0.27	61.64	38.09	1.24 ± 0.13	22.36 ± 6.83	1.90 ± 0.74	0.71 ± 0.32
Forestland	0.25	51.56	48.19	1.16 ± 0.12	21.99 ± 2.39	3.41 ± 1.97	0.79 ± 0.34

2.4. Data Analysis

Soil aggregate stability was evaluated using three key indices, mean weight diameter (MWD), geometric mean diameter (GMD), and aggregate particle fractal dimension (APD), which were calculated according to the standard methods described by Kemper and Rosenau (1986) [23]. These indices were calculated using the following formulae:

$$MWD = \sum_{i=1}^n \bar{x}_{ai} w_{ai} \quad (1)$$

$$GMD = \exp\left(\sum_{i=1}^n w_i \ln \bar{x}_{ai}\right) \quad (2)$$

$$(3 - APD) \lg(\bar{x}_{ai}/x_{max}) = \lg[w(\delta < \bar{x}_{ai})/w_0] \quad (3)$$

where $\bar{x}_{ai}$ is the mean diameter of the i size soil aggregate size class, mm; w_{ai} is the mass fraction of the i soil aggregates size class, %; x_{max} is the mean diameter of the largest aggregate size class, mm; and $w(\delta < \bar{x}_{ai})/w_0$ is the cumulative mass fraction of aggregates smaller than the i size class, %.

The formula for calculating the contribution of aggregates to organic carbon content across particle size is as follows:

$$R_{ai} = C_{ai} \times \omega_{ai} / \sum_{i=1}^n C_{ai} \omega_{ai} \quad (4)$$

where R_{ai} is the contribution of SOC from the i aggregate size, %; C_{ai} is the SOC content of the i aggregates size, g kg⁻¹; and ω_{ai} is the mass fraction of the i aggregate size, %.

Data variability was assessed using a one-way analysis of variance (ANOVA), followed by the least significant difference (LSD) test for post hoc comparisons. Pearson's correlation analysis was conducted to examine relationships between variables.

3. Results

3.1. Distribution Characteristics of Soil Aggregates

The distribution characteristics of soil aggregates in different land use types before freeze–thaw are shown in Figure 2A. Non-water-stable (NWS) aggregates showed no differences among land use types ($p > 0.05$, one-way ANOVA), but significant variations were observed across aggregate size ($p < 0.05$). Soil aggregates were predominantly composed of macroaggregates (>2 mm), with the 2–5 mm size range accounting for the highest proportion (31.53–36.26%), while the 0.25–0.5 mm size range represented the lowest proportion (5.09–7.59%).

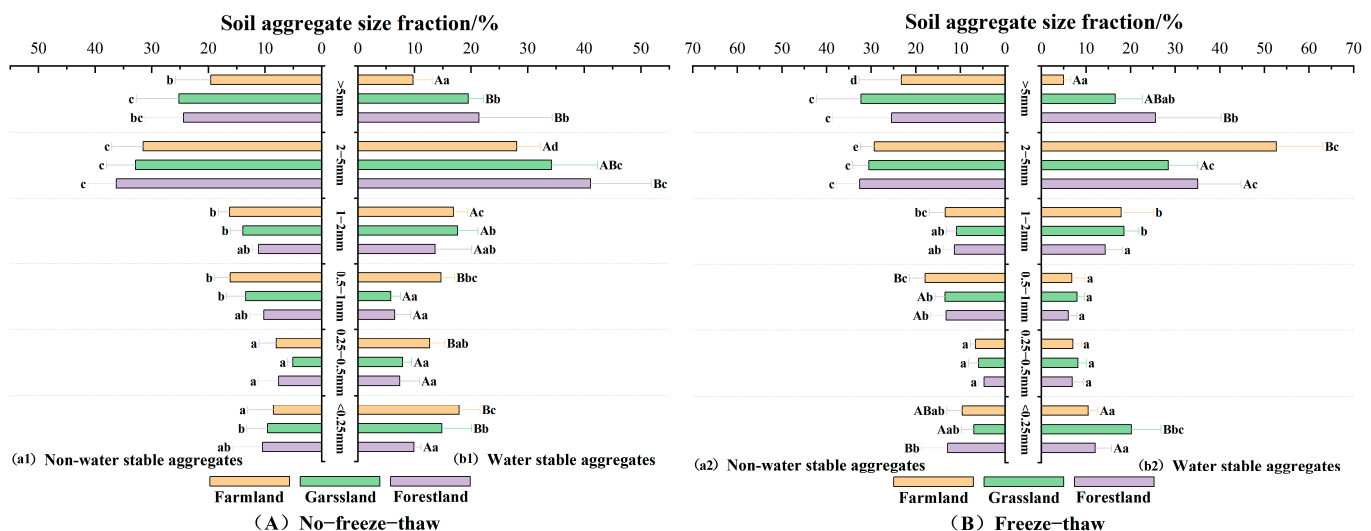


Figure 2. Distribution characteristics of soil aggregates. Note: capital letters indicate the significance variations between different land use types for the same particle size; lower case indicates the significance variations between different particle sizes for the same land use type ($p < 0.05$).

For water-stable (WS) aggregates, significant differences were observed among land use types ($p < 0.05$), with the exception of the 1–2 mm size range; additionally, significant variations were detected across aggregate size ($p < 0.05$). The WS aggregates were predominantly distributed in the 2–5 mm and <0.25 mm size ranges, with proportions ranging

from 28.06 to 41.11% and 9.90–17.92%, respectively. Specifically, forestland and grassland exhibited significantly higher proportions of >5 mm aggregates and significantly lower proportions of 0.5–1 mm and 0.25–0.5 mm aggregates compared with farmland ($p < 0.05$). Furthermore, forestland had a significantly lower proportion of <0.25 mm aggregates relative to both farmland and grassland ($p < 0.05$). These findings indicate that forestland aggregates possess superior stability under saturated soil conditions.

In both NWS aggregates and WS aggregates, the relative proportion of large aggregates (>2 mm) across land use types followed the order forestland (61–63%) > grassland (54–58%) > farmland (38–51%). Notably, the proportion of large aggregates in forestland did not differ significantly between NWS and WS fractions, indicating greater soil structural stability. In contrast, farmland and grassland exhibited significantly lower proportions of >5 mm aggregates and higher proportions of <0.25 mm aggregates in WS aggregates compared with NWS aggregate fractions ($p < 0.05$). These findings suggest that large aggregates in farmland and grassland are prone to disruption under saturated conditions, thereby facilitating the formation of microaggregates.

The distribution characteristics of soil aggregate particle sizes under different types of land use after freeze–thaw are shown in Figure 2B. For NWS aggregates, significant differences were observed among land use types for the 0.5–1 mm and <0.25 mm size fractions ($p < 0.05$), with additional significant variations detected across aggregate size fractions ($p < 0.05$). NWS aggregates were dominated by large macroaggregates (>2 mm), with the 2–5 mm size fraction accounting for the highest proportion (29.28–32.54%) and the 0.25–0.5 mm size fraction the lowest (4.67–6.61%); these patterns were consistent with pre-freeze–thaw observations. Relative to the pre-freeze–thaw period, all land use types exhibited an increasing trend in the proportion of >5 mm NWS aggregates, with grassland showing the most pronounced increase (28.15%); conversely, the 2–5 mm size fraction decreased, with forestland experiencing the largest reduction (10.25%). The size distribution of soil aggregates was altered by FTCs, with a decrease in 2–5 mm aggregates and an increase in both >5 mm and <2 mm aggregates.

For WS aggregates, significant differences were detected among land use types for the >5 mm, 2–5 mm, and <0.25 mm size fractions ($p < 0.05$), with additional significant variations across aggregate size ($p < 0.05$). The 2–5 mm size fraction accounted for the highest proportion of WS aggregates, with mass fractions ranging from 28.46% to 52.71%. Relative to the pre-freeze–thaw period, farmland exhibited an 87.82% increase in the 2–5 mm WS aggregate fraction and a 41.82% decrease in the <0.25 mm fraction—a pattern contrasting with the pre- to post-FTC changes observed in NWS aggregates. Following FTCs, an increase in macroaggregates coincided with a decrease in microaggregates in farmland soils. In contrast, grassland displayed the opposite trend: following FTCs, the mass fraction of macroaggregates decreased while that of microaggregates increased. This suggests that FTCs fragmented macroaggregates and promoted the formation of microaggregates in grassland soils.

3.2. Effects of Freeze–Thaw on the Stability of Soil Aggregates

The stability indexes of soil aggregates in different land use types are shown in Figure 3. Before freeze–thaw, for NWS aggregates, the MWD and GMD were in the order forestland > farmland > grassland, suggesting that forestland soils maintained a higher abundance and stability of large macroaggregates. Grassland exhibited the lowest GMD for NWS aggregates, suggesting a more uniform aggregate size distribution. Land use exerted minimal influence on the fractal dimension characteristics of NWS aggregates, with the APD ranging narrowly from 2.38 to 2.40; grassland displayed slightly lower AFD values than farmland and forestland. For WS aggregates, MWD was highest in forestland, with

significantly higher values in forestland and grassland than in farmland ($p < 0.05$). The GMD of WS aggregates followed the sequence forestland > grassland > farmland ($p < 0.05$), with farmland exhibiting the lowest GMD—indicating a more homogeneous aggregate size distribution under saturated conditions, likely reflecting structural vulnerability. Notably, forestland had significantly lower AFD values for WS aggregates than grassland and farmland, suggesting enhanced structural stability and erosion resistance in forestland soils, with reduced aggregate dispersion under rainfall events.

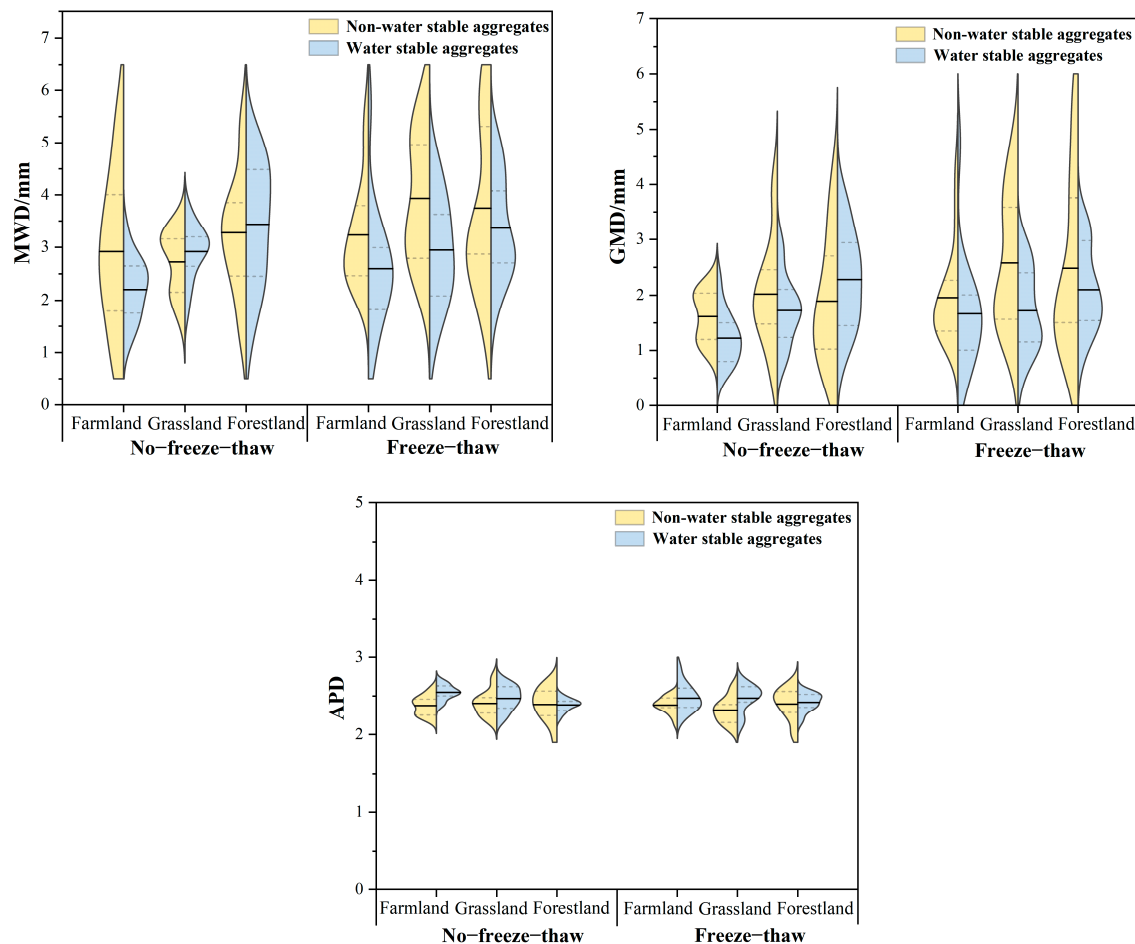


Figure 3. The stability indexes of soil aggregates in different land use types.

After freeze–thaw, the aggregate stability indices were observed with no significant differences among land use types ($p > 0.05$) for NWS aggregates and WS aggregates. However, the MWD and GMD of both NWS and WS aggregates were larger in forestland and grassland than in farmland, indicating superior aggregate stability in forestland and grassland soils under FTC conditions. The APD of NWS aggregates exhibited minimal variation across land use types under freeze–thaw conditions with a range of 2.32–2.39. Among WS aggregates, forestland displayed the lowest APD. Relative to the pre-FTC period, all land use types exhibited an increasing trend in MWD and GMD for NWS aggregates. For WS aggregates, only farmland showed a marked increase in MWD and GMD, while grassland remained relatively unchanged and forestland exhibited a slight decrease.

After freeze–thaw, NWS aggregates exhibited enhanced stability across all land use types; for WS aggregates, stability was enhanced in farmland and grassland but remained relatively unchanged in forestland. Additionally, the APD exhibited minimal variability

across land use types both before and after FTCs, indicating that aggregate structural homogeneity was not significantly altered by freeze–thaw-induced perturbations.

3.3. Effects of Freeze–Thaw on SOC in Soil Aggregates

The SOC contents of different soil aggregate particle sizes under different land use types are shown in Figure 4. For NWS aggregates, SOC contents across all size fractions followed the order forestland > grassland > farmland. Notably, forestland exhibited significantly higher SOC contents than farmland and grassland in the >5 mm and 2–5 mm sizes ($p < 0.05$). Across all land use types, SOC contents varied significantly among aggregate size fractions ($p < 0.05$), with each land use type exhibiting a unimodal distribution pattern: SOC contents increased with decreasing aggregate size up to a peak, then decreased. The maximum SOC content was observed in the 0.25–0.5 mm size fraction, with values of 18.40 g kg^{−1} (forestland), 15.12 g kg^{−1} (grassland), and 13.67 g kg^{−1} (farmland). Similarly, WS aggregates displayed SOC contents in the order forestland > grassland > farmland across all size fractions. The SOC content peaked in the 0.5–1 mm size fraction for all land use types, with values of 15.08 g kg^{−1} (forestland), 13.50 g kg^{−1} (grassland), and 11.04 g kg^{−1} (farmland). Forestland exhibited significantly higher SOC contents than farmland across all size fractions ($p < 0.05$); additionally, at the 1–2 mm, 0.25–0.5 mm, and <0.25 mm size fractions, forestland SOC contents were significantly greater than those of grassland ($p < 0.05$).

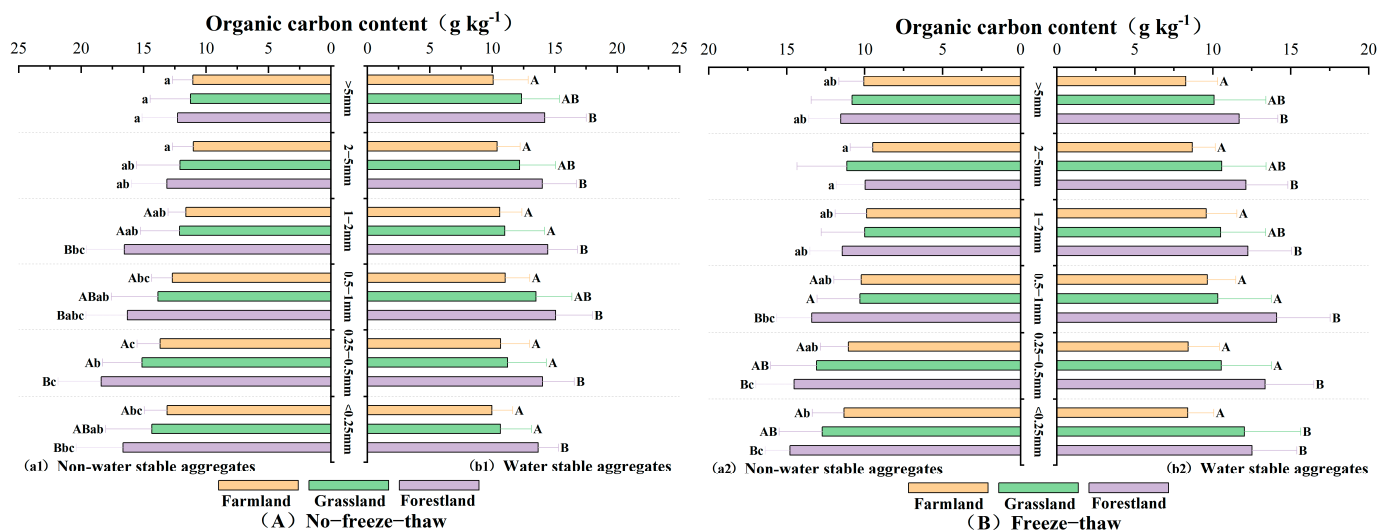


Figure 4. The SOC contents of different soil aggregates particle sizes under different land use types. Note: capital letters indicate the significance variations between different land use types for the same particle size; lower case indicates the significance variations between different particle sizes for the same land use type ($p < 0.05$).

These results indicate that both NWS and WS aggregates in forestland and grassland contained higher SOC contents than those in farmland. Specifically, NWS aggregates in the 0.25–0.5 mm size fraction and WS aggregates in the 0.5–1 mm size fraction exhibited a greater organic carbon sequestration capacity. Furthermore, SOC contents in WS aggregates were more evenly distributed across size fractions compared with NWS aggregates, suggesting a more stable SOC distribution in water-stable fractions.

After freeze–thaw, the SOC contents in both NWS and WS aggregates were higher in forestland and grassland than in farmland. Notably, for NWS aggregates, the 2–5 mm size fraction exhibited a distinct trend: SOC contents followed the order grassland > forestland > farmland, whereas all other size fractions maintained the pre-FTC pattern (forestland > grassland > farmland). For NWS aggregates, SOC contents peaked in the <0.25 mm size

fraction in farmland (11.32 g kg^{-1}) and forestland (14.79 g kg^{-1}), while grassland SOC contents peaked in the 0.25–0.5 mm size fraction (13.07 g kg^{-1}). For WS aggregates, SOC contents peaked in the 0.5–1 mm size fraction in both farmland (9.66 g kg^{-1}) and forestland (14.11 g kg^{-1}).

The SOC contents exhibited a reduction across all particle size fractions and land use types after freeze–thaw, with reduction rates ranging from 9.00 to 21.05% (farmland), 3.89–25.61% (grassland), and 4.75–30.88% (forestland). Notably, the magnitude of SOC reduction was more pronounced in non-water-stable (NWS) aggregates than in water-stable (WS) aggregates. These findings indicate that FTCs facilitate the release of SOC, with a more pronounced effect on NWS aggregates.

3.4. Contribution of Soil Aggregates to SOC

The SOC contribution proportions of soil aggregates across particle sizes under no-freeze–thaw condition are shown in Figure 5A. Across all land use types, NWS and WS aggregates exhibited consistent trends in SOC contribution. For NWS aggregates, the highest SOC contribution proportion was observed in the 2–5 mm size fraction, with values of 28.08% (farmland), 30.14% (grassland), and 28.08% (forestland). In contrast, the lowest contribution proportion occurred in the 0.25–0.5 mm size fraction, ranging from 9.27% (farmland) to 9.97% (forestland). For WS aggregates, the 2–5 mm size fraction also had the highest SOC contribution, with proportions of 27.89% (farmland), 28.99% (grassland), and 27.69% (forestland). The lowest SOC contribution was detected in the >5 mm size fraction, with values of 9.42% (farmland), 10.24% (grassland), and 9.78% (forestland).

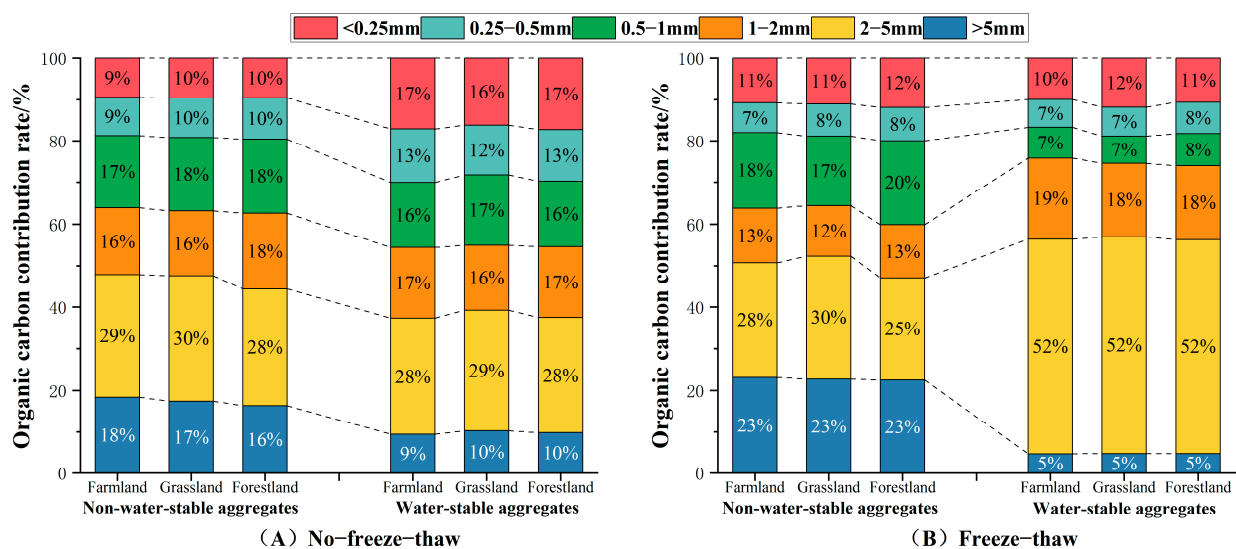


Figure 5. Contribution of soil aggregates to SOC.

The SOC contribution proportions of soil aggregates across particle sizes under freeze–thaw conditions are shown in Figure 5B. The highest SOC contribution proportion of NWS aggregates was observed in the 2–5 mm size fraction, with values of 27.53% (farmland), 29.55% (grassland), and 24.51% (forestland). In contrast, the lowest contribution proportion occurred in the 0.25–0.5 mm size fraction, ranging from 7.25% (farmland) to 8.06% (forestland). For WS aggregates, the highest SOC contribution proportion was detected in the 2–5 mm size fraction, with proportions of 51.77% (farmland), 52.23% (grassland), and 51.59% (forestland). Conversely, the lowest contribution proportion was found in the >5 mm size fraction, with values of 4.68% (farmland), 4.73% (grassland), and 4.72% (forestland).

The findings indicated that the SOC contribution of WS aggregates exhibited a more pronounced response to FTCs, with the most significant effect observed in the 2–5 mm size. Specifically, the SOC contribution of the 2–5 mm WS aggregates was enhanced by 85.63% (farmland), 80.18% (grassland), and 86.32% (forestland) relative to no-freeze–thaw conditions. These results confirm that the 2–5 mm size fraction of WS aggregates served as the primary contributors to SOC under freeze–thaw conditions.

3.5. Correlation Between Soil Aggregate Stability and SOC Content

The correlation between soil aggregate stability and SOC under no-freeze–thaw conditions is shown in Figure 6A. Correlations were assessed across the entire dataset. Across all particle size fractions, SOC exhibited positive correlations with both MWD and GMD and negative correlations with APD. Notably, SOC in the >5 mm aggregate size fraction displayed a significant positive correlation with GMD ($p < 0.05$), while SOC in the 2–5 mm size fraction exhibited significant positive correlations with both MWD and GMD ($p < 0.05$).

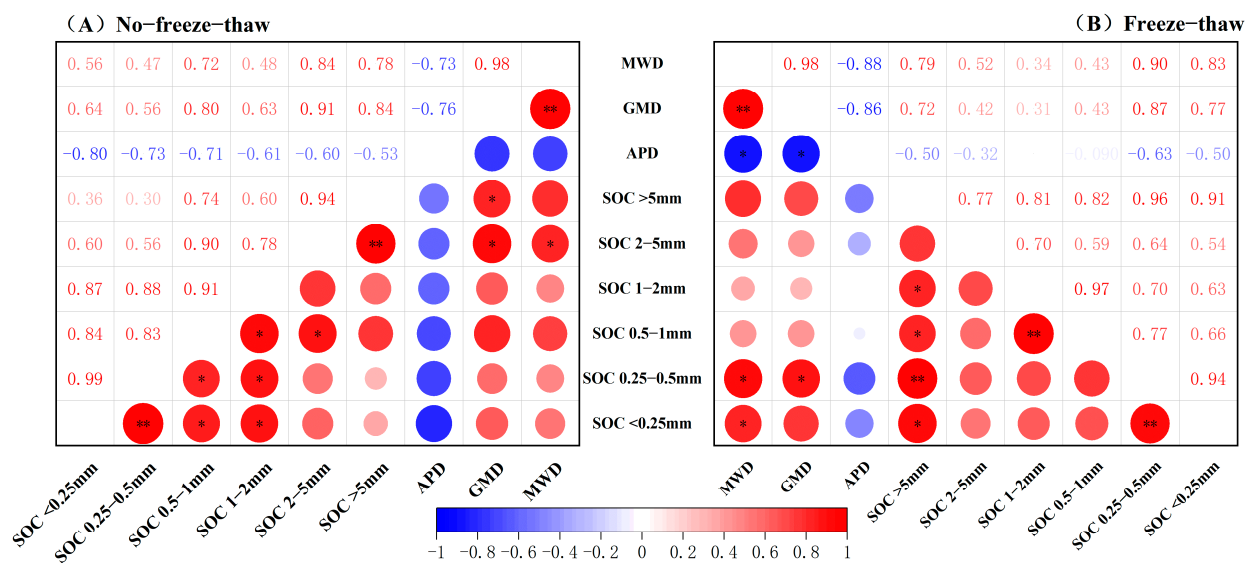


Figure 6. Correlation between soil aggregate stability and SOC content. Note: ** denotes a highly significant correlation ($p < 0.01$); * denotes a significant correlation ($p < 0.05$).

The correlation between soil aggregate stability and SOC under freeze–thaw conditions is shown in Figure 6B. Across all particle size fractions, SOC exhibited positive correlations with MWD and GMD, and negative correlations with APD—a pattern consistent with observations in no-freeze–thaw conditions, but the significance is different. Specifically, MWD and GMD were significantly positively correlated with the SOC content of aggregates in the 0.25–0.5 mm size fraction ($p < 0.05$), while MWD additionally showed a significant positive correlation with SOC in the <0.25 mm size fraction ($p < 0.05$).

The results indicated that FTCs weakened the correlation between the APD and SOC across all particle size fractions. For the correlations between MWD, GMD, and SOC, FTCs exhibited a threshold effect at the 0.5 mm particle size: specifically, the correlation was weakened in the >0.5 mm size fraction but strengthened in the <0.5 mm size fraction.

4. Discussion

4.1. Effects of Land Use Types on Soil Aggregate and Soil Organic Carbon Distribution

Soil aggregate size distribution serves as a key indicator of how distinct land use types influence soil structural stability. Previous studies have established that macroaggregates (>2 mm) contribute to optimal soil structural stability, with higher abundances of

these aggregates correlating with enhanced soil aggregation and structural integrity [24]. Consequently, soils with a higher macroaggregate content exhibit enhanced resistance to erosion. In the present study, soil aggregates were predominantly composed of macroaggregates (>2 mm), with the proportion in both NWS and WS aggregates following the order forestland (61–63%) > grassland (54–58%) > farmland (38–51%). These results indicate that forestland and grassland promoted the formation of macroaggregates, whereas farmland inhibited soil aggregation. These observations align with prior research demonstrating that farmland contains a significantly lower proportion of macroaggregates compared with grassland and forestland [25]. The enhanced macroaggregate proportions in forestland and grassland can be attributed to continuous organic matter inputs, root physical entanglement, binding by root exudates, and active microbial activity—all of which facilitate macroaggregate formation [26]. In contrast, anthropogenic disturbances in farmland disrupt aggregate structure, increase soil porosity, decrease macroaggregate abundance, and stimulate the formation of microaggregates (<0.25 mm) [27]. Specifically, tillage practices disrupt soil aggregates and accelerate organic matter decomposition, promoting the fragmentation of larger aggregates into smaller particles. Furthermore, WS aggregates in farmland and grassland exhibited a significantly lower proportion of >5 mm aggregates and a higher proportion of <0.25 mm aggregates compared with NWS aggregates. These findings indicate that macroaggregates in farmland and grassland are prone to dispersion into microaggregates under soil erosion conditions, whereas forestland aggregates demonstrate superior erosion resistance.

WS aggregate stability followed the order forestland > grassland > farmland, consistent with previous research demonstrating superior aggregate stability in forestland and grassland soils compared with farmland [28]. Furthermore, forestland exhibited significantly lower APD values for WS aggregates than grassland and farmland, indicating enhanced erosion resistance and reduced aggregate dispersion under rainfall events. In contrast, farmland soils are characterized by disrupted carbon inputs and the depletion of organic matter due to crop harvesting, suppressed microbial activity from intensive fertilizer application, and frequent tillage practices that fragment large aggregates and disrupt soil pore networks—collectively reducing aggregate cohesion and weakening both stability and erosion resistance [29]. Forestland exhibits greater aggregate stability than grassland, attributed to more extensive root systems and higher organic matter inputs. These factors stimulate microbial activity, promote the synthesis of binding agents, and strengthen aggregate cohesion [30]. Converting farmland to forestland or grassland enhances aggregate stability and structural homogeneity, reduces dispersion, and mitigates rainfall-induced erosion [31]. NWS aggregates in the 0.25–0.5 mm size fraction and WS aggregates in the 0.5–1 mm size fraction exhibited a greater SOC sequestration capacity. Across all land use types, both NWS and WS aggregates showed the highest SOC contribution from the 2–5 mm size fraction. Although the SOC content per unit mass of large aggregates was marginally lower than that of small aggregates, large aggregates dominated the total SOC pool owing to their higher mass fraction. These findings align with previous studies by Ayoubi et al. [32] and Bai et al. [33], which highlighted the pivotal role of large aggregates in SOC sequestration.

4.2. Effects of Freeze–Thaw on Soil Aggregate Stability and SOC Distribution

FTCs further influence the formation, fragmentation, and reorganization of soil aggregates by modifying soil structural properties, leading to changes in aggregate size distribution [34]. In this study, FTCs facilitated the aggregation of 2–5 mm NWS aggregates with smaller particles to form >5 mm aggregates. This phenomenon may be attributed to the fact that during FTCs, soil moisture freezes to form ice crystals, causing volume

expansion and generating radial extrusion pressure on surrounding aggregates; this forces 2–5 mm aggregates and adjacent smaller particles to bind tightly, forming larger temporary aggregates via physical adhesion [35]. Notably, after seven consecutive FTCs, the mass fraction of 2–5 mm WS aggregates in farmland increased by 87.82%, while that of <0.25 mm aggregates decreased by 41.82%. This trend was opposite to that observed in grassland, indicating that FTCs promoted the aggregation of small and medium-sized aggregates in farmland, thereby optimizing soil structure. Due to long-term tillage disturbances, the macroaggregates in farmland soil are disrupted, resulting in loosely arranged soil particles and a poor pore structure. During FTCs, the expansion and pressure from ice crystals force these dispersed soil particles and microaggregates to move closer together and reorganize into a denser, more compact structure [36]. In contrast, grassland soil characterized by dense root systems and high organic matter inputs forms an abundance of stable macroaggregates through root entanglement, rhizodeposits, and microbial products (e.g., fungal hyphae, polysaccharides), which act as effective binding agents. Although these organic cementing agents provide considerable stability, they are relatively vulnerable to the repeated tensile and shear stresses generated by ice crystals. FTCs may weaken or disrupt these critical biochemical bonds, leading to the disintegration of macroaggregates into smaller units.

FTCs exert biphasic effects—both promoting and inhibiting—on aggregate stability, with the direction and magnitude of these effects regulated by factors such as soil texture, initial aggregate size distribution, antecedent moisture content, organic matter content, freezing temperature, and FTC frequency [37,38]. Fu et al. [39] demonstrated that FTCs facilitate the disintegration of macroaggregates into microaggregates, thereby reducing aggregate stability. Similarly, Zhang et al. [40] observed that in sandy loam soils, the proportion of >1 mm aggregates decreased significantly with increasing FTC frequency, while the proportion of <0.5 mm aggregates increased progressively—resulting in a gradual decline in soil aggregate water stability. Conversely, Oztas et al. [41] identified a threshold at six FTCs: aggregate stability increased steadily up to six cycles, followed by a degradation phase thereafter. In the present study, NWS aggregate stability was enhanced across all land use types after FTCs, whereas WS aggregate stability increased in farmland but remained relatively unchanged in grassland and forestland. Notably, after FTCs, aggregate stability in forestland and grassland remained superior to that in farmland, indicating that FTCs did not eliminate the inherent differences in soil aggregate stability among land use types. The enhanced aggregate stability may be attributed to ice crystal formation during freezing: soil moisture freezing induces volume expansion, generating radial extrusion pressure on surrounding aggregates, which forces 2–5 mm aggregates to bond with adjacent microaggregates via physical adhesion, forming larger temporary aggregates [42]. Furthermore, ice crystal formation increases internal pore space within aggregates, induces the volumetric contraction of entrapped gases, and establishes a pressure equilibrium within aggregates or between adjacent agglomerates [43]—exerting negative pressure that inhibits physical disintegration.

FTCs induce alterations in soil physical, chemical, and microbial properties, which influence SOC distribution [44], facilitating the release of SOC and accelerating SOC mineralization rates [45]. Additionally, FTCs can eliminate 6–40% of soil microorganisms, which promotes the release of labile nutrients from lysed microbial cells, increasing dissolved organic carbon (DOC) concentrations [46,47]. The results showed that after FTCs, SOC content decreased across all aggregate size fractions and land use types, indicating that FTCs facilitate SOC release, with a more pronounced effect on NWS aggregates. This aligns with previous findings [48], which demonstrated that FTCs promote mutual transformation between aggregates of different size fractions, increasing the contact frequency between soil

organic matter and microorganisms [49]. This not only accelerates the decomposition and mineralization of soil organic matter but also enhances the release of various bioavailable nutrients from soil aggregates [50]. This apparent contradiction—increased aggregate stability alongside SOC loss after FTCs—can be attributed to the dual effect of FTCs. FTCs appear to break down weaker aggregates bound by transient organic matter (e.g., fungal hyphae and root exudates), thereby mineralizing and releasing their carbon. At the same time, FTCs promote the reorganization of soil particles into denser, more compact aggregates, where stability is derived less from organic binding and more from the physical interlocking of the particles themselves. Notably, FTCs exerted a more pronounced influence on the SOC contribution of WS aggregates, particularly in the 2–5 mm fraction, where contributions increased by 85.63% (farmland), 80.18% (grassland), and 86.32% (forestland). On the one hand, FTCs disrupt less stable >5 mm aggregates, causing them to fracture and reorganize into more densely structured 2–5 mm aggregates. On the other hand, 2–5 mm aggregates inherently possess ideal structural stability; they are capable of effectively encapsulating and physically protecting SOC while resisting disintegration induced by FTCs [51]. Furthermore, 2–5 mm aggregates are often rich in clay and silt particles, which facilitates the formation of organo-mineral complexes, thereby enhancing the chemical stability of carbon and mitigating SOC loss during FTCs.

FTCs attenuated the correlation between soil aggregate APD and SOC content across all size fractions. This observation indicates that FTCs disrupt the intrinsic relationship between soil structure and SOC, as repeated phase transitions induce cycles of the fragmentation and reorganization of aggregates, diminishing the specificity of carbon sequestration capacity in the original soil structure [52]. For MWD, GMD, and SOC content, correlations exhibited a threshold at the 0.5 mm size fraction: FTCs attenuated the positive correlation in the >0.5 mm fraction but strengthened it in the <0.5 mm fraction. Specifically, FTCs induced the fragmentation of >0.5 mm aggregates, impairing their physical protection of SOC and reducing carbon storage capacity, while facilitating <0.5 mm aggregates to become the primary carriers of secondary carbon pools. Additionally, FTCs selectively preserved mineral-bound carbon with higher stability in microaggregates and accelerated the loss of labile particulate organic carbon in macroaggregates [53]. These findings suggest that soil carbon pools may shift from macroaggregate-dominated physical protection to microaggregate-dominated chemical stabilization post FTC. Future research should incorporate analyses of carbon fractions (e.g., mineral-bound carbon and particulate organic carbon) to elucidate the effects of FTCs on SOC composition dynamics.

5. Conclusions

Forestland and grassland promoted the formation of macroaggregates with greater stability than those in farmland and exhibited higher SOC concentrations in these aggregates. FTCs modulated the homogeneity of WS aggregates, and WS aggregate stability in farmland was enhanced after FTCs. Additionally, FTCs induced a reduction in SOC content across aggregate size fractions. FTCs regulated the storage and distribution of SOC in WS aggregates, with the 2–5 mm size fraction serving as the primary carrier of SOC under FTC conditions. FTCs altered the correlation between soil aggregate stability and SOC content, suggesting a potential shift in soil carbon pools from macroaggregate-dominated physical protection to microaggregate-mediated chemical stabilization after FTCs. Converting farmland to forestland or grassland enhanced soil aggregate stability; to enhance soil carbon sink potential, SOC sequestration should be prioritized in 2–5 mm aggregates and the focus should be on preserving carbon sequestration in >0.5 mm aggregates during FTC periods. The findings of this study enhance our understanding of the freeze–thaw-driven mechanisms underlying soil carbon stabilization and aggregate dynamics, providing critical

insights for optimizing land management and soil conservation strategies in seasonally frozen regions.

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