

The effect of litter addition on soil microbial and enzyme indices after forest harvesting operations in Hyrcanian deciduous forests, twenty years trajectory

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Abstract

Improvement of soil characteristics in natural conditions after ground-based forest harvesting operations is necessary to maintain soil quality. Therefore, the present study was conducted to examine the effect of different tree litters on soil microbial properties and enzyme activity of skid trails over a 20-year period after skidding operations. A total of 216 soil samples were taken from three litter treatments (B; beech, B-H; beech-hornbeam, B-H-O; mixed beech) exposed to three traffic intensity classes (low, medium, and high) that were each assigned to three time periods (6, 10 and 20 years after harvesting). All combinations were replicated three times. The highest value of soil microbial characteristics and enzyme activity belonged to B-H-O treatment followed by > B-H > B and 20-years since harvest followed by > 10 years > 6 years. Recovery of microbial properties and enzyme activities under the influence of high-quality litter (mixed beech) was positively associated with other soil properties including pH, total N, available nutrients. Values of soil microbial properties including soil microbial respiration (15%), microbial biomass carbon (8.7 %), microbial biomass nitrogen (15.4%), NH_4^+ (13.6%), NO_3^- (9.8 %) and enzyme activities such as urease (10.2%), acid phosphatase (4.4%), arylsulfatase (8.8%), invertase (6.9%) in the B-H-O treatment measured 20 years after harvesting were less than the values of the undisturbed area. According to these results, it seems that the mixed beech litter treatment has been able to improve soil properties more than the other tested treatments.

Introduction

Forests are one of the most important ecosystems on earth that not only provide essential services such as climate control, habitat protection, erosion prevention, nutrient cycle, and biodiversity protection, but also maintain an important economic resource for many countries (Hume et al. 2018; Picchio et al. 2021). Based on the sustainable management of forests, there is an increasing demand for forest products, resulting in high environmental pressures towards forests (Acharya et al. 2019; Labelle et al. 2022).

Sustainable management is essential for improved protection and development of forests. In this regard, understanding soil properties is one of the principles of basic forest management (Chen et al. 2005). Ground-based mechanized operations performed by skidding machines on skid trails can create severe damage to soils and decline forest productivity (Jourgholami et al. 2018; DeArmond et al. 2020). Soil compaction and degradation following skidding operations by heavy machinery can, beyond changes to soil physical properties (Han et al. 2006), lead to significant changes in soil chemical and biological properties such as less gas exchange, reduced water penetration, reduced root and tree growth, reduced decomposition rates, alteration and disruption of the food cycle, resulting in an adverse effect on soil microbes (Crawford et al. 2021).

The use of organic and inorganic mulch (i.e., foliage, sawdust, and protective mats) and limiting the number of machine passes on skid trails as remedial treatments can mitigate soil disturbance in the short term (Han et al. 2009). However, under natural conditions, it is important to consider the type of forest stand and litter as an ecological solution to reduce the negative effects and accelerate the recovery time of soil properties after skidding operations (Jourgholami et al. 2019). In addition to the protective role of the soil surface (Li et al. 2014), forest floor litter contributes to the nutrient flow and carbon cycle (Kooch et al. 2018). In turn, high-quality litter plays a key role in stimulating soil organic carbon decomposition and the activity of soil microorganisms and microorganisms (Fang et al. 2011; Jourgholami et al. 2018).

In forest ecosystems, soil microbes govern the decomposition of organic matter, the food cycle, and the availability of plant-absorbable nutrients (Plante 2007). In the soil ambiance, the availability of usable carbon substrate is the most important factor limiting microbial activity, which results in an increase of the microbial population around the substrate once litter has been added to the soil (Kooch et al. 2020). In fact, soil microbial population is responsible for regulating the nutrient cycle in the soil layer and providing nutrients to the plant and thus, directly impacts plant growth and biomass production (Chen et al. 2007).

Trees can alter soil properties by changing the litter quantity and quality, organic matter, carbon to nitrogen ratio, moisture and acidity. These changes have a multiple effect on the abundance and diversity of earthworms, the activity of microbial communities, enzyme activities as well as nitrogen mineralization (Gei and Powers 2013; Tian et al. 2015). Meanwhile, soil microbial biomass carbon and nitrogen are the main components of a soil ecosystem (Burton et al. 2010) and are strongly influenced by the litter of forest stands. These components dictate many ecological processes such as the carbon and nutrient cycle, nitrogen mineralization, litter decomposition, and soil productivity (Gei and Powers 2013).

Furthermore, tree species with different litter quality contribute to the above-ground and underground environments, which in turn influences soil respiration. According to Thoms and Gleixner (2013), soil microbial respiration rate under beech, linden, maple, ash, and hornbeam litter is highly different and the highest value is observed in the stand of linden and maple species and the lowest value in beech stands. Soil enzyme activities can be a suitable indicator to quantify and monitor changes in the structure and activity of microbial communities as well as the dynamics of soil organic matter in response to forest harvesting operations (Trasar-Cepeda et al. 2008). Soil enzyme activity is often involved in the decomposition and synthesis of soil organic matter, cycle and nutrient availability, as well as soil fertility and quality (Wang et al. 2012; Moghimian et al. 2017). Soil enzyme activity is significantly associated with microbial biomass, which is also very important for soil structure formation (Ludwig et al. 2015). Research has shown that vegetation regeneration imports considerable amounts of nutrients into the soil environment, which in turn may increase soil microbial respiration and stimulate enzyme activity (Cui et al. 2019). Likewise, Moghimian et al. (2017) showed that soil enzyme activity is dependent on changes in microbial activity such as the content of nitrogen microbial biomass, because most enzyme activity occurs at the highest level of nitrogen microbial biomass.

So far, several studies have been conducted to determine the effect of land use change, forest harvesting operations and vegetation composition (litter) on physical, chemical, biological, microbial and enzyme properties of soil with conflicting results (Ponder and Tadros 2002; Jordan et al. 2003; Tan et al. 2008; Jourgholami et al. 2018; Kooch et al. 2020; Nazari et al. 2021). Studies have shown that soil compaction has negative effects on physical properties such as total porosity, pore size distribution, as well as air and water conductivity, which leads to a decrease in the soil carbon microbial biomass content (Tan et al. 2008). However, in other studies, no significant effect of soil compaction was observed on the carbon microbial content (Ponder and Tadros 2002; Jordan et al. 2003). These contentious results may be due to the unclear definition of factors affecting soil compaction during and after skidding operations, considering the short time interval between compaction event and sampling to analyze soil characteristics despite the long-term effects of soil compaction (Nazari et al. 2021).

Therefore, the present study aimed to test the effectiveness of adding litter of different tree species (leaf fall during natural processes) as a natural and ecological method to improve soil microbial and enzyme properties of soils on skid trails exposed to different traffic intensities compared to the undisturbed area over a 20-year period after skidding operations. The hypotheses of the present study were: (1) addition of litter on the compaction-induced soil can restore soil physio-chemical, microbial and enzyme properties in skid trails compared to the undisturbed area, (2) litter of different trees can have significant difference in recovery level of soil properties.

Materials And Methods

Site description

The present study was conducted in Namkhaneh and Gorazbon districts of Kheyroud forest in Hyrcanian forests (northern Iran), located at 51°36'50" E and 51°38'21" E longitude and 36°34'21" N and 36°33'34" N latitude (Fig. 1). The Namkhaneh and Gorazbon districts, with areas of 1080 and 1002 ha, are located in the altitude range of 1000–1150 and 1145–1360 meters above sea level, respectively. Based on the meteorological station data, the climate of the study area is very humid with cold winters with an annual rainfall of 1146 mm and a mean annual temperature of 8.6°C. The soils are mainly Alfisols with good drainage (Luvisols according to the World Reference Base for soil resources (WRB)), which has a loamy to loamy-clay texture with > 1 m depth. The forest type in the studied compartments is beech (*Fagus orientalis* Lipsky), beech- hornbeam (*Fageto Carpinetum*), oak - hornbeam (*Querceto-Carpinetum*), and mixed beech with maple (*Acer velutinum* Boiss.), alder (*Alnus subcordata* C.A. Mey), linden (*Tilia begonifolia*) and oak (*Quercus castanifolia* C.A.M.) species and on the southern slopes with cappadocian maple (*Acer cappadocicum* C.A.M.) and cherry tail (*Prunus avium*). The study area is covered by dominant herbaceous species including *Asperula odorata* L., *Euphorbia amygdaloides* L., *Hypericum androsaemum* L., *Polystichum* sp, *Rubus hyrcanus* Juz., *Oplismenus undulatifolius* (Ard.) Roem. & Schult.

The silvicultural treatment in the study area is a combination of single and group selection leading to the formation of heterogeneous stands. Felling and processing of trees was done with chainsaws (late winter) and transportation from the stump area to roadside landings was performed with a Timberjack 450C wheeled skidder during the spring. The Timberjack skidder is an articulated four-wheel drive vehicle with a weight of 10.3 tons (total weight with equipment) and an engine power of 177 hp. The front and rear straight axles are equipped with 775 × 813 mm tires inflated to 220 kPa and a ground clearance of approximately 0.6 m, with an overall width of 3.1 m. The width of skid trails was 3.5 m with a longitudinal slope of 5 to 20%. The last tree felling operation dates back to 6 years before the time of the study. The detailed information from the study area is given in Table 1.

Table 1

Description of skid trail/forest stand in the study area, northern Iran. B: Beech, B-H: Beech-Hornbeam, B-H-O: Mixed Beech, and UND: undisturbed area.

Harvesting period (years since harvest)	Forest stand	District (No. of Compartments)	Skid Trail Length (m)	Skid Trail Density (m ha^{-1})	Elevation (m)	Tree density (N ha^{-1})	Tree coverage (%)	Soil Texture
6	B	Gorazbon (C. 315)	255	65.5	1209	510	80	Clay
	B-H	Gorazbon (C. 316)	374	62.6	1174	496	72	Clay
	B-H-O	Gorazbon (C. 318)	310	70.4	1177	565	85	Silt clay loam
10	B	Gorazbon (C. 319)	240	76.5	1246	505	75	Clay
	B-H	Gorazbon (C. 320)	247	80.2	1345	520	80	Clay
	B-H-O	Gorazbon (C. 318)	360	85.5	1133	544	80	Silt clay loam
20	B	Namkhaneh (C. 215)	200	60.4	1040	495	85	Clay
	B-H	Namkhaneh (C. 220)	210	55.3	1115	482	75	Silt loam
	B-H-O	Namkhaneh (C. 214)	180	58.5	1010	510	80	Clay loam

Experimental design

To achieve the research objectives, skid trails with the same conditions in terms of silvicultural treatment, harvesting system and type of machinery were identified. Specifically, skid trails within areas of three different harvesting periods (6, 10, 20-years since harvest) with the same skidding direction and longitudinal slope (without considering the lateral slope) were selected in the forest. Each skid trail with three replications in three forest types (beech: B, beech-hornbeam; B-H, mixed beech: B-H-O) was identified based on litter type (Fig. 1). Since the present study is retrospective, the statistics and information of harvesting operations are based on the forestry plan documents and detailed reports from the plan managers.

According to the spatial distribution and extent of the skid trail network in relation to log landings, three traffic intensities (low, medium, and high) were identified. On the skid trails, in each treatment (e.g., a combination of litter type and traffic intensity), sampling plots with the dimensions of $4 \times 10 \text{ m}^2$ were designed. In each sample plot, three measurement lines were identified (Ezzati et al. 2012) and on each line, at a depth of 0–10 cm, soil

samples were taken at the right and left wheel tracks. Therefore, for each trail, three litter treatments and three traffic intensity classes and in each traffic class, three sampling plots were implemented (27 plots × 3 lines × 2 samples = 162). To compare the natural recovery between skid trail and undisturbed area, litter and soil samples (54 samples in each skid trail) were taken inside the forest stand at least 20–30 m away from the skid trail (based on the average height of dominant trees in the area without any effect of skidding operation). In this study, a total of 216 soil samples (3 harvesting periods × 3 litter treatments × 4 traffic intensities × 6 soil samples) were collected and analysed.

To sample soil microbial and enzyme properties, a sample plot (25 × 25 cm²) was designed in each skid trail located in three forest stands (litter of different trees) and in each tested traffic classes. Within these plots, a transect was established and 1 soil sample was taken from each of the two wheel tracks. Therefore, 18 samples were taken in each skid trail to study microbial and enzyme properties.

Data collection and laboratory analysis

Before soil samples were taken, the thickness of the litter layer was measured and the litter samples were collected in plastic bags, sealed and immediately transferred to the laboratory where they were oven-dried at 65 to 70°C for 48 hours. Litter carbon and nitrogen were measured using the burning method and the modified Kjeldahl method (Homer and Pratt 1961). Soil properties were measured in three separate sections for (1) physical properties, (2) chemical properties, and (3) microbial and enzymatic activity. To measure soil physical properties, samples were collected from mineral and surface soil (0–10 cm) using steel sampling cylinders (5 cm in diameter and 10 cm in length). Soil samples were transferred to the laboratory and oven-dried at 105°C until constant mass to obtain soil bulk density and moisture content (Blake and Hartge, 1986). Bulk density was calculated according to Eq. 1:

$$BD = \frac{M_s}{V_t}$$

1

where BD is the bulk density (g cm⁻³), M_s is the mass of soil (g), and V_t is the volume of cylinder (cm³).

The amounts of clay, silt, and sand (soil texture) were measured by hydrometric method (Bouyoucos 1962), and the particle density was measured by pycnometer method (Blake and Hartge 1986). Also, the total porosity was obtained by using the bulk density and particle density values (Pires et al. 2014).

To analyze soil chemical properties, 2 kg of soil was collected for other experiments and transferred to the laboratory. Soil pH was determined using an Orion Ionalyzer Model 901 pH meter in a 1:2.5, soil: water solution (Kooch et al. 2017). The Walkley–Black technique (Allison 1975) was used to determine soil organic carbon. Total N was measured using a Kjeldahl technique (Bremner and Mulvaney. 1982). Available phosphorous (P) was determined with a spectrophotometer using the Olsen method, and available potassium (K), calcium (Ca) and magnesium (Mg) (by ammonium acetate extraction at pH 9) were determined with an atomic absorption spectrophotometer (Kooch et al. 2014). The soil humic acid and fulvic acid were isolated and purified according to the method of the International Humic Substances Society (IHSS) (Sparks and Bartels 1996).

For microbial analysis and enzyme activity, another part of the soil samples was stored at 4°C. Evolved CO₂ was measured in a 3-day incubation experiment at 25°C in order to evaluate soil microbial respiration (Alef 1995). The microbial biomass carbon (MBC) and nitrogen (MBN) in the soils was determined by the chloroform fumigation–extraction method (Vance et al. 1987). Soil NH₄⁺ and NO₃⁻ were extracted with a 2 M KCl solution (soil: solution, 1:5) and determined using the colorimetric techniques (Yang et al. 2017).

Urease activity (EC 3.5.1.5) was determined using 200 µmol urea as substrate under incubated conditions (2 h at 37°C) (Moghimian et al. 2017). Acid phosphatase activity (EC 3.1.3.2) was measured using 15 mM p-Nitrophenyl phosphate disodium (PNP) as substrate in a modified universal buffer (MUB) at pH 6.5, incubated for 1 h at 37°C (Yang et al. 2017). Arylsulphatase activity (EC 3.1.6.1) was assayed following incubation of the soils with p-Nitrophenyl sulphate (25 mM) for 1 h at 37°C and measurement of the quantity of p-Nitrophenol liberated during enzymatic hydrolysis by spectrophotometry. To detect invertase, 1.2% sucrose solution was used for incubation at 3 h at 50°C (EC 3.2.1.26) (Schinner and von Mersi 1990).

Statistical analyses

The collected data was stored in Excel software as a database. Data normality was checked by the Kolmogorov-Smirnov test (the data was normal; sig > 0.05), while the homogeneity of variances was tested using the Levene's test (the variances were homogeneous; sig > 0.05). Three-way analysis of variance was used to investigate the recovery process (difference or no difference between the values of different litter and soil properties) in relation to litter type, traffic intensity and time since harvest. When significant differences among treatments were found by ANOVA, Duncan multiple comparison test was used to compare the mean recovery values of soil physical, chemical, microbial and enzyme properties. Pearson correlation was used to test the relationship between litter type, traffic intensity, time since harvest and soil properties. All statistical tests were performed by the SPSS software package (release 17.0; SPSS, Chicago, IL, USA). For multivariate correlation analysis of litter and soil properties with main components (litter type, traffic intensity and skid trail age) after standardizing the data with correlation matrix, the principal component analysis (PCA) method is used in PC-ORD software (Version 4, WILD BLUEBERRY MEDIA LLC, Corvallis, OR, USA).

Results

Litter and soil physico-chemical properties

ANOVA indicated that there were significant differences in the litter properties from different stands and time since harvest (Fig. 2). The highest amount of litter C (37.9%) belonged to the beech stand in the 20-years since harvest (6.2% less than the UND) and the lowest level (16.2%) at the mixed beech stand in the 6-years since harvest (21.7% less than the UND) (Fig. 2. a). Litter C/N ratio was significantly higher in the beech stand and in the 6-years since harvest (62.4% higher than the UND) than other stands and harvesting periods (years since harvest) (Fig. 2. c). Also, the highest litter N (2.5%) was observed in the mixed beech stand in the 20-years since harvest (8.5% less than the UND) and the lowest in beech and the 6-years since harvest (Fig. 2. b). The highest litter thickness belonged to the beech stand (10.9%) and in the 20-years since harvest (10.9% less than the UND) and the lowest level in mixed beech stand and 6-years since harvest (Fig. 2. d).

Results showed that there was a significant difference in the soil physio-chemical properties among different treatments (except BD, SM and Silt) (Table 2). Soil physical and chemical properties were significantly improved 20 years after harvesting with the addition of litter, where the recovery of soil physical and chemical properties was observed with an increase in litter quality (B-H-O > B-H > B) compared to the control treatment. The values of TP, SM, silt, clay, pH, N, available P, K, Ca, and Mg, fulvic and humic acid were at the highest level in the mixed beech treatment (B-H-O) followed by B-H > B, but these values were less than the undisturbed area even 20-years since harvest. In contrast, BD, sand, C and C/N ratio were at the lowest level in the mixed beech treatment (B-H-O) followed by B-H < B, but these values were higher than the undisturbed area over a 20-year period after skidding operations. Soil physical and chemical properties (except for clay, C and N) in the skid trail and litter treatments corresponding to the 20-years since harvest were significantly different to those measures at the undisturbed area (Table 2).

Table 2

Mean values (\pm SD) of the soil physical and chemical properties twenty years since harvest under litter treatments. B: Beech, B-H: Beech-Hornbeam, B-H-O: Mixed Beech, and UND: Undisturbed area. Soil physical properties (BD: bulk density, TP: total porosity, SM: soil moisture).

Litter treatments						
Soil Properties	B	B-H	B-H-O	UND	<i>F</i> test	<i>P</i> value
BD (g cm ⁻³)	1.11 \pm 0.00b	1.09 \pm 0.00b	1.05 \pm 0.00b	0.91 \pm 0.00a	10.258	0.000
TP (%)	57.81 \pm 0.11b	58.46 \pm 0.08b	60.04 \pm 0.07ab	65.55 \pm 0.11a	10.358	0.000
SM (%)	23.14 \pm 0.25b	23.68 \pm 0.18b	24.66 \pm 0.16b	31.96 \pm 0.23a	10.368	0.000
Sand (%)	44.82 \pm 0.29a	43.49 \pm 0.21a	42.73 \pm 0.19ab	41.89 \pm 0.28b	15.522	0.000
Silt (%)	30.36 \pm 0.23b	30.39 \pm 0.16b	30.51 \pm 0.15b	31.92 \pm 0.21a	5.721	0.000
Clay (%)	24.82 \pm 0.22b	26.12 \pm 0.16a	27.06 \pm 0.15a	26.19 \pm 0.21a	8.126	0.000
pH (1:2.5 H ₂ O)	5.61 \pm 0.01d	6.19 \pm 0.00c	6.68 \pm 0.00b	6.81 \pm 0.00a	4.597	0.001
C (%)	6.41 \pm 0.00a	5.41 \pm 0.00b	4.41 \pm 0.00c	4.23 \pm 0.00c	16.345	0.000
N (%)	0.31 \pm 0.00c	0.43 \pm 0.00b	0.51 \pm 0.00a	0.53 \pm 0.00a	11.319	0.000
C/N ratio	21.01 \pm 0.09a	12.69 \pm 0.07a	8.76 \pm 0.06b	7.94 \pm 0.09c	194.250	0.000
Available P (mg kg ⁻¹)	17.72 \pm 0.19b	22.72 \pm 0.13ab	26.71 \pm 0.13a	28.47 \pm 0.18a	11.971	0.000
Available K (mg kg ⁻¹)	173.61 \pm 0.17d	197.83 \pm 0.12c	221.75 \pm 0.11b	230.74 \pm 0.16a	99.445	0.000
Available Ca (mg kg ⁻¹)	146.04 \pm 0.38c	167.08 \pm 0.27b	183.90 \pm 0.25ab	199.91 \pm 0.36a	56.346	0.000
Available Mg (mg kg ⁻¹)	44.74 \pm 0.13d	54.75 \pm 0.09c	64.79 \pm 0.09b	74.72 \pm 0.12a	456.690	0.000
Fulvic acid (mg/100 g)	172.93 \pm 0.75d	208.80 \pm 0.53c	326.86 \pm 0.49b	349.15 \pm 0.69a	11.649	0.000
Humic acid (mg/100 g)	143.05 \pm 0.39d	163.22 \pm 0.28c	192.61 \pm 0.26b	219.40 \pm 0.37a	47.391	0.000

Note: Results of the ANOVAs (*F* test and *P* value) are given. Different letters within each treatment indicate significant differences by Duncan test ($P < 0.05$).

Soil microbial and enzyme properties

Soil microbial (except for NH_4^+ between B and B-H treatments 6-years since harvest and NO_3^- between B-H-O and UND from 20-years since harvest) and enzyme properties (except for Urease between B and B-H treatments from 20-years since harvest) were significantly different among treatments (litter type and skid trail age) (Tables 3 and 4). The recovery rate of soil microbial and enzyme properties increased with increasing litter type quality and time since harvest, but was still lower than the values under UND treatment. The highest value of soil microbial characteristics (SMR, MBC, MBN, NH_4^+ , and NO_3^-) and enzyme activity (Urease, Acid phosphatase, Arylsulfatase, and Invertase) belonged to B-H-O treatment followed by > B-H > B and 20-years since harvest followed by > 10 years > 6 years. The highest values of SMR ($0.51 \text{ mg CO}_2\text{-C g soil}^{-1} \text{ day}^{-1}$), MBC ($545.04 \text{ mg kg}^{-1}$), MBN (50.96 mg kg^{-1}), NH_4^+ (34.97 mg kg^{-1}) and NO_3^- (32.08 mg kg^{-1}) belonged to B-H-O and 20-years since harvest. Also, the highest values of Urease ($29.38 \text{ } \mu\text{g NH}_4^+\text{-Ng}^{-1} \text{ 2 h}^{-1}$), Acid phosphatase ($353.47 \text{ } \mu\text{g p-nitro phenol g}^{-1} \text{ h}^{-1}$), Arylsulfatase ($154.95 \text{ } \mu\text{g p-nitro phenol g}^{-1} \text{ h}^{-1}$) and Invertase ($186.34 \text{ } \mu\text{g Glucose g}^{-1} \text{ 3 h}^{-1}$) belonged to the B-H-O and 20-years since harvest. In contrast, the lowest values of these characteristics (soil microbial and enzyme properties) were measured in the beech treatment (B) and 6-years since harvest (Tables 3 & 4). Twenty years since harvest in mixed beech litter (B-H-O), the highest values of SMR, MBC, MBN, NH_4^+ , NO_3^- were less by 15%, 8.71%, 15.39%, 13.55%, and 9.76% than the undisturbed area respectively (Table 3). Also, twenty years since harvest, the values of urease, acid phosphatase, arylsulfatase, Invertase in B-H-O were 10.23%, 4.37%, 8.8% and 6.87% lower than the undisturbed area (Table 4).

Table 3

Mean values (\pm SD) of the soil microbial properties in different years since harvest under litter different treatments. B: Beech, B-H: Beech-Hornbeam, B-H-O: Mixed Beech, and UND: Undisturbed area. SMR: Soil microbial respiration, MBC: Microbial biomass carbon, MBN: Microbial biomass nitrogen.

Harvesting period (years since harvest)	Litter type	soil properties				
		SMR (mg CO ₂ -C g soil ⁻¹ day ⁻¹)	MBC (mg kg ⁻¹)	MBN (mg kg ⁻¹)	NH ₄₊ (mg kg ⁻¹)	NO ₃ ⁻ (mg kg ⁻¹)
6	B	0.24 \pm 0.00d	188.92 \pm 0.85d	23.65 \pm 0.11d	11.21 \pm 0.65c	9.45 \pm 0.09d
	B-H	0.34 \pm 0.00c	335.28 \pm 0.84c	34.14 \pm 0.11c	18.78 \pm 0.64c	17.87 \pm 0.08c
	B-H-O	0.44 \pm 0.00b	485.06 \pm 0.59b	44.82 \pm 0.11b	29.07 \pm 0.46b	27.97 \pm 0.06b
10	UND	0.60 \pm 0.00a	597.43 \pm 0.79a	60.26 \pm 0.1a	40.49 \pm 0.60a	35.52 \pm 0.08a
	B	0.27 \pm 0.00d	211.77 \pm 0.85d	26.74 \pm 0.11d	12.81 \pm 0.65c	11.38 \pm 0.09c
	B-H	0.38 \pm 0.00c	363.32 \pm 0.69c	37.59 \pm 0.09c	22.16 \pm 0.53b	19.59 \pm 0.07c
20	UND	0.6 \pm 0.00a	515.94 \pm 0.59b	47.78 \pm 0.08b	31.59 \pm 0.46ab	29.54 \pm 0.06b
	B-H	0.41 \pm 0.00c	599.24 \pm 0.79a	60.02 \pm 0.1a	36.28 \pm 0.6a	35.57 \pm 0.08a
	B-H-O	0.51 \pm 0.00b	241.17 \pm 0.85d	29.78 \pm 0.11d	15.01 \pm 0.65c	13.08 \pm 0.09c
	B	0.29 \pm 0.00d	392.92 \pm 0.60c	40.31 \pm 0.08c	24.95 \pm 0.46b	21.96 \pm 0.06b
	B-H	0.41 \pm 0.00c	545.04 \pm 0.57b	50.96 \pm 0.07b	34.97 \pm 0.43ab	32.08 \pm 0.06a
	B-H-O	0.51 \pm 0.00b	597.08 \pm 0.79a	60.23 \pm 0.1a	40.45 \pm 0.60a	35.55 \pm 0.08a

Note: Results of the ANOVAs (F test and P value) are given. Different letters after means within each treatment indicate significant differences by Duncan test ($P < 0.05$).

Harvesting period (years since harvest)	Litter type	soil properties				
		SMR (mg CO ₂ -C g soil ⁻¹ day ⁻¹)	MBC (mg kg ⁻¹)	MBN (mg kg ⁻¹)	NH ₄ ⁺ (mg kg ⁻¹)	NO ₃ ⁻ (mg kg ⁻¹)
<i>F</i> test		2.292	8.893	2.744	1.449	7.971
<i>P</i> value		0.0498	0.000	0.028	0.002	0.000

Note: Results of the ANOVAs (*F* test and *P* value) are given. Different letters after means within each treatment indicate significant differences by Duncan test ($P < 0.05$).

Table 4

Mean values (\pm SD) of the soil enzyme properties in different years since harvest under litter different treatments. B: Pure Beech, B-H: Beech-Hornbeam, B-H-O: Mixed Beech, and UND: Undisturbed Area.

Harvesting period (years since harvest)	Litter type	soil properties			
		Urease (μg $\text{NH}_4^+ - \text{N g}^{-1}$ 2 h^{-1})	Acid phosphatase ($\mu\text{g p-nitro phenol g}^{-1}$ 1 h^{-1})	Arylsulfatase (μg $\text{p-nitro phenol g}^{-1}$ h^{-1})	Invertase (μg $\text{Glucose g}^{-1} 3$ h^{-1})
6 years	B	10.56 \pm 0.09c	142.34 \pm 1.91d	54.42 \pm 0.32d	63.51 \pm 0.39d
	B-H	18.61 \pm 0.09bc	221.36 \pm 1.88c	92.25 \pm 0.32c	106.34 \pm 0.38c
	B-H-O	25.51 \pm 0.07b	320.51 \pm 1.34b	134.91 \pm 0.23b	157.95 \pm 0.27b
	UND	32.93 \pm 0.09a	378.51 \pm 1.34a	169.81 \pm 0.29a	200.15 \pm 0.36a
10 years	B	13.6 \pm 0.09c	158.33 \pm 1.77d	68.05 \pm 0.32d	
	B-H	20.46 \pm 0.08b	238.12 \pm 1.55c	103.44 \pm 0.26c	74.6 \pm 0.39d
	B-H-O	27.39 \pm 0.07ab	337.95 \pm 1.34b	144.72 \pm 0.23b	119.16 \pm 0.32c
20 years	B-H-O	27.39 \pm 0.07ab	381.26 \pm 1.77a	170.74 \pm 0.29a	171.92 \pm 0.27b
	UND	32.58 \pm 0.09a	175.18 \pm 1.91d	77.79 \pm 0.32d	200.36 \pm 0.36a
	B	15.59 \pm 0.09c	255.08 \pm 1.35c	114.73 \pm 0.23c	
	B-H	22.12 \pm 0.07c	353.47 \pm 1.26b	154.95 \pm 0.21b	84.72 \pm 0.39d
	B-H-O	29.38 \pm 0.06ab	369.61 \pm 1.77a	169.9 \pm 0.29a	133.37 \pm 0.27c
	UND	32.73 \pm 0.09a			186.34 \pm 0.26b
					200.08 \pm 0.36a
<i>F</i> test		19.946	10.508	14.619	27.066
<i>P</i> value		0.000	0.000	0.000	0.000

Note: Results of the ANOVAs (*F* test and *P* value) are given. Different letters after means within each treatment indicate significant differences by Duncan test ($P < 0.05$).

The results of PCA analysis in relation to litter type, time since harvest, traffic intensities and soil properties showed that the first and second axes accounted for 79.96 and 18.13% of the explained variance, respectively. The fractions of litter and soil physio-chemical properties (N litter, SM, sand, clay, silt, pH, soil N, available P, k, Ca, Mg, fulvic acid and humic acid), microbial properties (SMR, MBC, MBN, MBC/MBN, NH_4^+ and NO_3^-), enzyme activities (urease, acid phosphatase, arylsulfatase and invertase) were enhanced at sites with mixed beech litter (B-H-O), older skid trail (ST20) and undisturbed area (UND). Instead, all of these soil properties were negatively correlated with the treatments on the left side of the graph (ST6, ST10, B, B-H, HT, MT and LT). The left side of the graph in connection with beech litter (B), younger skid trail (ST6) and high traffic intensity (HT), increase in bulk density, soil C, litter C and litter C/N ratio and decrease in total porosity were observed (Fig. 3).

The results of correlation analysis showed that most of the measured properties of litter and soil have a significant correlation with each other. The key variables of litter and soil (such as litter and soil C, litter and soil C/N ratio, and litter thickness) have a significant negative correlation with soil microbial (SMR, MBC, MBN, MBC/MBN, NH_4^+ and NO_3^-), enzyme (urease, acid phosphatase, arylsulfatase and invertase) and chemical (pH, available P, k, Ca, Mg, fulvic acid and Humic acid) properties. In contrast, litter and soil N have a significant positive correlation with soil microbial, enzyme and chemical properties (Fig. 4). The correlation of litter variables with soil physical properties is not significant.

Discussion

Litter and soil physico-chemical properties

Results of the present study indicate a significant difference in litter quality among skid trails with different years since harvest, which can be attributed to the type of litter with different quality between skid trails due to difference in tree species. The quality of litter, its production and decomposition are some of the factors affecting soil properties that provide favorable conditions for the formation of soil organic matter and nutrient cycle (Kooch et al. 2021).

The high amount of nitrogen in the B-H-O treatment from all three skid trails with different years since harvest resulted in increase in the decomposition of the litter and finally reduced the litter thickness compared to other treatments. These results are in line with previous studies showing that beech litter in B treatment with high lignin and low nitrogen, has a higher C/N ratio, which ultimately leads to a reduction in decomposition rate (Jourgholami et al. 2018). Therefore, the highest litter thickness was observed in B treatment. In skid trails with different years since harvest, the B-H-O treatment with production of suitable quality litter (higher nitrogen and less carbon) compared to beech-hornbeam (B-H) and beech (B) treatments (less nitrogen and higher carbon) leads to increase in organic matter quality and improve soil properties. In this regard, Zhang et al. (2015) pointed out that different tree species have varying litter qualities due to differences in carbon content, nitrogen and carbon to nitrogen ratio of litter, which influences on the intensity of their decomposition. The more nitrogen in the leaf, the faster it decomposes (Moghimian et al. 2017). Twenty years since harvest, litter layer characteristics showed the highest recovery compared to 10-years and 6-years since harvest, as it may take more than a decade to fully recover litter organic properties after harvesting operations (Mo et al. 2003).

Physical properties of the soil under litter treatments (pure beech, beech-hornbeam and mixed beech) were significantly recovered 20 years since harvest. Long-term recovery of soil properties under natural conditions has also been reported in previous studies (Labelle and Jaeger, 2011; Sohrabi et al. 2019; DeArmond et al. 2020). Changes in bulk density and total porosity can be attributed to the litter decomposition rate of soil surface and the amount of organic matter (Jourgholami et al. 2018). The decrease in bulk density (1.11 to 1.05 g cm^{-3}) and the increase in total soil porosity (58–60%) from pure beech to mixed beech can be related to the rapid decomposition of the litter (low carbon and high nitrogen).

Previous studies have reported that soil moisture increases with an increase in the amount of soil litter (Kader et al. 2017). The results of the present study showed that soil moisture did not differ significantly in different litter treatments. However, soil moisture did increase with an increase in the thickness of the litter layer.

Although soil texture is one of the constant characteristics of soil, changing the type of vegetation and litter can affect the changes in soil texture components over a long period of time (Meyfroidt et al. 2013). In this study, it was also observed that by changing the litter type from pure beech to mixed beech, the proportion of soil particles (sand and clay) changed. In general, the litter layer, as a protective cover, intercepts the erosive power of raindrops and reduces the clay particle detachment (Jourgholami et al. 2020).

Many of the chemical reactions that affect access to nutrients are influenced by the chemical environment, in particular soil pH (Schoenholtz et al. 2000). Species differences in the production of different organic acids resulting from the litter decomposition, which changes the ratio of exchange bases (such as Ca and Mg) and acidic cations (such as Fe and Al) in the soil can be effective on soil pH (Finzi et al. 1998). Results of this study showed that the soil in B treatment has a lower pH than B-H and B-H-O treatments ($B < B-H < B-H-O$) 20 years since harvest. In other words, the production of low-quality litter in pure beech treatment resulted in more acidic ambience in soil (Guckland et al. 2009; Jourgholami et al. 2018).

The quality of the litter layer and its diversity can result in an enhanced soil structure because high quality litter has a significant amount of N, which leads to the lowest C/N ratio in the litter, thus increasing the decomposition rate and natural recovery of compacted soil (Cools et al. 2014). In other words, the degree and extent of mineralization and the produced humus type depend on the type and amount of chemical compounds in the organic matter (Jourgholami et al. 2018). Current results showed that N was more B-H-O treatment, but (B), the amount of C and consequently C/N ratio were higher in beech litter treatment instead. Previous studies have also concluded that tree species with higher lignin and lower N have a significant decomposition rate, which lead to a lower C/N ratio on both the litter layer and in mineral soil (Guckland et al. 2009).

The highest recovery level of available soil nutrients (i.e., P, K, Ca, and Mg), humic and fulvic acids in the B-H-O treatment compared to B-H and B treatments can be attributed to low lignin content and high soil nitrogen content (Aponte et al. 2013; Jourgholami et al. 2021). Since soil pH in mixed beech treatment (due to high quality litter production) is higher than in the B and B-H treatments, the conditions are favorable for more activity of microorganisms, which ultimately resulted in an increase in the soil nutrients (Kader et al. 2017; Jourgholami et al. 2018). Also, the increase of nutrients in this treatment can be related to the increase of clay and the decrease of sand within the soil. The high sand content and associated low clay content on the one hand cause proper aeration, but on the other hand cause lack of storage and waste of nutrients and its transfer to the subsoil layers (Kooch et al. 2021). In line with the results of this study, previous studies also reported an increase in humic and fulvic acids by planting trees and increasing litter to the soil surface (Lima et al. 2006; Jourgholami et al. 2020). Furthermore, Abakumov et al. (2013) reported humic and fulvic acids increased with the site age in the reclaimed land by planting alder.

Soil microbial and enzyme properties

Soil microbial activity in forest habitats is affected by several factors including litter quality, organic matter content, nitrogen content, moisture content, soil texture, tree cover and the intensity of forest interference (Gorobtsova et al. 2016). This study showed that with increasing time since harvest and litter quality, SMR, MBC, MBN, NH_4^+ and NO_3^- were recovered in comparison with the undisturbed area. According to the results, the B-H-O treatment in the 20-years since harvest had the highest amount of SMR, MBC, MBN,

MBC/MBN, NH_4^+ and NO_3^- , while the B treatment in the 6-years since harvest showed the lowest amount of these characteristics. The high soil microbial respiration can be attributed to the high quality of mixed litter and soil (high N content low C/N ratio of mixed litter and soil) and alkaline pH under the B-H-O treatment (Burton et al. 2010; Moghimian et al. 2017; Kooch, et al. 2020; Jourgholami et al. 2020). Soil nutrient content (e.g., P, K, Ca, and Mg) acts as a major regulator of soil microbial respiration under different land cover (Tardy et al. 2014). According to the results of Tardi et al. (2014), the decrease in soil fertility, due to limited nutrient resources available to the soil microbial population, can have a negative impact on soil microbial respiration, which was also observed in the current study (Table 3).

The litter layer regulates fluctuations in soil moisture and temperature, and ultimately soil microbial activity (Fang et al. 2011; Kader et al. 2017; Jourgholami et al. 2020). According to previous studies, soil moisture content can also be an effective parameter in soil carbon dioxide emissions (Zhao et al. 2019). In fact, the amount of microbial activity in soils with lower moisture content is reduced (Kooch, et al. 2020). With the gradual increase of soil moisture, soil conditions become a favorable ambience for the activity of many microorganisms, which lead to increased microbial activity and the greater respiration in the soil (Sotta et al. 2006). In the current study, the highest soil moisture was observed in the 20-years since harvest with the highest litter thickness and B-H-O treatment, which also had the highest amount of microbial respiration. Changing the litter type of surface layer can affect the amount of microbial respiration by changing the ratio of soil texture components (Kooch et al. 2021). The presence of vegetation and litter layer on the soil surface prevents losses and erosion of soil particles and leads to the formation of heavy-textured soil. Liao et al. (2012) found that forest cover could improve soil texture and ultimately soil microbial biomass by increasing litter quantity. In other words, the components of soil texture affects the spaces between soil particles as well as the carbon dioxide emission. The amount of microbial biomass is high in soils with high clay contents (kooch et al. 2021). According to the present study, the highest percentage of clay was allocated to mixed beech litter treatment and was closely related to microbial respiration 20 years since harvest (Fig. 4).

Microbial biomass is an important component of forest ecosystems. A small change in microbial biomass can have a major impact on plant nutrient availability, at least in the short term. Based on the current results, amounts of SMR, MBC, MBN, NH_4^+ and NO_3^- were at the highest level in the B-H-O treatment followed by B-H > B; in contrast, the lowest value of these properties was in pure beech treatment due to the low quality and acidity of the litter and the lower decomposition rate (Aponte et al. 2013). In line with the results of this study, Allen and Schlesinger (2004) found that high nitrogen content increases microbial biomass of nitrogen in the soil mineral layer. This result indicates that the diversity in the microbial biomass nitrogen in this study can be controlled by soil nitrogen. According to the findings of Singh et al. (2012), soil nitrogen restriction can reduce microbial activity at high C/N ratios (negative correlation between microbial activity and C/N ratio: Fig. 4), which is more pronounced in pure beech litter treatment.

PCA analysis showed that there was a high correlation between microbial activity with litter and other soil properties (pH, N, NH_4^+ , NO_3^- , and microbial respiration) in B-H-O treatment. The results showed that the amount of NH_4^+ and NO_3^- in the B-H-O treatment was higher than other treatments. The mineralization rate of available nitrogen (NH_4^+ and NO_3^-) is strongly influenced by the type and quality of tree litter, so that under mixed litter and of good quality (B-H-O treatment), it is higher than pure and low-quality litter (B treatment). This

is likely due to more litter N, lower C/N ratio, and faster decomposition rate of organic matter in mixed litter than pure litter (Kooch et al. 2017). In this regard, Kader et al. (2017) concluded that the activity of nitrogen-fixing bacteria, which convert N_2 released to ammonia (NH_3) leading to an increase in N content. According to the results of previous studies, increasing the pH leads to increasing the nitrogen mineralization rate (Kooch et al. 2021). In the results of the present study, it was found that pH has a strong correlation with the amounts of NH_4^+ and NO_3^- (Fig. 4).

The results of our study showed that the highest enzyme activity (i.e., urease, acid phosphatase, aryl sulfatase and invertase) was obtained 20 years since harvest in B-H-O treatment. Previous studies indicated that there is a strong positive correlation between the enzyme activity with pH, N and other nutrients (Zhang et al. 2015; Kader et al. 2017; Moghimian et al. 2017; Jourgholami et al. 2020; Kooch et al. 2021). In addition, improving soil physical properties through the creation of a more favorable environment along with changes to soil nutrients (suitable conditions such as pH, C and N) will lead to increased microbial and enzyme activity (Moghimian et al. 2017). The results of the relationship between soil properties showed that there was a positive correlation between the enzyme activities with pH, N, available nutrients, while they had a negative correlation with C and C/N ratio (Fig. 4). In line with these results, the study of Zeng et al. (2009) showed that there was a positive correlation between invertase activity and pH, C, N and available P in soil.

Conclusions

In the present study, the effectiveness of adding litter of different trees (leaf fall during natural processes) as a natural and ecological modification method was evaluated to improve the soil physical, chemical, microbial and enzyme properties of skid trails over a 20-year period after skidding operations. Results showed that the addition of litter of different trees (B, B-H, B-H-O treatments) can gradually improve the physical, chemical, microbial properties of the soil as well as enzyme activity over a 20-year period after harvesting compared to the undisturbed area, which support the first research hypothesis. However, soil properties recovery and enzyme activity in skid trails were still lower than in undisturbed areas. Recovery of microbial properties and enzyme activities under the influence of high-quality litter (mixed beech) is positively associated with other soil properties including pH, N, available nutrients. Based on our results, twenty years was not long enough to return the soil properties to pre-harvest levels (with the value of UND treatment used as a reference point). Based on the second hypothesis, the recovery rate of soil properties was different among litter treatments, so that the highest soil recovery was obtained in B-H-O treatment followed by B-H > B and 20-years since harvest followed by 10 years > 6 years. Therefore, the results of the present study indicate the positive effect of litter of different trees as one of the factors to improve soil properties. Overall, it seems that mixed beech litter treatment has been able to improve the soil properties under study more than other treatments. In general, the management activities that can be done to improve soil properties under natural and ecological forest conditions are summarized as follows:

- Prevent the removal of litter cover from the soil surface in skid trails by using soil protection activities (use of different foliage mulch and brush mats, limiting ground-based skidding operations to gentle slope gradient, reduce the number of machine passes, choice of operating season and meteorological conditions) during forest skidding operations.

- As a rapid ecological response to soil disturbance after skidding operations, the addition of tree litter, depending on its quantity and quality, improves soil properties.
- According to the results of this study, mixed beech litter (beech, hornbeam, alder, linden, maple, Cappadocian maple, etc.) is considered as the preferred treatment.
- As a long-term aim in forestry plans, it is recommended to plant native and suitable deciduous species for the restoration of degraded natural forests, at least in skid trails (area with high disturbance intensity).

Declarations

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Authors' contributions Hadi Sohrabi and Meghdad Jourgholami conceived and designed the experiments. Material preparation and data collection were performed by Hadi Sohrabi and Meghdad Jourgholami. Hadi Sohrabi and Meghdad Jourgholami performed the experiments and analyzed the data. All authors wrote, read and approved the final manuscript.

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Figures

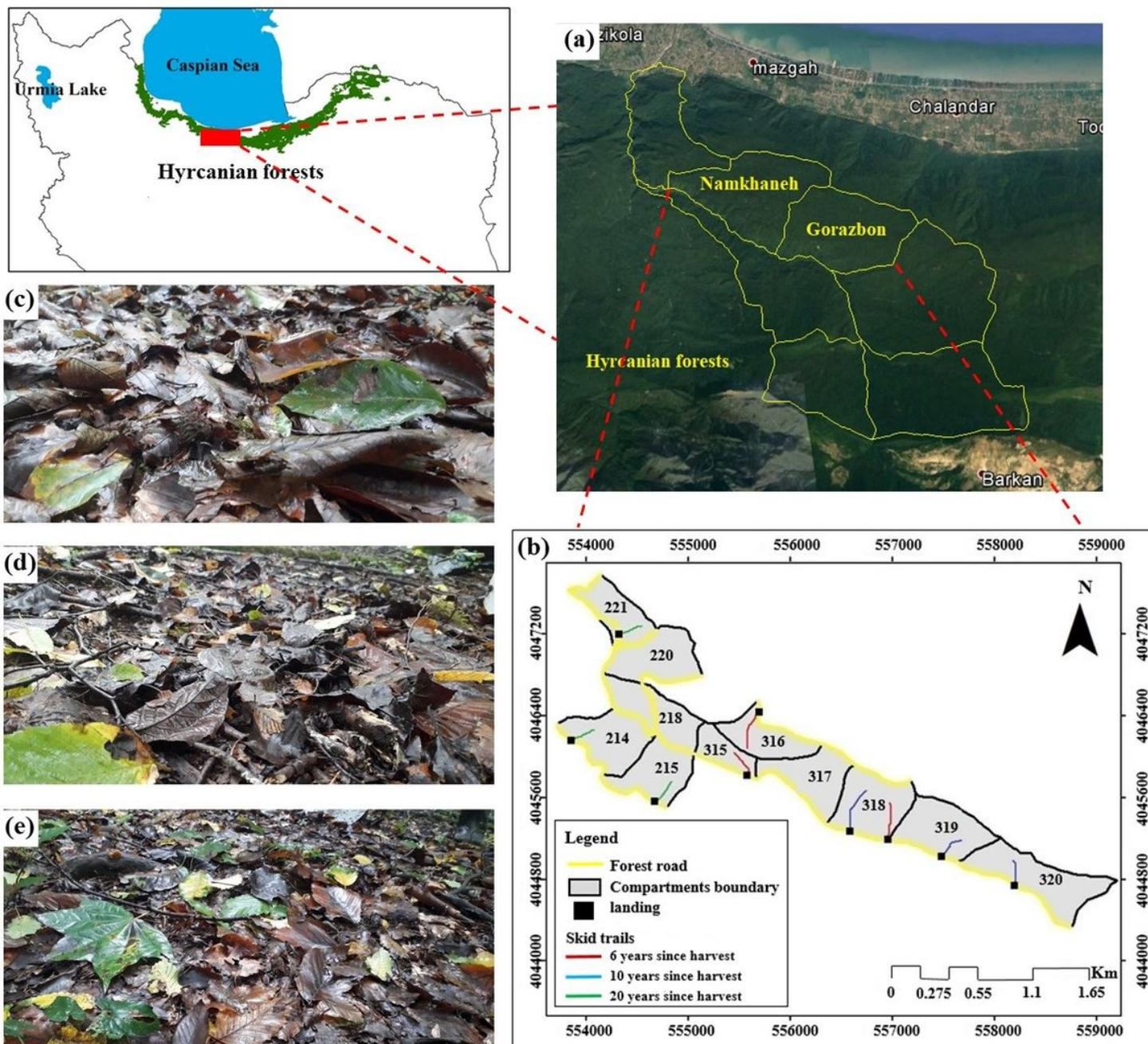


Figure 1

Study area (Namkhaneh and Gorazbon districts of Kheyroud forest) in northern Iran (a); the location of skid trails with different harvesting periods (6, 10, and 20 years since harvest) in the selected compartments after logging operations (b); Beech litter (c), Beech-Hornbeam litter (d) and mixed Beech litter (e) on the skid trail after logging operations.

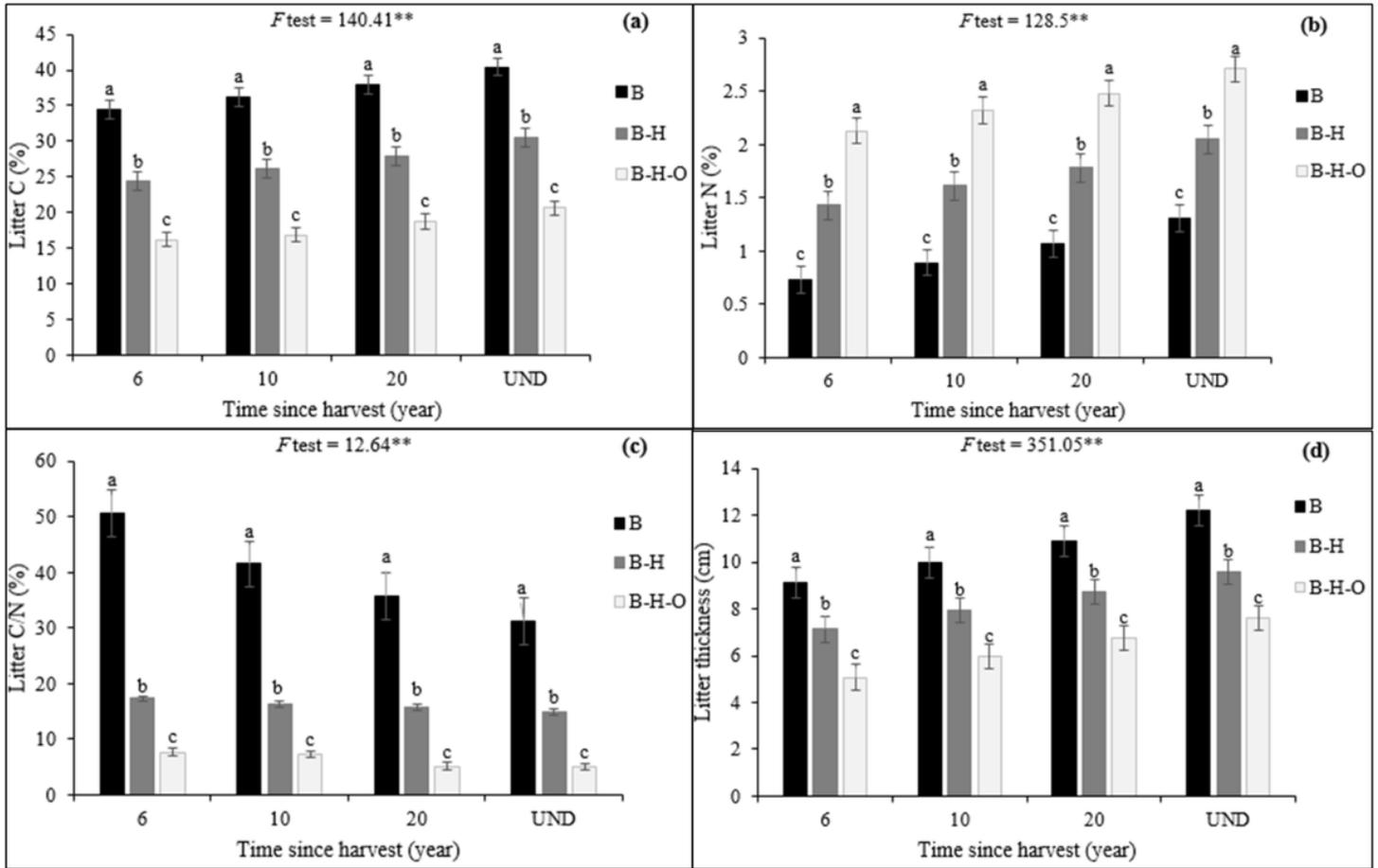


Figure 2

Mean (\pm SE) contents of litter C, N, C/N ratio and litter thickness under litter treatments and time since harvest (years). * $P < 0.05$, ** $P < 0.01$. B: Beech, B-H: Beech-Hornbeam, B-H-O: Mixed Beech, and UND: Undisturbed area. Different letters indicate a significant difference in mean by Duncan test ($P < 0.05$).

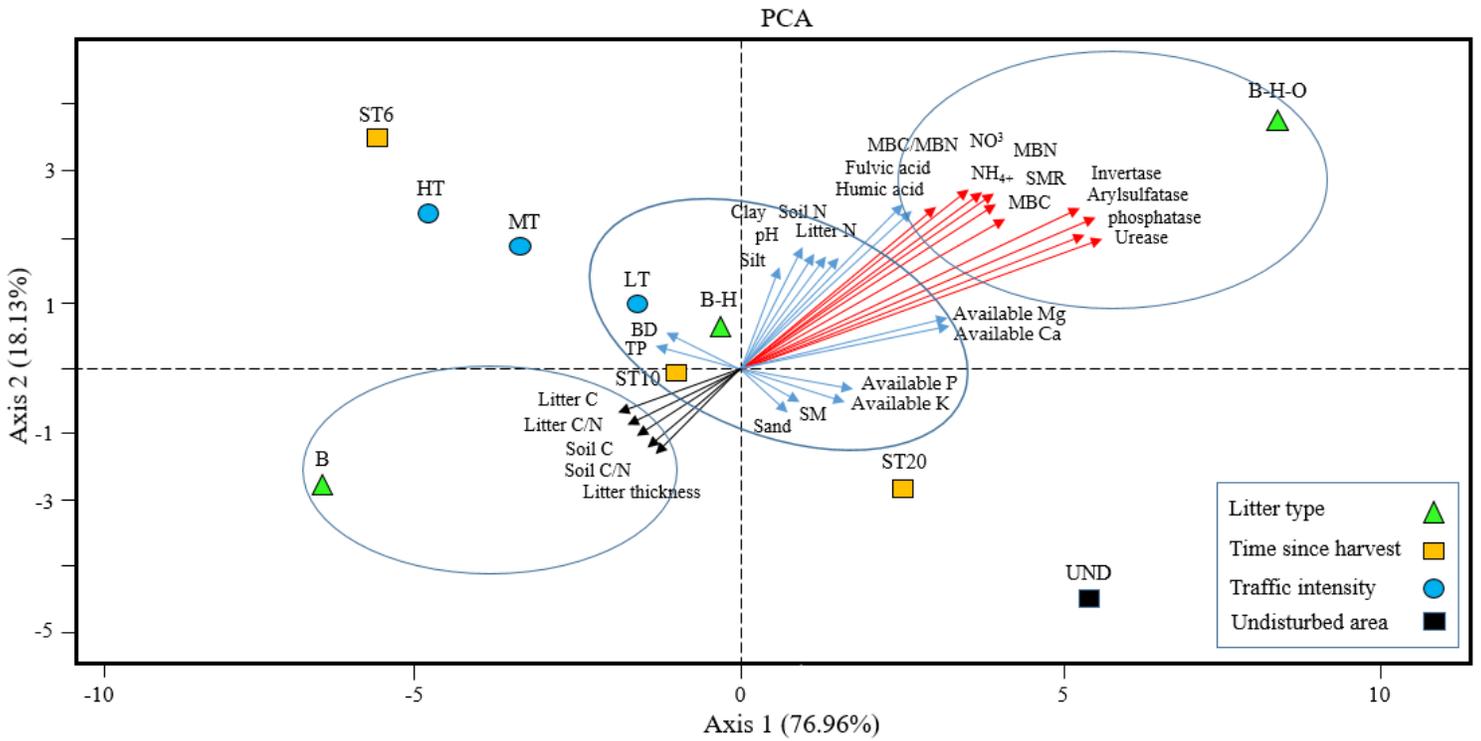


Figure 3

Principal component analysis (PCA) of litter properties and soil physio-chemical properties and enzyme activities at various treatments (Litter type: B; Beech, B-H; Beech-Hornbeam, B-H-O; Mixed Beech; time since harvest: ST6; 6 years, ST10; 10 years, ST20; 20 years; Traffic intensity: HT; High traffic, MT; Medium traffic, LT; Low traffic, and UND: Undisturbed Area. Soil physical properties (BD: bulk density; TP, total porosity; SM, soil moisture). SMR: Soil microbial respiration, MBC: Microbial biomass carbon, MBN: Microbial biomass nitrogen.

