

Article

Fractionation of Inorganic Phosphorus in Cold Temperate Forest Soils: Associating Mechanisms of Soil Aggregate Protection and Recovery Periods after Forest Fire Disturbance

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Abstract: The soil aggregate is the fundamental unit of soil structure. The fractionation characteristics and influencing factors of phosphorus (P) in soil aggregates inherently link its geochemical characteristics and recycling mechanism. This work investigated the fractionation characteristics of inorganic P in cold temperate forest soils and studied the impacts of recovery periods after forest fires and soil aggregate protection mechanisms on P fractionation. Our results showed that the TP, active P, stable P, and total organic carbon (TOC) contents varied with increasing recovery years after forest fire disturbance. The TP content in the coarse particulate organic matter fraction (cPOM) exhibited an increasing trend with the number of recovery years. Redundancy analysis (RDA) and correlation analysis indicated that TOC played a crucial role in influencing the dynamics of P fractionation during the recovery process. The order of TP levels in different soil aggregate fractions was as follows: μ Clay > dClay > LF > cPOM > dSilt > μ Silt > iPOM, with significant contributions from the cPOM and dSilt fractions. The ranking of P fractions in bulk soils was as follows: Aca-P > Fe-P > Oc-P > Or-P > De-P > Al-P > Ex-P. The protective mechanism of soil aggregates had a more significant effect on TOC than TP, with the order of protective abilities being: Phy \times biochem-protected > Biochem-protected > Phy-protected > Non-protected mechanism. TOC and recovery years emerged as critical factors influencing the dynamics of different P fractions during post-fire recovery. Soil aggregate protection mechanisms demonstrated significantly higher effects on TOC than on TP. This study provides insights into the fractionation mechanisms of P in the soil–forest ecosystem of the Greater Khingan Mountains, contributing to the sustainable development and utilization of cold temperate forest ecosystems.

Keywords: Greater Khingan Mountains; restoration years; soil aggregates; inorganic phosphorus fractionation; protective mechanism



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1. Introduction

The concentration, fractionation, and cycling processes of phosphorus (P) in soils directly impact its bioavailability, making them a crucial factor in determining the structure and functioning of ecosystems [1–3]. It has been indicated that the distribution of P forms in soils is a result of multiple interacting factors, including soil pH, clay content, organic carbon content, microbial activity, free ions of iron and aluminum, active calcium carbonate, and the surface hydrology conditions [4–8]. Although significant progress has been made in understanding the characteristics of soil particle composition [9,10], the ecological effects resulting from changes in soil particle composition [9,11,12], and the influence of vegetation types on soil particle composition [13,14], many studies have predominantly focused on bulk soils. Consequently, understanding the biogeochemical cycling mechanisms of soil

elements in soil particles during the pedogenesis process and their ecological effects in soil formation still presents several challenges.

In fact, soil consists of a mixture of particles of different sizes [9], existing in the form of mineral–organic matter complexes. Hence, a soil fractionation method was proposed based on the mechanisms of organic carbon protection and the size of aggregates in 2000, dividing soil into macroaggregates ($>250\ \mu\text{m}$), microaggregates ($53\text{--}250\ \mu\text{m}$), and easily dispersed silt-sized components ($<53\ \mu\text{m}$) [15,16]. Studies have shown a close correlation between the distribution of soil nutrients and the distribution of soil aggregates, with the horizontal and vertical differentiation of soil particle sizes being the main cause of variations in soil nutrient content [17]. The distribution of P fractions and P's bioavailability in soils is closely related to the distribution of soil aggregates [18–21], especially as smaller aggregates keep more P reserves, while larger aggregates exhibit a higher P supplying capacity [22]. In conclusion, the composition of soil aggregates is determined by factors such as soil parent material, vegetation type, and climate, while the composition of soil aggregates significantly influences the distribution characteristics of organic matter and nutrients in soil [23,24]. Therefore, aggregates serve as the fundamental units of soil structure and the fractionation, distribution characteristics, and influencing factors of P within soil aggregates inherently reflect the biogeochemical characteristics and cycling mechanisms of P.

Soil is the substantial foundation of forest ecosystems, and its physicochemical properties and the transformation of nutrients play a crucial role in the development and distribution of forests, thus sustaining and shaping the overall functioning of forest ecosystems [25,26]. Among numerous influencing factors, wildfires are ecologically significant drivers in northern forest ecosystems as they directly impact biological activities and alter the environmental conditions of the soil system [27–29], including the soil structure, organic matter content, microbial conditions, and geochemical cycling of nutrients, by modifying the soil temperature and oxidation conditions [30,31]. Consequently, wildfires have a profound influence on the structure and functionality of forest ecosystems. In recent years, extensive research has focused on the impact of various wildfire intensities and management strategies on forest ecological recovery after fire disturbances [32]. However, investigations on the recovery patterns of forest ecosystems, particularly soil systems, at different post-disturbance stages after wildfires have been relatively limited.

The geochemical fractionation of P within forest soil aggregates is a crucial research focus in the field of nutrient maintenance and sustainable utilization in forest ecosystems. The Greater Khingan Mountain region in China is the only cold temperate forest region, and its unique geographical location and climate conditions result in distinct characteristics of inorganic P distribution in the soil compared to forests in the southern region. Over the years, numerous scholars have focused on carbon and nitrogen in forest soils in the Greater Khingan Mountains [33,34]. However, P, as one of the essential nutrients for plant growth, has received relatively less attention, especially regarding the geochemical fractionation of inorganic P in soil aggregates. Therefore, this study focuses on the Greater Khingan Mountains, considering the spatial representation of the sites and the valuable soil samples from severely burned areas in 1968, 1987, 2009, and 2016. By utilizing combined physical and chemical fractionation methods coupled with a sequential extraction method for P fractions, we systematically study the fractionation characteristics of inorganic P in soil aggregates and its influencing factors.

Our objectives are to (1) determine the fractionation characteristics of inorganic P in cold temperate forest soils from the Greater Khingan Mountains, (2) explore the mechanisms of soil aggregate protection and the impact of recovery periods after wildfires on the fractionation of inorganic P within bulk soils and soil aggregates, and (3) understand the migration and transformation patterns of P in forest soil after wildfire disturbances. This work provides insights into the biogeochemical cycling mechanisms of P in cold temperate forest soils and contributes to our understanding of the ecological recovery and functional evolution processes of forest ecosystems.

2. Materials and Methods

2.1. Study Area

The forest region of the Greater Khingan Mountains (50°10′–53°33′ N, 121°12′–127°00′ E) is located in the northeastern area of the Inner Mongolia Autonomous Region and north of Heilongjiang Province. It stretches approximately 1200 km in a northeast–southwest direction, with a width of about 200–300 km; the elevation ranges from 1100 to 1400 m [35]. This region is characterized by a cold temperate continental monsoon climate, with an annual average temperature ranging from −6 to 1 °C. The average temperatures in winter and summer are approximately −28 °C and 18 °C, respectively, with the lowest temperature dropping to −52 °C. The annual precipitation ranges from 240 to 442 mm, predominantly occurring in July and August, and the snow cover can last for up to 6 months during winter [36]. The major soil types in this forest area include dark brown soil, gray forest soil, and brown coniferous forest soil. Dark brown soil is primarily distributed in the southern and eastern hilly regions of the area, while brown coniferous forest soil is mainly located in the northern and central parts of the Greater Khingan Range. Gray forest soil mainly occurs in the western area close to the grassland [37]. The dominant vegetation type in this region is cold temperate coniferous forest, with the dominant species being *Larix gmelinii*. Other significant tree species include *Pinus sylvestris*, *Betula platyphylla*, *Quercus mongolica*, and so on [38].

2.2. Soil Sample Collection

In this work, a total of 30 sampling sites were established, as illustrated in Figure 1. The soil samples were collected at two layers, namely 0–10 cm and 10–20 cm depths, at each site. In this study, we adopted a space-for-time substitution approach to investigate the effects of different recovery periods after fire on P fractions in soils. In the study area, we identified four sample areas representing recovery periods of 1, 8, 30, and 49 years after fire. The collected soils were carefully placed into self-sealing bags and transported to the laboratory. The samples were naturally air-dried and subsequently processed by removing impurities such as plant roots and gravel using a 2 mm sieve for further analysis and experiments.

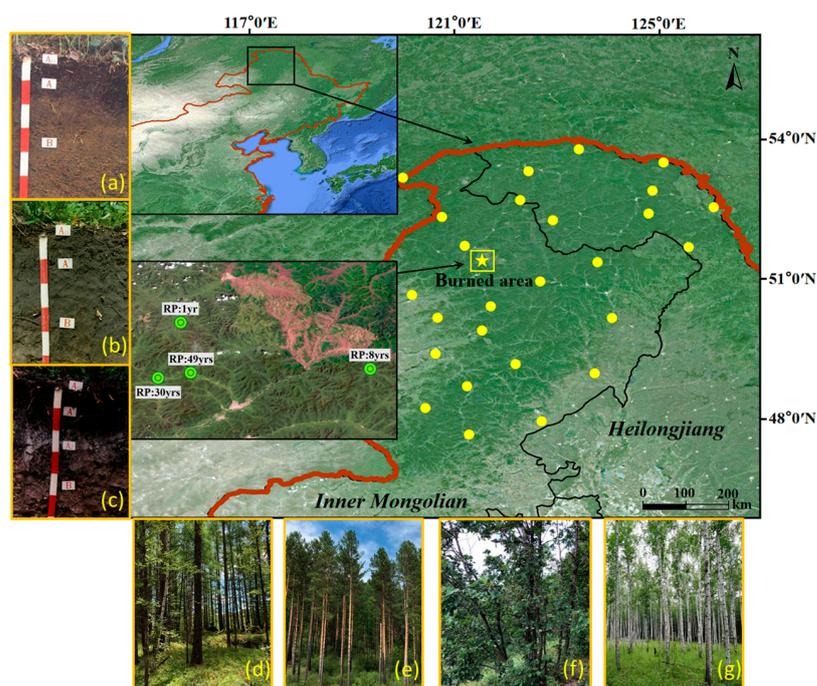


Figure 1. Locations of sampling sites. (a) Dark brown soil; (b) gray forest soil; (c) brown coniferous forest soil; (d) *Larix gmelinii* forest; (e) *Pinus sylvestris* forest; (f) *Quercus mongolica* forest; and (g) *Betula platyphylla* forest.

2.3. Soil Grouping Method and P Fractionation

In this study, we adopted the combined physical and chemical fractionation technique proposed by Stewart et al. [39]. The procedure involved the following steps: Firstly, the soil samples were dispersed and physically separated into four fractions: coarse particulate organic matter without protection (cPOM, 250–2000 μm), microaggregates (μagg , 53–250 μm), and dispersible silt and clay (dSilt and dClay, <53 μm). Secondly, the microaggregates (μagg) were further classified by employing density flotation, dispersion, flocculation centrifugation, as well as other processes. This facilitated the isolation of fine particles of organic matter without protection (light fraction, LF), organic matter protected in microaggregates (>53 μm , iPOM), and silt and clay protected in microaggregates (μSilt , μClay). Thirdly, the separated fractions were air-dried at 60 °C and subsequently weighed. Lastly, the total organic C (TOC) content was determined using a TOC/total N analyzer (Multi N/C 3100, Jena, Germany). Meanwhile, we applied the sequential extraction procedure to investigate the fractions of P in the bulk soils and their aggregates [40–43], in which the soil P were extracted as exchangeable or loosely sorbed P (Ex-P), Al-bound P (Al-P), Fe-bound P (Fe-P), authigenic apatite plus CaCO_3 -bound P plus bigenic apatite (Aca-P), detrital apatite plus other inorganic P (De-P), and organic P (Or-P). The content of P in the extract was determined by a colorimetric method (UV-2550, Shimadzu, Japan). Soil pH was determined in a 5:1 water:soil ratio using a digital pH meter (Mettler Toledo, China).

3. Results

3.1. P Fractionation and Influencing Factors in Bulk Soils

3.1.1. TP and P Fractionation

The results revealed a significant increasing trend for TP in bulk soils along with the recovery periods. Similar patterns were also observed in the soil layers at depths of 0–10 cm and 10–20 cm (Figure S1, Supplementary Material). At a recovery period of 1 year after the disturbance, the average concentration of TP in the 0–10 cm soil layer was measured as 677.73 mg/kg, while at 49 years of recovery it increased to 981.47 mg/kg. For the 10–20 cm soil layer, the average TP content was 424.17 mg/kg at 1 year of recovery and significantly increased to 799.66 mg/kg at 49 years of recovery.

The distribution of various P fractions in the bulk soils is presented in Table S1 (Supplementary Materials). The concentration ranges of Ex-P, Al-P, and Fe-P were 0.99–3.82, 1.92–11.43, and 74.10–371.71 mg/kg, with average concentrations of 2.78, 5.76, and 201.25 mg/kg. The Fe-P concentrations accounted for approximately 29.33% of the TP. The concentrations of Oc-P, Aca-P, De-P, and Or-P were 87.90–332.21, 152.95–213.17, 18.16–100.10, and 32.09–94.61 mg/kg, with averages of 207.63, 179.06, 55.11, and 63.74 mg/kg, respectively. In total, these stable fractions accounted for approximately 70.67% of the TP. Overall, Oc-P, Fe-P, and Aca-P exhibited the highest levels among the different fractions, accounting for approximately 29.03%, 28.13%, and 25.03% of the TP in bulk soils, respectively.

3.1.2. Influencing Factors

To investigate the key factors influencing the dynamics of P fractionation during the recovery process following forest fire disturbances, we conducted a correlation analysis of P fractions and TP contents with environmental factors such as soil pH, TOC, soil texture, and recovery years (RY) (Figure 2). The results revealed that the active P fractions, Ex-P, Al-P, and Fe-P, were positively correlated with TOC. Fe-P and Oc-P showed significant positive correlation with RY. De-P exhibited significant positive correlations with $R_{0.053-0.25}$ (aggregates with size of 0.053–0.25 μm) and $R_{<0.053}$ (aggregates with size < 53 μm), while showing a significant negative correlation with $R_{>0.25}$ (aggregates with size > 0.25 μm). Or-P exhibited a significant positive correlation with TOC, while TP showed significant positive correlations with TOC and RY. Furthermore, the correlations between various P fractions revealed a significant positive correlation between Al-P and Or-P. Fe-P exhibited significant positive correlations with Oc-P, while showing a significant negative correlation

with De-P. Fe-P and Oc-P both showed significant positive correlations with TP. Overall, these findings indicate a close association between the dynamics of P fractions and TOC as well as RY.

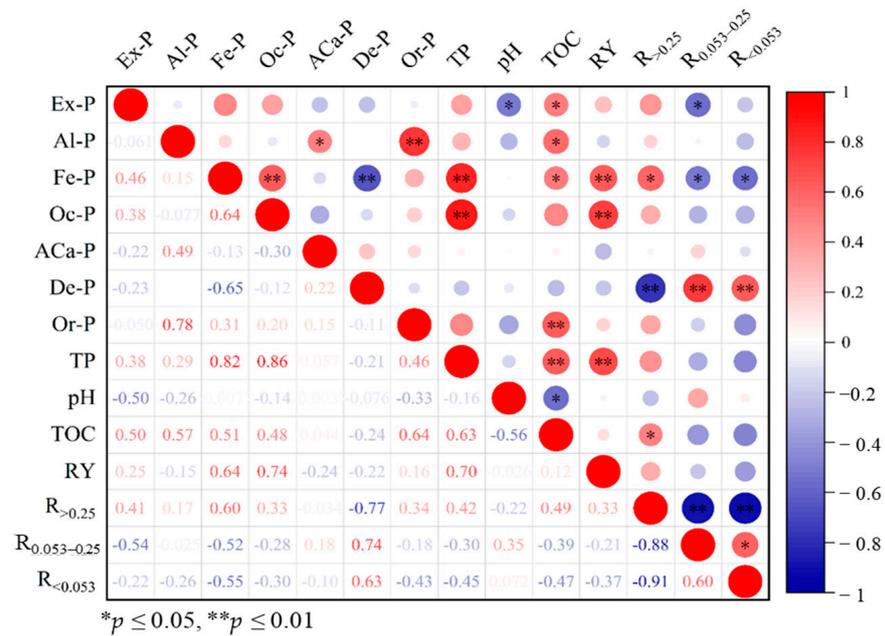


Figure 2. Correlations between P fractions and environment factors.

In this study, redundancy analysis (RDA) was conducted using P fractions and TP contents as response variables and soil TOC, pH, R_{<0.053}, R_{0.053-0.25}, R_{>0.25}, and RY as explanatory variables (Figure 3). The RDA results revealed that the first axis (RDA1) explained 52.2% of the variation in P fractions and 89.9% of the correlation between P fractions and environmental factors. The second axis (RDA2) accounted for 4.6% of the variation in P fractions. According to Monte Carlo permutation tests, it was found that TOC, pH, R_{<0.053}, R_{>0.25}, and RY collectively explained 61% of the variation in P fractionations.

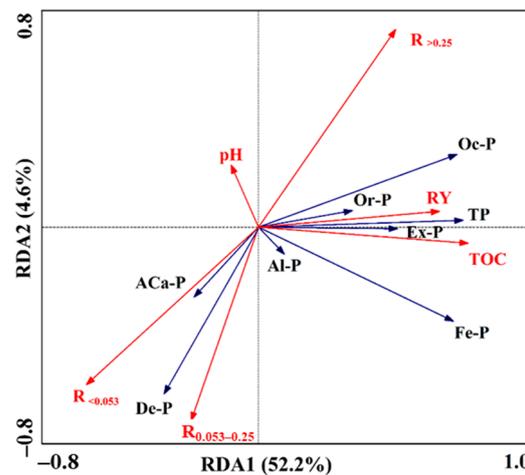


Figure 3. Redundancy analysis (RDA) of P fractions during different restoration periods.

3.2. P Fractionation in Soil Aggregates

The distribution of TP in various soil aggregates for different recovery periods was presented in Table S2 (Supplementary Materials). The aggregates of μ Clay, dClay, and LF showed the highest TP levels, with averages of 2164.94, 1584.27, and 860.14 mg/kg, respectively. On the other hand, dSilt, iPOM, and μ Silt exhibited the lowest TP contents, with averages of 679.13, 509.98, and 414.87 mg/kg, respectively.

The P fractionations in various soil aggregates for different recovery periods was illustrated in Figure 4. The fractions of ACa-P, Fe-P, and Oc-P were found to be the most abundant in all soil aggregates after forest fire disturbance, with average concentrations of 277.51, 247.93, and 241.01 mg/kg, respectively. These fractionations accounted for 30.52%, 27.27%, and 26.51% of the TP, making them the dominant fractionations in the different soil aggregates.

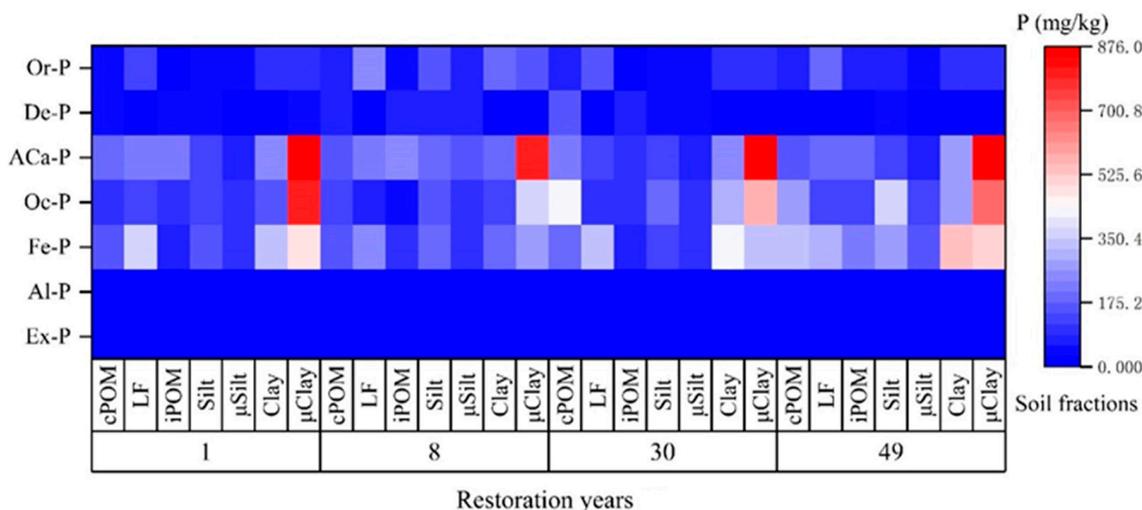


Figure 4. Distribution of P fractions in soil aggregates.

4. Discussion

4.1. P Fractionation Characteristics in Bulk Soils

4.1.1. The Trend for TP during Different Restoration Periods

The TP patterns in soils suggested that the severe fire disturbance resulted in the depletion of aboveground vegetation and litter, leading to the death of microorganisms and roots [44], which broke the recycling of P in the forest ecosystem. Additionally, the fire destroyed the forest understory, disrupting the stability of the soil structure and diminishing the protective role of vegetation, consequently accelerating the P loss. Therefore, in the early stage of post-fire recovery (1 year), the concentration of TP in the regenerating soils was relatively low. As secondary succession progressed following the fire disturbance, soil conditions favored an increase in species diversity, forest density and soil biomass, and vegetation flourish [45]. Consequently, there was an increase in the quantity of plant and animal residues entering the soil, contributing to the visible upward trend in soil fertility. As a result of this process, the TP content in the soil gradually increased. Furthermore, as vegetation gradually recovered, root systems and microorganisms continued to grow, enhancing the stability of the soil structure. The increasing forest canopy closure, along with reduced sunlight penetration in the understory, provided effective protection for the soil [46]. These factors drastically reduced the loss of P and other soil nutrients. Moreover, the improved canopy closure reduced light intensity in the understory that, coupled with increased plant transpiration, resulted in improved water and heat conditions [46]. This, in turn, increased microbial abundance and activity, promoting the decomposition of litter and facilitating the accumulation of P in the soils. As a result, with prolonged recovery periods, the TP content in the soils continued to increase.

4.1.2. The Characteristics of P Fractionation

The ranking order of P fractions was as follows: Oc-P > Fe-P > ACa-P > Or-P > De-P > Al-P > Ex-P (Table S1). Among these fractions, Ex-P, Al-P, and Fe-P represented the active P fractions that were readily available for plant uptake. Fe-P exhibited the highest level among the active fractions, indicating its dominance as the main active P fraction. On the other hand, Oc-P, ACa-P, De-P, and Or-P exhibited relatively stable properties, making it

challenging for plants to directly uptake and utilize them. Fe-P refers to P that is bound to iron oxides and hydroxides through surface bonds, while Oc-P mainly refers to P enclosed within iron and aluminum oxide films. The relatively high levels of these fractions may be attributed to the higher concentrations of iron oxides in soils from the studied area. These findings highlighted the significant role of Oc-P, Fe-P, and ACa-P, shedding light on the P dynamics and availability in the study area. The spatial patterns of various P fractions in bulk soils are shown in Figure S2 (Supplementary Materials).

The changes in different fractions of P in soils after various numbers of restoration years are presented in Figure 5. Overall, both the active fractions of P (Active-P) and stable fractions of P (Stable-P) gradually varied with the increase in restoration periods. This can be attributed to the gradual recovery of vegetation, resulting in increased forest density and a significant increase in litter input, thereby providing more sources of P. Additionally, with the increasing restoration age after the forest fire, soil microbial biomass and related enzyme activities significantly increased, enhancing the decomposition of litter and the accumulation of soil P. As the dominant fractions of P in the study area, Fe-P and Oc-P exhibited similar changing trends, i.e., their concentrations increased with the restoration age. A previous study demonstrated that coniferous trees preferentially absorb $\text{NH}_4^+\text{-N}$ from the soil, leading to the generation of equivalent amounts of H^+ and a decrease in soil pH under coniferous forests [47]. As the forest ecosystem gradually recovers, *Larix gmelinii* becomes the dominant tree species, resulting in a noticeable decrease in soil pH. As the pH decreases, the concentration of H_2PO_4^- in the soil solution continuously increases. Compared to the HPO_4^{2-} species, the hydroxyl groups on the surface of iron oxides are more likely to undergo ligand exchange reactions with H_2PO_4^- [48]. Therefore, as the pH decreases, the adsorption capacity of iron oxides for P gradually increases. As a result, with the increase in restoration age, the ability of iron oxides to adsorb inorganic P strengthens, in turn leading to a significant increase in Fe-P and Oc-P levels. In addition, the concentrations of ACa-P and De-P exhibited a significant decrease at the restoration times of 30a and 49a, which may be due to the relatively low soil pH during these two periods, enhancing the dissolution of calcium-bound P.

The RDA results showed that TOC showed the most significant effect on the variation in P fractionations, explaining 32% of the variance (Figure 3). This suggests that TOC plays a crucial role in shaping the distribution and fractionations of P in bulk soils from the study area. RY and pH followed TOC, explaining 14% and 7% of the variance, respectively. The importance ranking of the environmental factors influencing the variation in P fractionations was as follows: $\text{TOC} > \text{RY} > \text{pH} > R_{>0.25} > R_{<0.053} > R_{0.053-0.25}$.

These findings highlight the significant role of TOC in regulating the dynamics of P fractionations in the soil and the importance of considering RY and pH as additional influential factors during the recovery process following forest fire disturbance. The restoration process of forests influences soil, plant, and microbial activities, resulting in changes in the distribution characteristics of TOC and consequently affecting the distribution of P fractionations. During the recovery process after fire disturbance, forest density, soil biomass, and species diversity continuously increase [49]. This leads to an increase in the quantity of plant and animal residues, as well as litterfall entering the soil, resulting in significantly elevated TOC levels. Organic matter plays a significant role as a source of soil P, releasing inorganic P during the mineralization process [2,50,51]. Moreover, the mineralization decomposition of organic matter generates organic acids that promote the transformation of stable inorganic P into active P. Additionally, the presence of the fulvic acid polyanions in organic matter can enter mineral particles through selective adsorption, competing with phosphate anions and thereby facilitating the release of inorganic P [52] and increasing the activity of soil P. Therefore, the restoration periods are significantly positively correlated with both TP and various fractions of active P, with TOC playing a crucial role in this process.

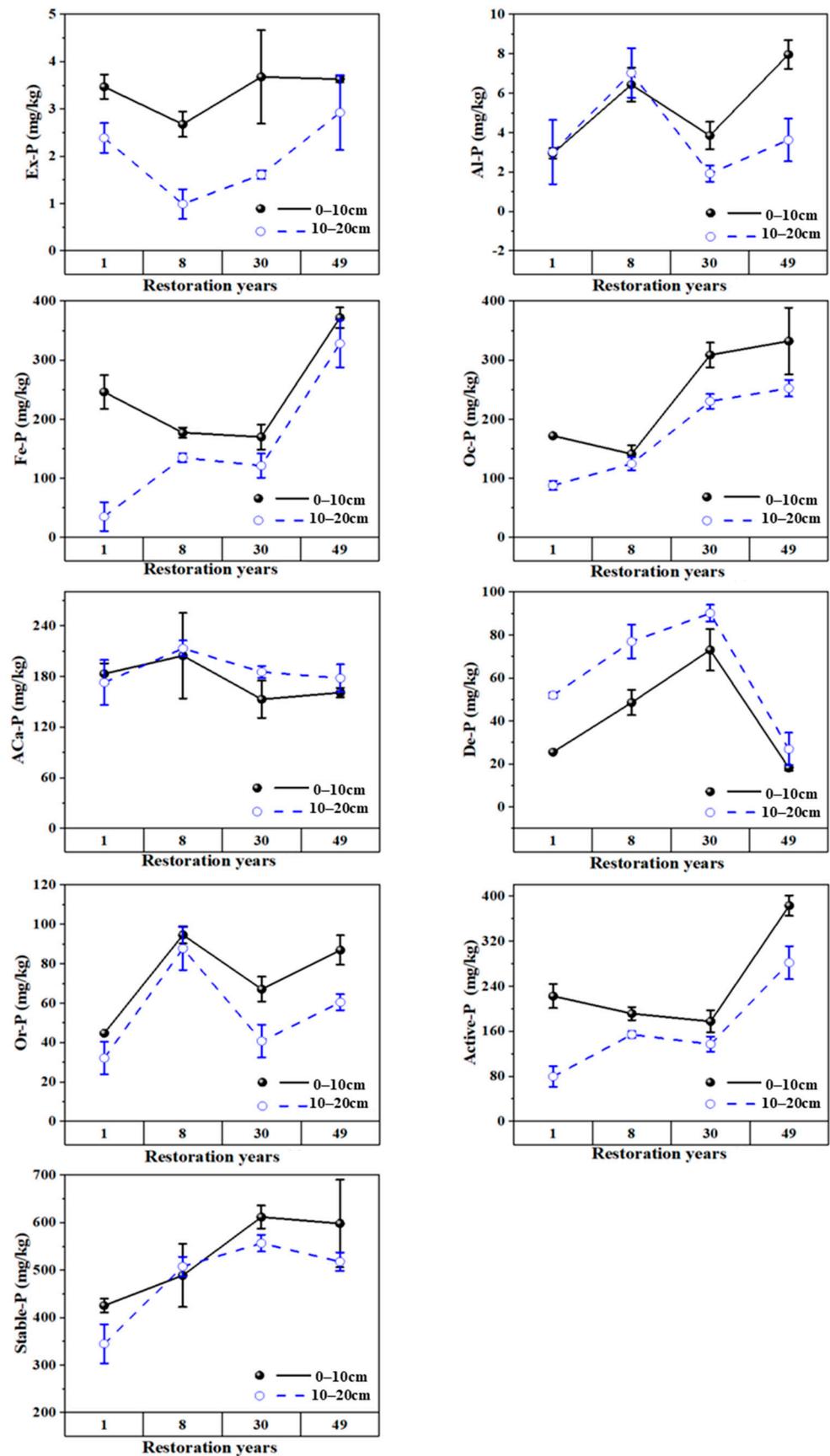


Figure 5. P fractions in bulk soils during different restoration periods.

4.2. P Fractionation Characteristics in Soil Aggregates

4.2.1. TP Characteristics

The distribution trend of TP in each soil layer was as follows: $\mu\text{Clay} > \text{dClay} > \text{LF} > \text{cPOM} > \text{dSilt} > \text{iPOM} > \mu\text{Silt}$ (Table S2). The aggregates of dClay and μClay consist of clay-sized particles that have small particle sizes and larger specific surface areas, resulting in a strong adsorption capacity for P. As a result, P tends to accumulate in these two soil aggregates. The LF aggregate represents unprotected fine particles, mainly consisting of plant roots, residues, charcoal, and microbial structures. Therefore, this aggregate exhibited higher organic matter and P levels. Both dSilt and μSilt are composed of silt-sized particles that are characterized by a weaker adsorption capacity and, consequently, lower TP contents. The spatial patterns of TP in soil aggregates are shown in Figure S3 (Supplementary Materials).

As can be seen from Figure 6, with an increase in the recovery period, only the TP content of the cPOM aggregate exhibited a significant increase. The cPOM aggregate, which represents the largest proportion of soils, mainly includes decomposed plant and animal residues as well as microbial remains, in addition to the sandy fraction. It is considered as the non-protected fraction of soil, susceptible to external disturbances such as land-use practices and soil management influences [53]. Hence, as the recovery period lengthens and vegetation gradually recovers, the TP content in the cPOM aggregate showed a corresponding increasing trend. For the LF aggregate, the TP content in the 0–10 cm soils was significantly higher than that in the 10–20 cm soils, demonstrating a clear surface accumulation pattern (Figure 6). The LF aggregate consists of plant residues at different stages of decomposition and some microbial structures. The surface soil layer (0–10 cm) tends to accumulate more litter, fine roots, and other external plant debris [54], suggesting a greater input of organic matter and P compared to the lower soil layer (10–20 cm).

4.2.2. P fractionation Characteristics

Generally, Fe-P, being an active and soluble phosphate, is easily dissolved in the soils due to factors such as pH, redox potential, and biological processes. Its relatively high levels suggested that the soils in the study area had a high P supply potential. The environmental conditions in the Greater Khingan Mountains are relatively humid and mobile elements are leached out as a result of the weathering and decomposition of silicate minerals. Consequently, elements like iron and aluminum, which are characterized by relatively stable chemical properties, tend to accumulate [55]. Therefore, the high content of iron oxides in the soil was beneficial for the formation of Fe-P. As mentioned above, the pH of forest soils in the study area was very low (4.5–6.5), which induces phosphate ions to be adsorbed by free iron oxides in the soils. Additionally, the acidic conditions promote the conversion of stable P fractions to Fe-P [56,57]. Hence, the fraction of Fe-P was found to be one of the dominant fractionations in forest soils from the Greater Khingan Mountains. As for various soil aggregates, the maximum of each P fraction for different recovery periods was observed in the dClay and μClay aggregates, with $\mu\text{Clay} > \text{dClay}$. This aligned with the higher TP content observed in these two aggregates. The smaller particle size and stronger adsorption capacity of clay fractions, along with their protective effect on microaggregates, contribute to this phenomenon. Consistent with the investigations of bulk soils, an increasing trend in P fractionations for each soil aggregates was also observed with the increasing recovery period. This suggested that the forest recovery process also involved a gradual accumulation of soil P.

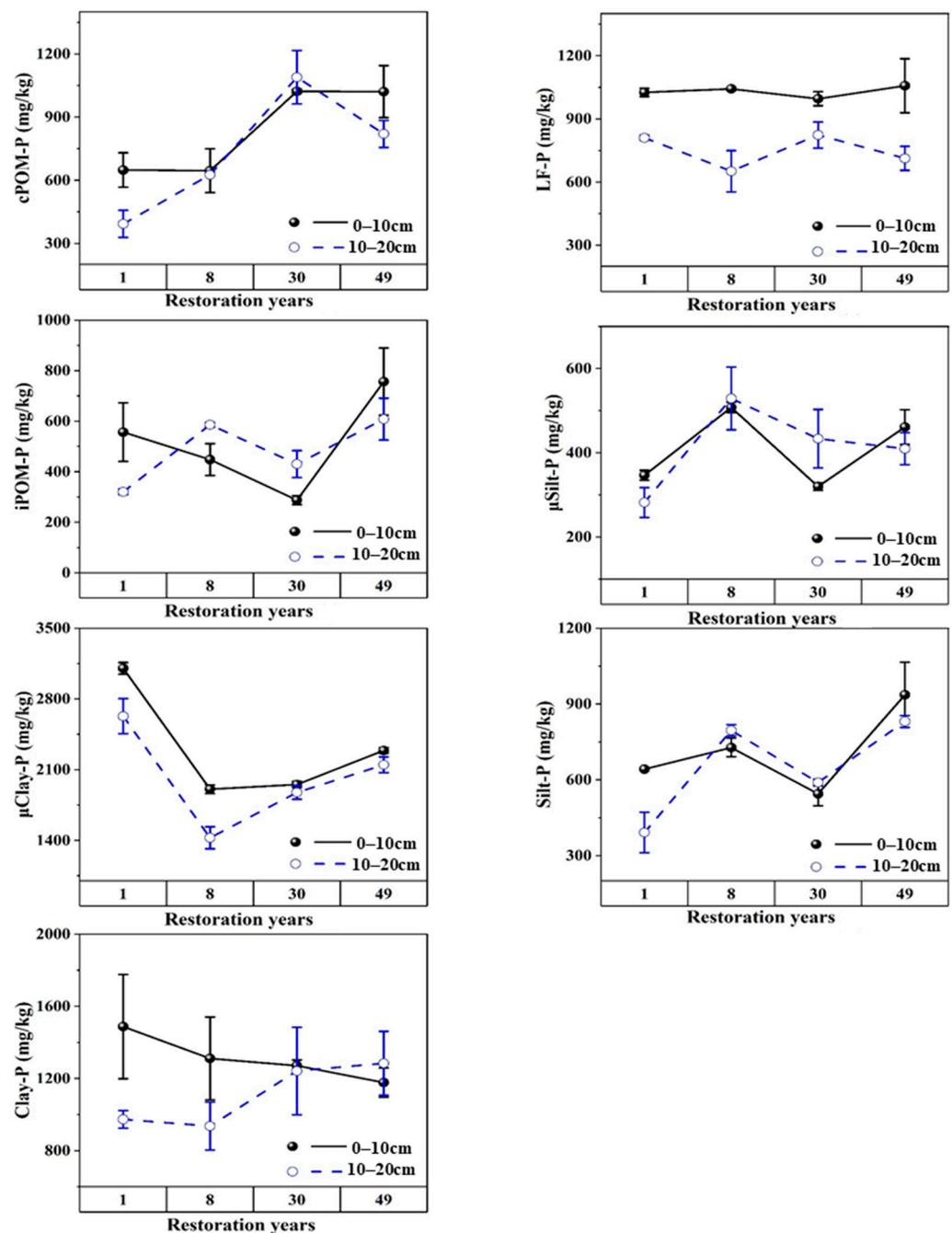


Figure 6. Changes in TP in soil aggregates with different restoration years.

4.3. Soil Aggregate Protection Mechanisms

4.3.1. Characteristics of C and P under Various Protection Mechanisms

In order to explore the effects of aggregate protection mechanisms on the migration and transformation of C and P in soils, this study applied the soil grouping method proposed by Stewart et al. [39] to divide the soil into four protective mechanism fractions: the unprotected fraction (cPOM + LF), physically protected fraction (μ agg), biochemically protected fraction (dSilt + dClay), and physically–biochemically coupled protected fraction (μ Silt + μ Clay). Figure 7 shows that the levels of TOC and TP were highest in the unprotected fraction and relatively lower in the physically protected and physically–biochemically coupled protected fractions. Generally, the unprotected fraction consists of soil macroaggregates ($>250 \mu\text{m}$) and free light fractions, with the macroaggregates being weakly physically protected, resulting in rapid turnover of internal organic matter and

rapid decomposition of organic binding agents. Therefore, macroaggregates are unstable and prone to disintegration [58]. On the other hand, the free light fraction contains C and P that are weakly bound to soil particles, making them unstable and fast in turnover. Consequently, the unprotected fraction is susceptible to anthropogenic disturbances and external inputs of quantity and quality and the processes of organic matter and nutrient consumption and storage within the fraction occur rapidly. Typically, the residues of animals, plants, and microorganisms in soils are first retained by macroaggregates and the free light fraction after decomposition, leading to C and nutrient enrichment [59,60]. Moreover, since this fraction accounted for a large proportion in the soils, the concentrations of TOC and TP were significantly higher than for other aggregate fractions. Subsequently, organic matter and nutrients undergo a series of geochemical processes, finally becoming fixed in the biochemically protected fraction by binding with soil minerals (dSilt + dClay). Some parts of biochemically protected fraction and particulate organic matter can be encased within microaggregates, receiving physical protection from the microaggregates and becoming the physically protected fraction, while the parts of the biochemically protected fraction that enter aggregates are subjected to physically–biochemically coupled protection. As a result, the organic matter and nutrients that ultimately enter microaggregates were relatively low in content within the physically protected and physically–biochemically coupled protected aggregates.

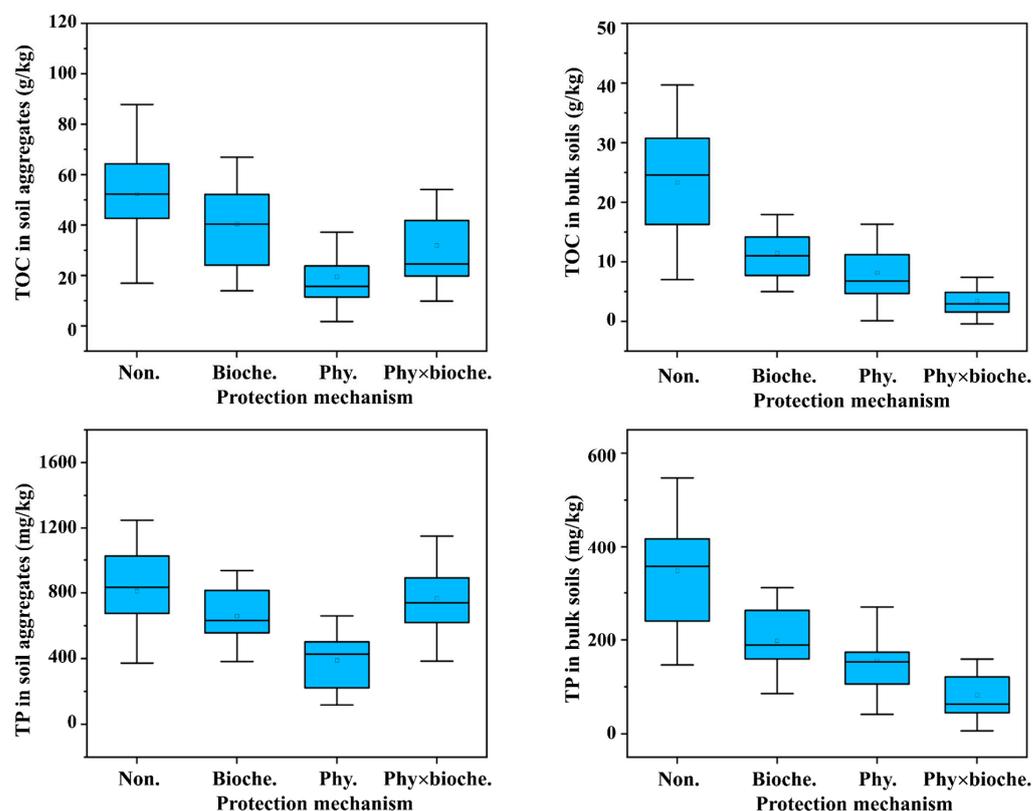


Figure 7. TOC and TP in different protection mechanisms for soil aggregates.

4.3.2. Association between C and P under Various Protection Mechanisms

The geochemical cycling of C and P in soil is influenced by various factors, with soil aggregates playing a key role as protective mechanisms. Soil aggregates are formed under the co-effects of physical, chemical, and biological processes, facilitating the sequestration and transformation of C, P, and other elements. These processes lead to their storage in different protective mechanisms. Accordingly, the response of different elements to the protective mechanisms of aggregates varies. Our findings revealed significant positive correlations between C and P in all protective mechanisms (Figure 8). Specifically, the

concentrations of TP increased with TOC contents, with different protective mechanisms displaying varying slopes: $\text{Phy} \times \text{biochem-protected} > \text{Biochem-protected} > \text{Phy-protected} > \text{Non-protected}$ (Figure 8). Moreover, approximately 90% of soil C is preserved within aggregates [61], indicating the substantial influence of aggregate protective mechanisms on TOC. In contrast, P, being a depositional element with slower participation in biogeochemical cycling, exhibits a lesser response to aggregate protection. The unprotected fraction, lacking stability in structure, exhibits relatively lower affinity for the binding of organic matter and nutrients to soil particles, leading to weaker degrees of protection and faster turnover rates. Consequently, during changes in the external environmental conditions, both TOC and TP contents showed higher variability compared to other aggregate fractions, resulting in smaller slopes in the fitting curves. The physical protective fraction provides a physical barrier in the form of microaggregates, which protects TOC from microbial decomposition and mineralization processes [62,63]. Additionally, the anaerobic conditions within these microaggregates further inhibit microbial activity. As a result, TOC within the physical protective fraction experienced better preservation, displaying lower susceptibility to environmental changes compared to TP, as illustrated by the higher slope in the fitting curve of this fraction than that of the unprotected fraction. The biochemical protective fraction involves the adsorption of organic matter by soil minerals and free iron–aluminum colloids through mechanisms such as ligand exchange and multivalent cation bridging [64]. The ligand exchange process involves the formation of covalent bonds between organic functional groups and the active inorganic carboxyl groups on the mineral surface, resulting in a relatively stable association [64]. Therefore, the TOC within this fraction enjoyed higher stability and protection compared to the unprotected and physical protective fractions. However, the protective effect on P was observed as being relatively consistent. Accordingly, the fitting curve slope of this fraction was higher than that of the previous two fractions (Figure 8). Finally, the $\text{Phy} \times \text{biochem-protected}$ fraction represents a scenario where organic carbon binds to soil minerals before being enclosed within microaggregates. This fraction benefits from a combination of physical and biochemical protective mechanisms, resulting in the highest levels of protection and stability for TOC. Consequently, this fraction exhibits the least susceptibility to environmental changes. However, no significant difference in the protective effect on P were observed in this fraction compared to the other fractions, resulting in the largest slope in the fitting curve among all aggregate fractions.

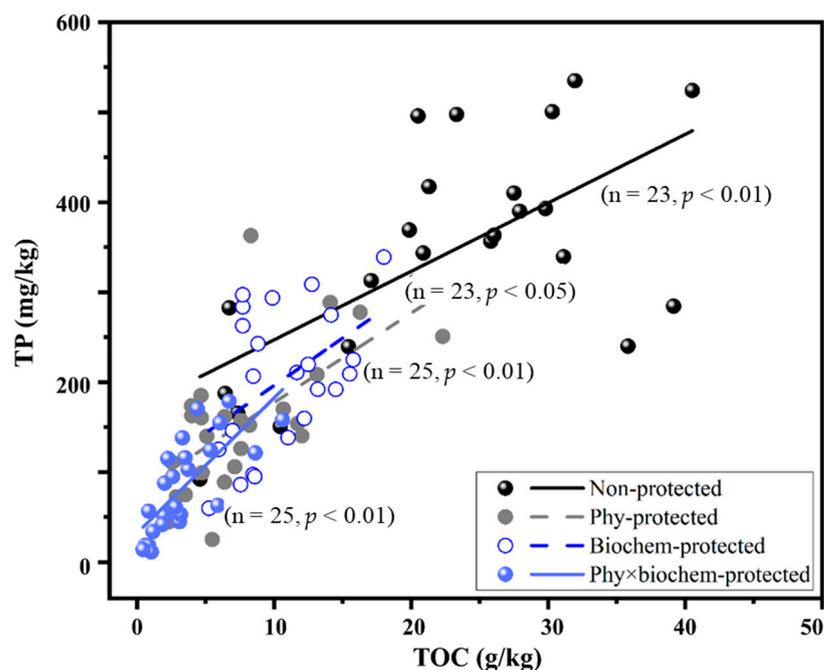


Figure 8. Difference of soil aggregate protection mechanisms for TOC and TP.

5. Conclusions

The geochemical fractionation of P in forest soil aggregates is an important research focus in the fields of nutrient maintenance mechanism and the sustainable utilization of forest ecosystems. Considering the spatial representation of the sites and the valuable soil samples from severely burned areas in 1968, 1987, 2009, and 2016, this work employed combined physical and chemical fractionation methods and a sequential extraction procedure to systematically investigate the fractionation mechanism of inorganic P in bulk soils and their aggregates in the Greater Khingan Mountains. Our findings revealed the sequence of various P fractions in soils as follows: A_{Ca}-P > Fe-P > Oc-P > Or-P > De-P > Al-P > Ex-P. Among the various soil aggregates, the levels of TP were found to be highest in the aggregate fraction of μ Clay, followed by dClay, LF, cPOM, dSilt, μ Silt, and iPOM, with cPOM and dSilt making significant contributions to soil TP. The TP content in the cPOM fractions showed an increasing trend with longer recovery periods. RDA and correlation analysis indicated that TOC and RY explained 32% and 14% of the variations in P fractionation, respectively, emerging as critical factors influencing the dynamics of different P fractions during post-fire recovery. The relative importance of environmental factors in affecting the variations in P fractionation was ranked as follows: TOC > RY > pH > $R_{>0.25}$ > $R_{<0.053}$ > $R_{0.053-0.25}$. Soil aggregate protection mechanisms demonstrated significantly higher effects on TOC than on TP, with the ranking of protective abilities as follows: Phy \times biochem-protected > Biochem-protected > Phy-protected > Non-protected. The findings of this study hold important implications for understanding the processes of ecosystem recovery and functional evolution and furthering our understanding of the biogeochemical cycling mechanisms of phosphorus in cold temperate forest soils.

Supplementary Materials: The following supporting information can be downloaded at: <https://www.mdpi.com/article/10.3390/f15050875/s1>. Figure S1. TP variation in bulk soils during different restoration periods; Figure S2. Spatial patterns of various P fractions in bulk soils; Figure S3. Spatial patterns of TP in soil aggregates; Table S1. P fractions in bulk soils during different restoration years; Table S2. Distribution of TP in soil aggregates during different restoration years.

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