

# The effect of mechanical site preparation on sandy soil properties in Scots pine plantations

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**Abstract:** Sandy soils represent an extreme environment for tree growth. Traditionally, site preparation before planting involves removing logging residues (LRR) and ploughing. An alternative method is incorporating logging residues (LRI) into the topsoil which may enhance tree regeneration and seedling growth. The aim of this study was to assess whether and how different site preparation techniques affect soil physico-chemical and microbial properties over the long term. The study was performed in the Záhorská nížina lowland (Slovakia) in September 2020. Soil samples were taken in two 25-year-old *Pinus sylvestris* (L.) plantations along five soil profiles in each stand, down to a depth of 30 cm. Results showed a significant increase in carbon and nitrogen concentration and soil moisture in the LRI plot. However, soil pH and phosphorus content significantly decreased. No significant differences were observed in calcium, magnesium, and potassium concentrations between the differently treated plots. The LRI plot also exhibited a significant increase in microbial biomass carbon, N-mineralisation, and catalase activity. The results indicate that different mechanical site preparation methods may impact soil properties over the long term, likely through improved seedling survival and tree growth.

**Keywords:** logging residues; microbial properties; physico-chemical properties; *Pinus sylvestris* (L.); soil preparation

Sandy soils are defined as containing less than 18% of clay and more than 68% of sand within the first 100 cm of the solum. These soils have usually developed on recently deposited sandy materials such as alluvium or dunes. Sandy soils are often characterised by weak or absent structure, low or negligible humus content, poor water retention properties, high permeability, and high susceptibility to compaction with many negative consequences (Bruand et al. 2005). Therefore, sandy soils represent an extreme environment for tree growth. Over time, forestry practice has shown that the most suitable tree species for such sites is Scots pine [*Pinus sylvestris* (L.)]. Pine is a light-demanding pioneer

tree species with a marked tolerance to nutrient-poor and dry soils (Diers et al. 2021). The species offers several advantages, including (i) tolerance to a wide range of climatic and edaphic conditions, (ii) stabilisation of eolian sands, and (iii) suitability for production and market demands (Zachar 1965).

Scots pine is typically cultivated in monocultures on these sites and harvested by clear-cutting in alternate years (100–130 years). After clear-cutting, intensive site preparation techniques are often employed to prepare the site for planting, including mechanical preparation (deep ploughing) and removal of logging residues. Ploughing helps eliminate unwanted weeds, sedges, and may-beetle

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(*Melolontha melolontha*) (Moffat et al. 2011). An alternative site preparation method involves incorporating logging residues into the mineral soil using various techniques (Konôpka et al. 2012). This practice can not only increase soil organic matter content but also moderate soil temperature, enhance soil structure, improve water retention and provide protection from erosion (Sanchez et al. 2009).

Harvesting regimes and site preparation techniques used in forest ecosystems can have a significant impact on soil organic matter and nutrient budgets (Pérez-Batallón et al. 2001). Soil properties, vegetation, and soil microbiota influence each other and interact in complex ways (Jing et al. 2014; Wu et al. 2017). Vegetation absorbs nutrients from the soil transformed by microbes and returns them to the soil via litter and root exudates. Soil properties, in turn, influence the rate of microbial decomposition and transformation, affecting plant growth (Hou et al. 2021). If the logging residues are removed, the nutrient balance can be significantly disrupted, potentially leading to habitat degradation, especially in low-trophic environments (Novotný et al. 2011).

Incorporating logging residues into forest soils has been shown to increase soil carbon and nutrient stocks, improve seedling survival, and enhance tree growth (Sanchez et al. 2003). However, these findings are based on early stand establishment, and it remains unclear whether these effects persist in the long term. Therefore, the main objective of this study was to assess whether and how mechanical site preparation techniques affect the physico-chemical and microbial properties of soils in pine plantations on sandy soils in the long term. Currently, 25 years after the application of alternative management practice involving the incorporation of logging residues into sandy soils in the Záhorská nížina lowland (Slovakia), positive changes in the growth of pine stands are evident. We hypothesise that improved tree growth and higher stand density have led to increased organic matter input to the soil, influencing soil properties, particularly humus content and microbial communities, even longer time after the stand establishment.

## MATERIAL AND METHODS

**Site description.** This study was conducted in the Záhorská nížina lowland, which is characterised by a moderately warm and humid climate with an average annual precipitation of 630 mm, an av-

erage annual temperature of 9.5 °C, and an average growing season temperature of 15 °C. The main soil type in this area is Regosol.

The study sites were located in the Scotch pine [*Pinus sylvestris* (L.)] plantations, which represent the typical and widespread plantations in the Záhorská nížina lowland. Two sites with different techniques of site preparation were selected:

- (i) Site with whole tree harvesting and removal of logging residues and forest floor (LRR), representing traditional management; after stump removal, the surface was ploughed to a depth of approximately 60 cm (48°37'47.80"N, 17°16'1.69"E).
- (ii) Site with mechanical incorporation of logging residues and forest floor into the upper 25 cm of the mineral soil (LRI) (48°37'48.79"N, 17°15'55.11"E).

The pine stands at both sites were of the same age (25 years) but differed significantly in their height and canopy density (Figure 1) and in the presence and thickness of the surface organic layer (the O-horizon). The LRR site had poorer seedling establishment, lower tree height and canopy density, lacked the herb layer, and the O-horizon reached a maximum thickness of 2 cm but was absent in some places. The LRI site exhibited higher tree height and canopy density, the presence of the O-horizon (locally up to 4 cm), and the herb layer, especially mosses.

**Soil sampling and analyses.** Soil sampling was performed in September 2020. In each stand, representing a different site preparation technique, soil samples were collected at five sampling points at 10 m intervals along the transect. Samples were taken from the soil profiles in 10 cm intervals up to a depth of 30 cm. The O-horizon was not included. In the laboratory, soil samples were divided into two parts. The first part of each sample was air-dried, sieved through a 2 mm sieve to remove larger organic particles, and used for the quantification of physico-chemical properties. The second part of the sample used for microbial analyses was stored at 4 °C in field-moist condition until laboratory analyses were performed. These soil samples were not sieved to preserve natural conditions for microbiota.

Soil moisture and dry weight were determined gravimetrically by oven-drying soil samples at 105 °C for 24 h. Soil pH was measured potentiometrically in 1 M CaCl<sub>2</sub> suspension after 24 h. Total carbon (C) and nitrogen (N) contents and

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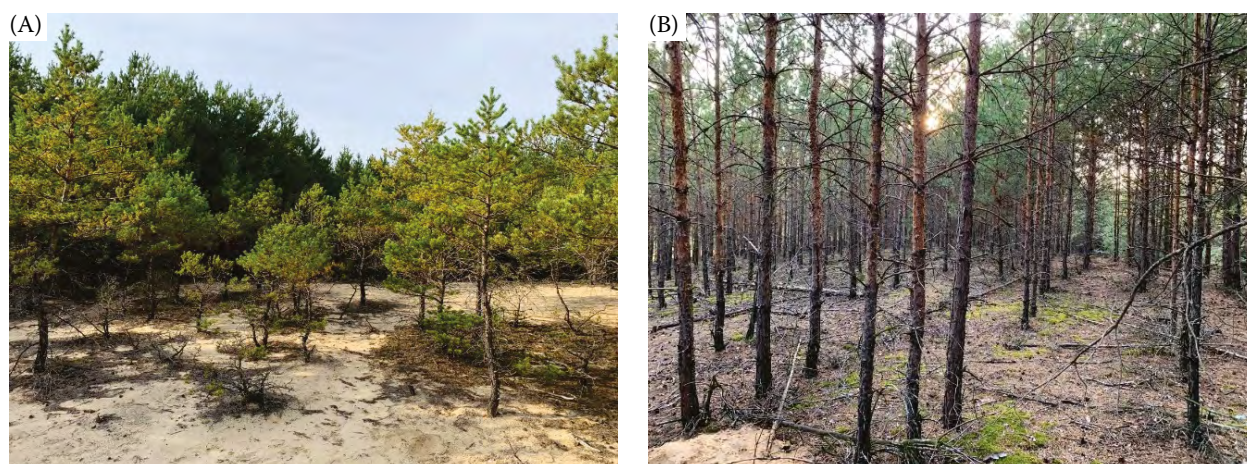


Figure 1. Forest stand after 25 years (A) where logging residues were removed and (B) where logging residues were incorporated into the soil

the C:N ratio were measured using the Vario MACRO elemental analyser (CNS version; Elementar, Germany), which employs the dry combustion method. Since the soils are acidic and do not contain any carbonates, the total carbon represents the organic carbon. Plant-available phosphorus was determined by the Bray-Kurtz method (Bray, Kurtz 1945) and exchangeable calcium (Ca), magnesium (Mg), and potassium (K) were determined in the  $\text{NH}_4\text{Cl}$  extract by atomic absorption spectrometry (AAS).

Microbial biomass carbon ( $C_{\text{mic}}$ ) was assessed using the microwave-irradiation procedure following Islam and Weil (1998). N-mineralisation (N-min) was determined using the laboratory anaerobic incubation procedure described by Kandeler (1993). Basal soil respiration ( $BR$ ) was quantified according to Isermeyer (Alef 1991). Catalase activity ( $Catal$ ) was evaluated using the method described by Khaziev (1976). Community-level physiological profiles (CLPPs) of microbial communities were determined in two ways: employing BIOLOG<sup>®</sup> EcoPlates (Biolog Inc., USA) (Insam 1997) and using a rapid microtiter-plate method measuring the evolved  $\text{CO}_2$  using the whole soil (Campbell et al. 2003).

**Data evaluation.** All values of microbial characteristics were converted per unit of the dry matter of the soil.

The richness of the soil microbial community was assessed as the number of wells with a positive response (violet-coloured wells) observed following incubation, i.e. the number of different substrates used by the microbial community. To assess the

alpha diversity ( $Div$ ) of soil microbial functional groups based on the Biolog<sup>®</sup> approach, we used Hill's index (Hill 1973), see Equation (1):

$$Div = \frac{1}{\sum_i p_i^2} \quad (1)$$

where:

$p_i$  – frequency (relative abundance) of the  $i^{\text{th}}$  functional group, calculated as the ratio of the activity on a particular substrate to the sum of the activities on all substrates.

Basic statistical descriptions (means, standard deviations) were calculated for each soil characteristic within each site with a different preparation technique. Differences in physico-chemical characteristics and parameters of the soil biological activity among individual forest management practices were tested by two-way analyses of variance. Pairwise differences were subsequently tested by Tukey's post-hoc test. The software package Statistica (Version 12.0, 2018) was used for all calculations.

## RESULTS AND DISCUSSION

According to a two-way analysis of variance (ANOVA), site preparation had a more pronounced effect on soil properties than the depth of sampling (Table 1). However, some interactions were also significant. Different site preparations significantly affected the soil acidity, C and N content, C:N ratio ( $P < 0.001$ ), and soil moisture. Among soil microbial characteristics, significant differences were

Table 1. Analysis of variance of soil physico-chemical and microbial properties including community-level physiological profiles of microbial communities (*F*-tests)

Factor	Depth		Management		Depth × management	
	<i>F</i> -test	<i>P</i> -value	<i>F</i> -test	<i>P</i> -value	<i>F</i> -test	<i>P</i> -value
<b>Physico-chemical soil properties</b>						
Soil moisture	0.31	n.s.	9.96	**	1.40	n.s.
Soil pH	1.27	n.s.	39.48	***	1.07	n.s.
C	8.20	**	26.40	***	6.62	**
N	9.64	***	27.63	***	6.63	**
C:N	7.43	**	77.97	***	3.65	*
Ca	0.36	n.s.	2.56	n.s.	3.40	n.s.
Mg	0.33	n.s.	1.37	n.s.	0.55	n.s.
K	1.60	n.s.	2.05	n.s.	0.20	n.s.
P	0.22	n.s.	4.36	*	1.13	n.s.
<b>Microbial soil properties</b>						
Catalase activity	12.45	***	13.14	**	4.75	*
Basal respiration	3.71	*	0.29	n.s.	1.55	n.s.
Microbial biomass carbon	8.73	**	14.76	***	8.24	**
N-mineralisation	6.19	**	10.31	**	6.69	**
Diversity index	0.50	n.s.	6.07	*	2.91	n.s.
Richness of functional groups	1.16	n.s.	–	n.s.	3.00	n.s.
<b>Utilisation of substrates by microbial communities (MicroResp™ method)</b>						
α-ketoglutaric acid	2.29	n.s.	6.94	*	0.93	n.s.
Arginine	0.61	n.s.	1.56	n.s.	0.00	n.s.
Asparagine	0.17	n.s.	7.46	*	0.19	n.s.
Cellulose	0.04	n.s.	0.14	n.s.	0.04	n.s.
Malic acid	3.10	n.s.	2.95	n.s.	0.16	n.s.
Methylglucamine	0.86	n.s.	0.05	n.s.	0.27	n.s.
Phenylalanine	7.68	n.s.	14.62	***	4.11	n.s.
Serine	1.34	n.s.	0.35	n.s.	0.35	n.s.
Mannose	0.30	n.s.	0.52	n.s.	0.06	n.s.
Glutamine	0.98	n.s.	0.68	n.s.	1.40	n.s.
Malonic acid	0.31	n.s.	4.33	*	0.03	n.s.
Xylose	0.61	n.s.	2.41	n.s.	1.23	n.s.

\**P* < 0.05; \*\**P* < 0.01; \*\*\**P* < 0.001; n.s. – non-significant

found in the diversity index (*P* < 0.001), N-mineralisation (*P* < 0.01), catalase activity, and microbial biomass carbon (*P* < 0.05). On the other hand, non-significant differences were in Ca, Mg, K content, basal respiration, and richness (Table 1). The different site preparations significantly affected the utilisation of some substrates, especially that of phenylalanine.

Surprisingly, samples from different depths differed significantly in physico-chemical properties

only in the C and N content and C:N ratio. Significant differences were found in most of the microbial characteristics except the diversity index and richness. On the other hand, no significant differences were observed in the utilisation of substrates.

Most of the soil properties reached higher values in the first 10 cm of the soil profile than in the depths of 10–20 cm and 20–30 cm (except soil reaction, amount of Ca, Mg, K, basal respiration, diversity index and functional group richness) (data not

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shown). In the case of factor interaction, the main differences were in the first 10 cm and the differences decreased with increasing depth (Figure 2).

As previously mentioned, significant differences were observed in the soil physico-chemical properties between the LRR and LRI stands. Specifi-

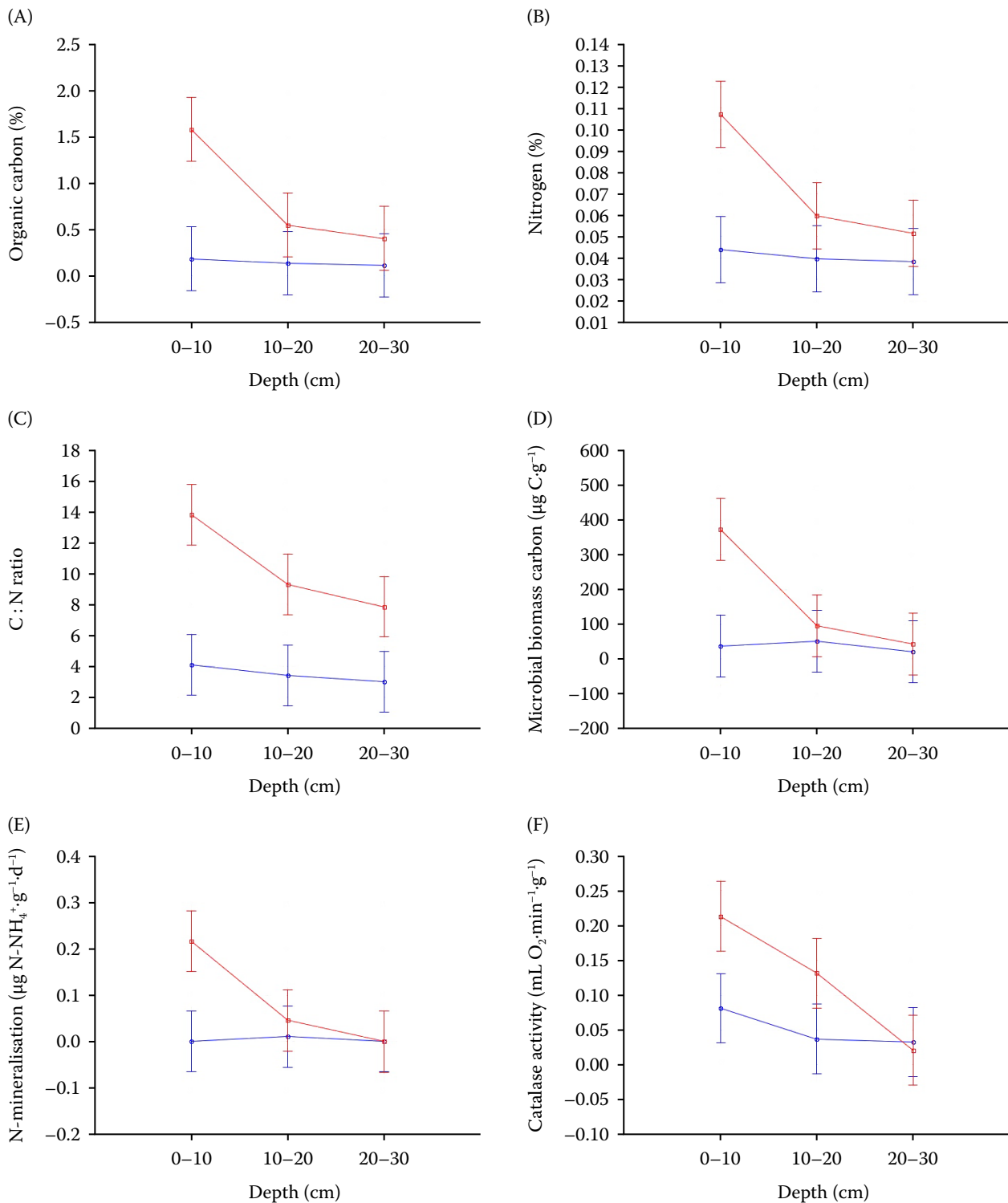


Figure 2. Means and standard deviations of (A) carbon content, (B) nitrogen content, (C) C:N ratio, (D) microbial biomass carbon, (E) N-mineralisation, and (F) catalase activity at plots with different treatment and in different soil depths. Red line – LRR (removal of logging residues) treatment; blue line – LRI (incorporation of logging residues) treatment.

cally, soil moisture, the content of C, N, and the C:N ratio increased significantly at the plot with the incorporation of logging residues into the topsoil ( $P < 0.05$ ). In contrast, soil pH and P content decreased there significantly, while no significant differences were observed between the two site preparation techniques for Ca, Mg, and K contents (Table 2). Sanchez et al. (2003) demonstrated that incorporating forest slash during site preparation effectively increases soil C and nutrients, but their findings were based on early stand establishment. Our results, however, show that the organic C content at the LRI plot was five times higher than at the site using the traditional site preparation method even 25 years after the treatment.

It is well known that soil organic matter performs several functions, including improving water retention. Thus, the higher soil moisture at the LRI plot is consistent with the increase in organic carbon. Moreover, a continuous surface organic layer was present at the LRI plot, which likely acts as an insulating layer, reducing soil evaporation (Pérez-Batallón et al. 2001). We suppose that the incorporation of logging residues, which improves water retention, led to better tree establishment and growth. Over time, this enhanced tree growth likely contributed to increased litterfall on the soil surface. In 30-year-old Scots pine stands, annual litterfall can vary and reach 2–8 Mg·ha<sup>-1</sup> (Kacálek et al. 2018). Pine litter was found to be nutrient-poor (Johansson 1995), and coniferous litter is generally more acidic than that of deciduous

trees (Burgess-Conforti et al. 2019). As the cation exchange capacity of sandy soils is very low, it is not surprising that soil acidity increased at the LRI plot. Similar findings of increased organic carbon and soil acidity with stand age, likely due to increasing litter input, were reported by Šurda et al. (2021). Novotný et al. (2011) observed that the initial release of elements from milled material can positively affect soil pH, increasing its values. However, acidification tends to increase over time probably due to the large carbon amount in the milled material, especially wood. The combined effects of tree uptake and increased acidity may also explain the observed decrease in phosphorus content. At the LRI plot, phosphorus could be largely taken up by trees or immobilised, as acidic conditions reduce phosphorus mobility by fixing it in insoluble compounds (Ouro et al. 2001). Further research is required to confirm this assumption.

The analysis of soil microbial characteristics as potential indicators of site preparation techniques revealed a significant increase in microbial biomass carbon, N-mineralisation, and catalase activity in the plot with the incorporation of logging residues into the topsoil ( $P < 0.05$ ). Similar effects have been observed in other forest plantations (Pérez-Batallón et al. 2001; Karlsson, Tamminen 2013; Węgiel et al. 2023) and are likely due to increased carbon availability and higher moisture, both of which promote microbial population growth. In contrast, the removal of logging residues impacts soil C pools by reducing the formation of soil organic matter, as stumps and coarse roots are removed, and the topsoil is mixed with subsoil (Mäkipää et al. 2023). Foote et al. (2015) found that forest harvesting practices that removed more than just the tree bole significantly reduced total soil nitrogen, microbial biomass carbon and nitrogen in loblolly pine forests. These effects were still evident 15 years after the intervention. The lower microbial biomass carbon observed after the removal of logging residues can be attributed to reduced C availability and lower soil moisture content. This finding aligns with other studies (Hendrickson et al. 1985; Ross et al. 1995), which also reported reduced soil microbial C in whole-tree harvested stands.

A significant decrease in the microbial diversity index based on the Biolog® method was observed, while the richness of the functional groups remained unaffected (Table 3). However, the dif-

Table 2. Tukey's pairwise post-hoc test of the differences of means  $\pm$  SD between plots for physico-chemical properties

Site treatment	LRR	LRI
Soil moisture (%)	1.31 $\pm$ 0.82 <sup>b</sup>	2.93 $\pm$ 2.66 <sup>a</sup>
pH	4.77 $\pm$ 0.10 <sup>a</sup>	4.06 $\pm$ 0.43 <sup>b</sup>
C (%)	0.15 $\pm$ 0.07 <sup>a</sup>	0.85 $\pm$ 0.73 <sup>a</sup>
N (%)	0.04 $\pm$ 0.01 <sup>b</sup>	0.07 $\pm$ 0.03 <sup>a</sup>
C:N	3.50 $\pm$ 1.24 <sup>b</sup>	10.32 $\pm$ 3.63 <sup>a</sup>
Ca (mg·kg <sup>-1</sup> )	369.66 $\pm$ 39.48 <sup>a</sup>	395.34 $\pm$ 52.91 <sup>a</sup>
Mg (mg·kg <sup>-1</sup> )	76.21 $\pm$ 8.98 <sup>a</sup>	79.65 $\pm$ 6.21 <sup>a</sup>
K (mg·kg <sup>-1</sup> )	15.04 $\pm$ 4.80 <sup>a</sup>	17.34 $\pm$ 3.89 <sup>a</sup>
P (mg·kg <sup>-1</sup> )	15.00 $\pm$ 8.72 <sup>a</sup>	8.37 $\pm$ 6.21 <sup>b</sup>

<sup>a, b</sup> Plots with the same letters do not differ significantly; SD – standard deviation; LRR – removal of logging residues; LRI – incorporation of logging residues

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Table 3. Tukey's pairwise post-hoc test of the differences of means  $\pm$  SD between plots for microbial properties

Site treatment	LRR	LRI
Basal respiration ( $\mu\text{g CO}_2\cdot\text{g}^{-1}\cdot\text{h}^{-1}$ )	$0.05 \pm 0.03^a$	$0.04 \pm 0.03^a$
Catalase activity ( $\text{mL O}_2\cdot\text{min}^{-1}\cdot\text{g}^{-1}$ )	$0.05 \pm 0.04^b$	$0.12 \pm 0.10^a$
Microbial biomass carbon ( $\mu\text{g C}\cdot\text{g}^{-1}$ )	$36.00 \pm 25.04^b$	$171.25 \pm 194.26^a$
N-mineralisation ( $\mu\text{g N-NH}_4^+\cdot\text{g}^{-1}\cdot\text{d}^{-1}$ )	$0.00 \pm 0.01^b$	$0.09 \pm 0.13^a$
Diversity index	$13.55 \pm 1.55^a$	$12.09 \pm 1.85^b$
Richness of functional groups	$21.20 \pm 1.21^a$	$21.20 \pm 1.52^a$
<b>Microbial utilisation of:</b>		
Arginine	$0.14 \pm 0.01^b$	$0.15 \pm 0.01^a$
Asparagine	$1.06 \pm 0.97^a$	$1.58 \pm 1.12^a$
$\alpha$ -ketoglutaric acid	$1.85 \pm 0.77^b$	$5.84 \pm 5.08^a$
Cellulose	$1.40 \pm 0.43^a$	$1.34 \pm 0.44^a$
Xylose	$1.03 \pm 0.81^a$	$1.60 \pm 1.00^a$
Mannose	$0.95 \pm 0.82^a$	$1.03 \pm 0.83^a$
Malic acid	$1.45 \pm 0.49^b$	$3.17 \pm 1.72^a$
Methylglucamine	$0.14 \pm 0.01^a$	$0.14 \pm 0.01^a$
Phenylalanine	$3.52 \pm 2.32^a$	$2.97 \pm 1.34^a$
Serine	$2.94 \pm 1.29^a$	$2.53 \pm 1.43^a$
Glutamine	$2.19 \pm 1.80^b$	$3.65 \pm 1.68^a$
Malonic acid	$1.44 \pm 1.19^a$	$2.51 \pm 2.26^a$

<sup>a, b</sup> Plots with the same letters do not differ significantly; SD – standard deviation; LRR – removal of logging residues; LRI – incorporation of logging residues

ference in the diversity was minor (diversity index 13.55 at LRR vs 12.09 at LRI, respectively). Both basal respiration and utilisation of individual carbon sources by microorganisms were generally low, consistent with sandy soil properties, such as higher acidity, low humus content and soil moisture. Nevertheless, our results demonstrate that LRI had a considerable effect on the utilisation of several substrates compared to the traditional site preparation technique. An analysis of the microbial functional group composition using the MicroResp<sup>TM</sup> assay revealed a significant increase in the utilisation of malic acid, glutamine,  $\alpha$ -ketoglutaric acid and arginine when logging residues were incorporated into the topsoil ( $P < 0.05$ ). The community structure based on the Biolog<sup>®</sup> method also showed differences in the utilisation of two substrates. The utilisation of  $\alpha$ -ketobutyric acid and  $\alpha$ -cyclodextrin was significantly higher at the LRI plot compared to the LRR plot ( $P < 0.05$ ; data not shown). The higher utilisation of different carbon sources found after incorporating logging residues into the topsoil can be explained by higher C availability and soil mois-

ture content. Overall, research on the incorporation of logging residues into forest soils is limited, as this treatment has only recently become more widely accepted in forestry practices. Nevertheless, our results indicate that different site preparation techniques can lead to slight changes in the microbial community activity and structure.

## CONCLUSION

Site preparation generally aims to enhance tree regeneration by promoting seedling survival and growth. Incorporating logging residues is a recommended practice in forest management, offering significant benefits, including increased nutrient supply and gradual release over time, increased organic matter content in the area, and improved water retention. This method may be particularly effective for soils prone to degradation and nutrient loss, such as sandy soils, which are poor in soil organic matter and nutrients, and have low water holding capacity, i.e. conditions that hinder plant nutrient uptake and water availability. Applying this technique to vulnerable soils is expected to sta-

bilise or even improve their properties. Our results showed that the post-harvest management of logging residues can significantly influence soil properties even 25 years after the treatment and indicate that this technique is associated with an improvement in the soil C pool and water content, as well as alterations in microbial activity and biomass involved in the organic matter decomposition and nutrient release. On the other hand, this site preparation technique led to a decrease in soil pH and phosphorus concentration, which could potentially hinder tree growth over time. An important question remains whether the addition of deciduous trees, such as oak, into pine stands could help mitigate soil acidity and enhance nutrient availability. Therefore, further research is needed to explore the relationships between tree growth and soil properties in such extreme environments.

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