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Article

Significant Differences in Microbial Soil Properties, Stoichiometry, and Tree Growth Occurred within 15 Years after Afforestation on Different Parent Material

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Abstract: Soil formation results from the weathering and alteration of bedrock, influenced by climatic conditions and biotic factors, including living organisms. The mineralogical composition of the parent material, along with plant species and soil microorganisms, forms the foundational elements of an ecosystem's energy cycle. Afforestation in arid-semi arid regions plays a crucial role in preventing erosion and enhancing soil quality, offering significant economic and ecological benefits. This study assessed the impact of parent material diversity on soil physicochemical properties, microbial basal respiration (MR), and microbiological characteristics, as well as their subsequent effects on plant growth, 15 years post-afforestation. For this purpose, various soil physicochemical parameters, MR, soil microbial biomass carbon (C_{mic}), stoichiometry (microbial quotient = $C_{mic}/C_{org} = q_{Mic}$ and metabolic quotient = $MR/C_{mic} = q_{CO2}$), and tree growth metrics such as height and diameter were measured. The results of the one-way ANOVA indicated that afforested soils on limestone bedrock exhibited significantly better physicochemical and microbiological properties, as well as tree growth parameters, compared to those on andesite. Notably, sensitive microbial properties such as C_{mic} , MR, and q_{Mic} were positively influenced by afforestation. The highest C_{mic} (323 g C kg⁻¹) and MR (1.3 CO₂-C g⁻¹ h⁻¹) values were recorded in limestone-derived soils. Currently, the measured properties of soils can be ranked as LS>AS>LC>AC. Conversely, the highest q_{CO2} values were observed in the control plots of andesite parent material (7.22). This study highlights the importance of selecting appropriate parent material and understanding soil characteristics to optimize afforestation efforts in bare areas. As a result, the higher plant growth capacity and carbon retention of limestone soil indicate that it has high microbial biomass and microbial activity (q_{Mic}).

Keywords: parent material; afforestation; scots pine; microbial biomass C; microbial respiration; plant growth

1. Introduction

Soil is formed under the influence of bedrock, climate, vegetation, and topographic factors. However, the dominance of one factor over the others can lead to variations in soil properties and land productivity [1]. Semiarid and arid ecosystems, for example, are primarily influenced by parent material and the continental climate, with summer drought being a critical stress factor [2,3]. The physical properties, mineral composition, and texture of the parent material significantly affect the soil formation rate and the characteristics of the resulting soil [4]. The hardness of rocks is also crucial; soils formed on hard rocks have lower formation rates than the rate of soil loss through erosion, leading to poor habitat characteristics. Birkeland [5] ranked bedrock stability against soil formation factors as follows: quartzite, chert > granite, basalt > sandstone, siltstone > dolomite, limestone.

The parent material contributes to the nutrient cycle by releasing nutrients through weathering and maintaining a balance between nutrient loss and storage [1,4]. After climate, plant and microorganism activities play the most important roles in soil formation. Trees, in particular, influence parent material through biomechanical, chemical, and biological effects [1,3]. Traditionally, studies have focused on the biochemical, ecological, and edaphic influences on soils, but the significance of biomechanical effects is increasingly recognized [6–8]. Most research on the

biomechanical effects of vegetation has centered on tree root growth, but trees also impact the physical displacement of regolith materials and the formation of root debris and exudates [9]. Plant roots mechanically break down rocks during growth, accelerating soil formation processes. Additionally, root exudates and mechanical root activities help reduce erosion, stabilizing soil ecosystems [10]. Organic acids and exudates produced by plants and microorganisms enhance the chemical weathering of minerals and the release of nutrients [11].

Over the last 20 years, the Turkish Ministry of Agriculture and Forestry has conducted afforestation activities to increase forest cover to 30% of Turkey's surface area. Scots pine and black locust are commonly used for erosion control due to their high tolerance to drought and poor soil conditions [12]. Planting native tree species has accelerated natural succession [13] and carbon sequestration [14]. Plants, known as the organisms with the largest biomass in terrestrial ecosystems, contribute greatly to the formation and development of soils through litter residues, leaves, branches, roots, and exudates of their aboveground biomass [15,16]. In addition, vegetation affects the organic carbon (OC) concentration added to soils and the diversity, quality, and quantity of soil microorganisms [17]. For example, the biomass of microbial organisms in soils converted from forest to agriculture decreases significantly [18,19]. Similarly, agricultural land contains less soil organic C (SOC) and C_{mic} compared to native forests [20]. Vegetation type strongly influences soil-forming processes, contributing significantly to organic matter and microbial community development and activity [21,22]. Microbial stoichiometric indices such as microbial basal respiration (MR), microbial quotient (q_{Mic}), metabolic quotient (qCO_2), and C mineralization are related to the SOC cycle in the environment. With the help of these stoichiometric indices, the effects of changes in land use or ecosystem on soil quality, microbial populations, and microbial activity rates can be estimated [23,24].

While many studies suggest that microbial biomass is controlled by soil physicochemical properties [25,26], others indicate that aboveground vegetation type has a stronger influence on microbial biomass and activity [27,28]. Kara et al. [10] found positive effects of ecosystem changes on microbial biomass and activity in afforested areas in both the short and long term. Research on afforested ecosystems shows that vegetation enhances soil-forming processes, leading to organic matter accumulation and microbial community development [29–31]. Trees provide nutrients to the soil through dead plant residues and root secretions, which microorganisms break down and mineralize, forming a usable nutrient source for plants [32]. Microbial organisms decompose this litter, returning essential organic nutrients to the soil. This process enriches the growing environment, enhancing the productivity of the soil. As a result, plant growth parameters, such as tree height (TH) and diameter at breast height (DBH), improve significantly. The upper soil horizon, a reservoir for soil microbial biomass, is crucial for decomposition and nutrient cycling [33]. This symbiotic relationship underpins the nutrient cycle, essential for ecosystem health.

Soil microorganisms are the most rapidly affected by environmental conditions, various forestry management practices, and sudden changes on or within the soil surface. Consequently, measuring microbial activities and properties following forestry management activities is essential for monitoring changes in soil fertility. Changes in microbial properties can provide early indications of potential ecosystem disruptions [32,34–37]. Assessing microbial biomass in soils is crucial in many soil monitoring studies and programs [35], but it should be considered alongside other habitat parameters for a comprehensive ecological assessment. Although recent studies focus on the effects of soil microbiological properties on soil fertility, there is still limited information on their relationship with tree growth. This research aimed to new perspectives on the effects of physicochemical, microbial properties, and stoichiometry of soils derived from different bedrocks on seedling growth parameters, highlighting the importance of evaluating soil quality parameters in forests.

2. Materials and Methods

2.1. Study Sites

The study was conducted in Gümüşhane Province, located in the Northeast Black Sea mountainous region of Turkey (40°19'–40°23' N and 39°30'–39°38' E). The elevation ranges from 1,400 to 1,600 meters, with steep slopes (45–50%) covered by shallow soil (typically 50 cm or less) on a west-southwest aspect. The region has a "Warm, dry-summer continental climate" (Dsb) according to the Köppen-Geiger climate classification (www.koeppen-geiger.vu-wien.ac.at/). Weather data, including long-term averages (1990–2018) for monthly air temperature and precipitation in Gümüşhane Province, were obtained from the Turkish State Meteorological Service (www.mgm.gov.tr). The annual mean temperature is 9.4°C, with a high of 39.5°C in July and a low of –19.0°C in February. The topsoil temperature drops below 0°C and freezes during the winter. Annual precipitation is 461 mm, with about 54% occurring during the growing season between April and September. The main soil type in the area is Entisol, characterized by minimal horizon development. The rugged topography consists of andesitic and limestone bedrock. Table 1 provides a detailed characterization of the study site.

Table 1. Study site characteristics.

Environmental Factors	Limestone		Andesite	
	Sample	Control	Sample	Control
Longitude	39° 33' 19"	39° 33' 46"	39° 33' 43"	39° 33' 23"
Latitude	40° 21' 08"	40° 21' 23"	40° 21' 20"	40° 21' 07"
Altitude mean (m)	1480	1520	1500	1450
Slope degree mean (%)	50	40	45	50
Afforestation date	2005	-	2005	-
Vegetation type	Scotch Pine	Bare space	Scotch Pine	Bare space
Stand canopy (%)	70-100	-	70-100	-
Soil texture	Loamy clay	Clay loam	Sandy clay loam	Sandy loam
pH class	Moderately	Slightly	Moderately	Slightly
	Alkaline	Alkaline	Alkaline	Alkaline

2.2. Experimental Design, Soil Sampling, and Pretreatment

This study was conducted in the spring of 2019 at Scotch pine (*Pinus sylvestris* L.) afforestation sites, planted over 30 hectares in 2005 by the General Directorate of Afforestation of Türkiye. The sites were initially prepared using an excavator, and irrigation and fertilization were applied during the first five years after planting. The control area was significantly degraded grassland with less than 5% vegetation cover, exposed to erosion and not fertilized. A grid-based sampling design was used to select three 10x10 m² experimental areas from different bedrocks, representing a total of 30 hectares. Tree growth parameters, such as tree height (TH) and diameter of breast height (DBH), were directly measured within each 100 m² area using a Blume-Leiss and compass, respectively. Soil samples were taken at the beginning of the vegetation period in the Scots pine forest, with the bare land used as a control. A total of 40 sample points were systematically selected from two different parent materials afforested and control areas (LS= limestone sample; LC= limestone control; AS= Andesite sample; AC= Andesite control). Before collecting disturbed mineral soil samples, the litter, root debris, and stones were removed, and stored in plastic bags. In the field study, microbial soil samples were first sieved using <2 mm sieve, then distilled water was added until 50-60% moisture content was reached and quickly transferred to the laboratory for storage in a 4°C refrigerator before

analysis. The disturbed and microbial soil samples were collected from the topsoil layer at 0-20 cm depth. Other soil samples were dried by spreading them in drying cabinets to make them air-dried, after drying the soils were ground and sieved (<2 mm) to determine the physicochemical parameters of the soil.

2.3. Physical and Chemical Properties of Soils

Soil moisture content was determined gravimetrically by oven-drying samples at 105°C until a stable weight was achieved. Soil particle size (PS) was measured using the hydrometer method [38]. Soil pH and electrical conductivity (EC) were assessed using a 1:2.5 soil/water suspension for pH and a 1:5 soil/water suspension for EC, measured with appropriate meters. Soil organic carbon (SOC) was estimated using the potassium dichromate oxidation method [39], total nitrogen by the Kjeldahl method [40], while total lime (CaCO_3) content was determined using the Scheibler calcimeter method [41].

2.4. Microbial Biomass C (C_{mic})

The chloroform-fumigation-extraction method was performed to determine Soil microbial biomass carbon (C_{mic}) [42]. Each soil samples were divided into 2 portions and the first half of the soils stored for microbial analyses were fumigated with ethanol-free chloroform at 25°C for 24 h in a vacuumed desiccator, while the second portion of samples was not fumigated. Both fumigated and non-fumigated soil samples were mixed in 0.5 M K_2SO_4 (1:4 w/v), followed by shaking the suspension for 30 minutes at 200 rpm in an oscillator. The extracts were filtered through the Whatman-42 filter paper and stored before titration. Total organic carbon in both fumigated and non-fumigated extracts was estimated as specified by the Walkley Black method [43,44]. C_{mic} was calculated from the difference in extractable organic C between fumigated and unfumigated soil samples using the formula: biomass C = $2.64 \times \text{EC}$, where EC refers to the difference in extractable organic C between the treatments; 2.64 is the proportionality factor for biomass C released by fumigation extraction [42,44]. The microbial quotient (C_{mic}/C_{org} or q_{Mic}) was calculated by expressing microbial C as a percentage of total soil organic C.

2.5. Microbial Respiration

Microbial respiration was determined using the absorption method to quantify carbon dioxide (CO_2) [45]. Moist soil samples (50 g dry weight equivalent) were placed in 60-mL beakers and incubated in the dark at 25°C in 500 mL airtight, sealed jars along with 10 mL 0.1 M NaOH. After 7-day incubation, the CO_2 generated was measured by titration of the excess NaOH with 0.05 M HCl [46]. An empty jar without soil was used as the control. The difference in the volume of HCl consumed between the treatment and the control was used to calculate CO_2 emission from soil microorganisms. The metabolic quotient q_{CO_2} is an important parameter that can be used to estimate soil quality and stress response. It was calculated as the microbial respiration rate ($\mu\text{g CO}_2\text{-C h}^{-1}$) per mg of C_{mic} [45].

2.6. Statistical Analyses

The data were tested for normality and homogeneity of distribution using skewness, kurtosis, and Shapiro–Wilk tests. Parameters that deviated from normal distribution were log-transformed ($\log_{10}$) before analyzing variance (ANOVA). After confirming that the data met the assumptions for parametric tests, one-way ANOVA was applied to assess the effects of parent material on all parameters, followed by Duncan's multiple range tests ($\alpha = 0.05$). Pearson's linear correlation analysis was also conducted to assess the relationships between soil properties and plant growth characteristics. Discriminant analysis (DA) was employed to analyze the differences between groups of multivariate data using one or more discriminant functions to maximally separate the identified groups. DA was specifically used to differentiate between the tree growths based on soil variables.

All statistical analyses were carried out using the Statistical Package for the Social Sciences (SPSS Inc., Chicago, IL, USA) and figures were generated by using the XLSTAT Windows software package.

3. Results

Under similar ecological conditions, soils formed from different bedrocks exhibited significant differences in physical and chemical properties, including sand, silt, clay content, pH, EC, SOC, TN, and C/N ratio. These variations also extended to microbial activity indicators such as MR, C_{mic} , q_{Mic} , and qCO_2 . Consequently, these differences in soil properties and microbial activity led to significant variations in the growth characteristics of the seedlings planted in the area (Table 2).

Table 2. F statistics of the one-way ANOVA for the effects of parent material on soil physicochemical, microbial properties, and plant growth characteristics.

	<i>San</i>	<i>Sil</i>	<i>Clay</i>	<i>pH</i>	<i>E</i>	<i>SO</i>	<i>T</i>	<i>C/</i>	<i>CaC</i>	<i>Cm</i>	<i>M</i>	<i>qM</i>	<i>qC</i>	<i>DB</i>	<i>T</i>
	<i>d</i>	<i>t</i>	<i>y</i>		<i>C</i>	<i>C</i>	<i>N</i>	<i>N</i>	<i>O₃</i>	<i>ic</i>	<i>R</i>	<i>ic</i>	<i>O₂</i>	<i>H</i>	<i>H</i>
<i>Parent</i>															
<i>Mater</i>	***	**	***	**	**	***	**	**	***	***	**	***	***	***	**
<i>ial</i>		*		*	*		*	*			*				*
<i>F-</i>	355.	42.	117,	60,	9.	21.	48.	24.	123.2	84.3	62.	44.5	27.9	56.	85.
<i>value</i>	8	6	4	5	8	5	8	3			6			0	6
<i>s</i>															

Abbreviations: EC=electrical conductivity (dSm⁻¹); TN=total soil N (%); SOC=soil organic carbon (%); C_{mic} =carbon in the microbial biomass (μg g⁻¹); MR= microbial basal respiration (μg CO₂-C g⁻¹ soil h⁻¹); q_{Mic} = microbial quotient; qCO_2 = metabolic quotient; DBH = diameter at breast high and TH= tree high. a No significant differences are indicated by ns. * Significant at P ≤ 0.05. ** Significant at P ≤ 0.01. *** Significant at P ≤ 0.001.

3.1. Physicochemical Properties of the Soil

The results indicated significant differences in the analyzed properties of soils developed on different parent materials, with notable improvements in soils from post-afforestation environments. The ANOVA test revealed that the physical, chemical, and microbiological properties of these soils were significantly different from each other and the control areas. These differences in soil properties influenced soil fertility, resulting in varying development rates among the seedlings planted in the different areas. These findings are consistent with other studies [1,3,47]. The mean values of some physicochemical properties and plant growth parameters are presented in Table 3, while the microbial indices and microbial basal respiration are depicted in Figures 1 and 2.

According to international soil texture classifications, the soils at the research sites displayed varying textures based on parent material. In the afforestation site, soils derived from limestone were classified as clayey, while those in the limestone control site were also clayey. Soils from basaltic andesite were identified as sandy clayey in the afforestation sites and sandy loam in the control area. The percentages of sand, silt, and clay differed significantly depending on the parent material. Soils from basaltic andesite had significantly higher sand content compared to those from limestone (Table 3). Conversely, the average silt and clay contents were significantly higher in limestone-derived soils (Table 3). In other words, andesite produced coarse-grained soils with significantly higher sand and lower silt and clay percentages. Notably, the afforestation sites exhibited a decrease in sand and dust percentages compared to the control sites, while the clay percentages increased (Table 3).

The mean soil reaction (pH) values and electrical conductivity (EC) of the limestone soils statistically differed from the andesite soils (P < 0.001) (Table 2). The mean pH and EC values in all soil samples including controls were found to be the highest in the control sites on limestone bedrock

(Table 3). However, the lowest pH and EC values were consistently observed in the soils of the areas with significant andesite bedrock. Moreover, the mean pH (8.08 and 8.18) and EC (0.17 and 0.18 dSm⁻¹) in the limestone bedrock sampling site were higher than andesite afforestation and control sites with 7% and 8%; 30%, and 42%, respectively. The average pH and EC values in all soil samples, including controls, were found to be the highest in afforestation sites on limestone bedrock (Table 3). However, the lowest pH and EC values were consistently observed in soils from areas with significant amounts of andesite bedrock. The pH of limestone bedrock was moderately alkaline (8.08–8.18) in afforestation and control sites and slightly alkaline (7.59–7.57) in andesite bedrock afforestation and control sites. Again, the EC values, like the pH in limestone bedrock control and afforestation sites, were higher than those observed in andesite bedrock. EC values were found below the salinity threshold level (0.20 dS cm⁻¹).

As expected, SOC and TN contents differed significantly among the parent materials ($p < 0.0001$). The average SOC contents of the afforestation sites were 1.65% in andesite and 2.40% in limestone, while TN content was 0.10% in andesite and 0.19% in limestone. SOC and TN amounts in afforestation sites increased by 300% in limestone and 333% in andesite compared to control sites. These results indicate that SOC and TN levels were highest in afforestation sites constructed on limestone parent material (Table 3). The highest annual mean C/N ratio was observed in the andesite control site (22.74), followed by the limestone control site (18.76), the andesite afforestation site, and finally, the lowest in the limestone afforestation site (12.61). The mean C/N ratio of the andesite control site was 17%, 29%, and 44% higher than the limestone control, andesite afforestation, and limestone afforestation sites, respectively (Table 3). Additionally, the soil CaCO₃ percentage was significantly higher in soils derived from limestone compared to andesite. After afforestation, this value decreased by 30% (from 32.90% to 23.09%) in limestone parent material and by 36% (from 4.85% to 3.08%) in andesite parent material compared to the control sites.

Table 3. Selected soil characteristics developed from different parent materials at the topsoil depths.

Parent Material					
		Limestone (n = 30)		Andesite (n = 30)	
		Sample	Control	Sample	Control
SOIL PHYSICOCHEMICAL PROPERTIES	<i>Sand (%)</i>	53,50±0,94 ^a	59,74±1,00 ^b	72,58±,13 ^c	75,28±1,55 ^d
	<i>Silt (%)</i>	15,40±2,89 ^b	22,08±0,91 ^c	10,60±1,06 ^a	13,66±0,87 ^b
	<i>Clay (%)</i>	31,10±2,65 ^c	18,18±1,68 ^b	16,82±2,15 ^b	11,02±1,71 ^a
	<i>pH (H₂O)</i>	8,05±0,1 ^b	8,18±0,09 ^b	7,59±0,09 ^a	7,57±0,15 ^a
	<i>EC (mS cm⁻¹)</i>	0,17±0,04 ^b	0,18±0,02 ^b	0,12±0,02 ^a	0,13±0,01 ^a
	<i>SOC (%)</i>	2,40±0,8 ^c	0,80±0,11 ^a	1,65±0,05 ^b	0,61±0,12 ^a
	<i>TN (%)</i>	0,19±0,05 ^c	0,04±0,00 ^a	0,10±0,02 ^b	0,03±0,01 ^a
	<i>C/N</i>	12,61±1,88 ^a	18,76±1,20 ^b	16,22±2,39 ^b	22,74±3,31 ^c
	<i>CaCO₃ (%)</i>	23,09±3,42 ^b	32,90±6,31 ^b	3,08±1,24 ^a	4,85±2,04 ^a
	<i>C_{mic}</i>	323,19±64,96 ^c	60,74±9,03 ^a	183,10±9,31 ^b	35,09±3,65 ^a
	<i>MR</i>	1,3±0,24 ^c	0,34±0,04 ^a	0,61±0,15 ^b	0,25±0,024 ^a
	<i>qMic</i>	1,39±0,20 ^d	0,77±0,12 ^b	1,11±0,08 ^c	0,59±0,09 ^a

	$q\text{CO}_2$	$4,06\pm0,65^a$	$5,71\pm1,23^b$	$3,37\pm0,85^a$	$7,22\pm0,67^c$
GROWTH PROPERTIES	DBH	$6,63\pm0,26^b$	-	$5,54\pm0,38^a$	-
	TH	$10,81\pm0,70^b$	-	$8,68\pm0,19^a$	-

3.2. Soil Microbiological Properties

The significant changes were determined in the C_{mic} values of the areas after afforestation. The highest average C_{mic} values in soil samples were found in the LS site ($323.19\text{ g C kg}^{-1}$), while the lowest was found in the AC site (35.09 g C kg^{-1}) (Figure 2a). Again, The highest average microbial respiration rate ($1.3\text{ }\mu\text{g CO}_2\text{-C g}^{-1}\text{ h}^{-1}$) from different soil samples was found in LS soil, followed by AS ($0.61\text{ }\mu\text{g CO}_2\text{-C g}^{-1}\text{ h}^{-1}$), LC ($0.34\text{ }\mu\text{g CO}_2\text{-C g}^{-1}\text{ h}^{-1}$) and AC ($0.25\text{ }\mu\text{g CO}_2\text{-C g}^{-1}\text{ h}^{-1}$), respectively (Figure 2b). MR and C_{mic} values of soils increased by approximately 530% with afforestation on both bedrocks. The highest mean value of the C_{mic}/C_{org} ($qMic$) ratio was found in the LS site at 1.39, while the lowest value was observed in AC sites at 0.59 (Figure 2c). MR, C_{mic} , and $qMic$ values were ranked from largest to smallest as $LS > AS > LC > AC$. The highest and lowest $q\text{CO}_2$ values of soil samples were recorded in AC (7.22) and AS (3.37) sites, respectively (Figure 2d). The $q\text{CO}_2$ values of control sites were found to be significantly higher than afforested sites. Unlike other microbial indices, the $q\text{CO}_2$ index order is $AC > LC > LS > AS$.

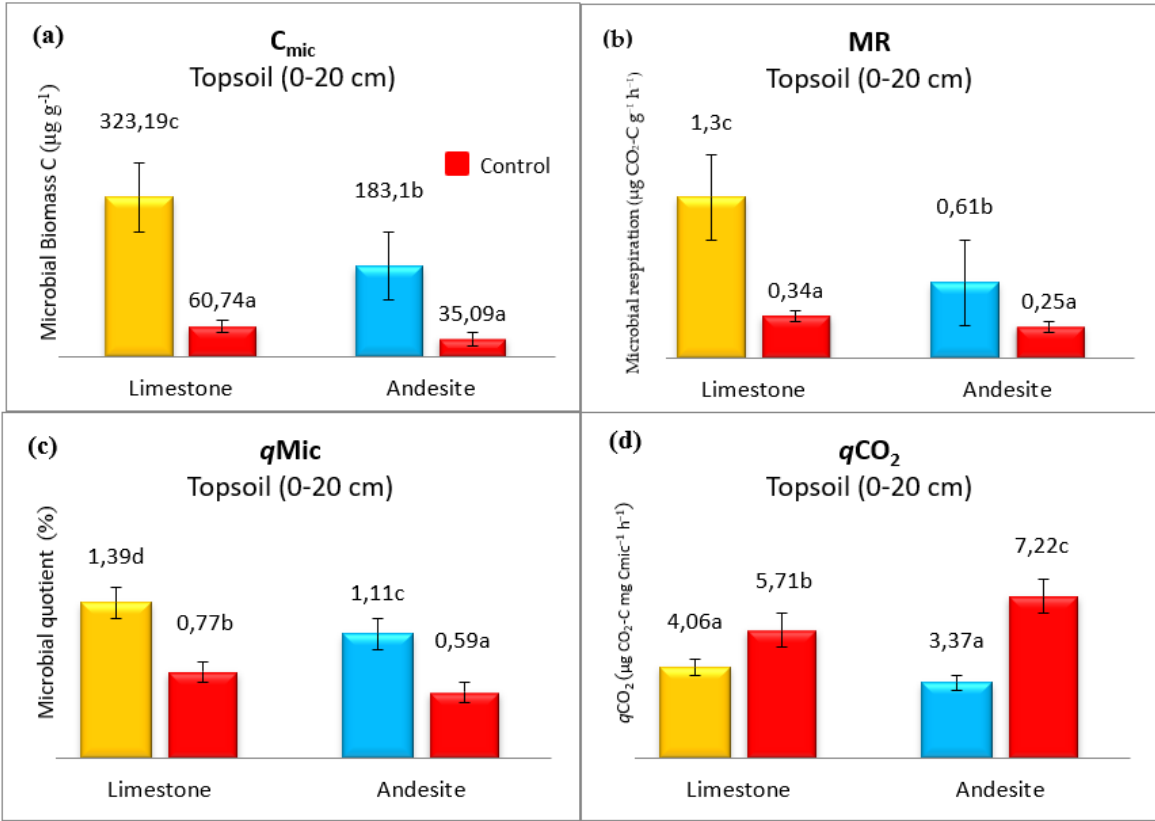


Figure 2. Changes in microbial biomass C (C_{mic}) (a), microbial respiration (MR) (b), Microbial quotient ($C_{mic}/C_{org}=qMic$) (c), and metabolic quotient ($q\text{CO}_2$) (d) with afforestation and difference of parent material. Different letters above the bars indicate significant differences at $p < 0.05$ among the land use types.

3.3. Plant Growth Characteristics

The average height of the saplings on limestone bedrock was 10.81 m, compared to 8.68 m on andesite parent material. Similarly, the average diameter at breast height (DBH) of saplings on

limestone parent material was 6.63 m, while on andesite parent material; it was 5.54 m (Figure 2). These results indicate that, following afforestation, the average height and diameter of saplings on limestone parent material were 20% and 25% higher, respectively, than those on andesite parent material, despite the presence of the same tree species and ecological conditions.

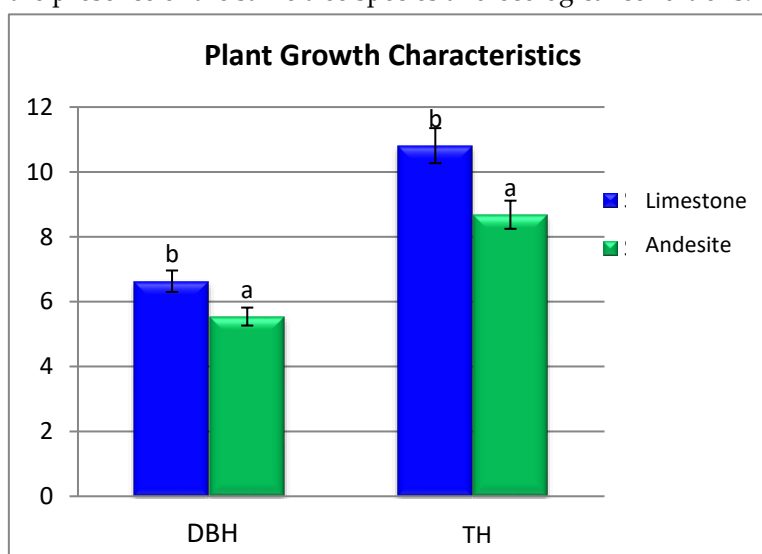


Figure 2. Changes in plant growth characteristics in different parent material sites. Different letters above the bars indicate significant differences at $p < 0.05$ among the land use types. DBH: Breast High Diameter and TH: Tree High.

The Pearson correlation coefficients between TH, DBH, C_{mic} , MR, $qMic$, qCO_2 , SOC, TN, pH, EC, $CaCO_3$, sand, silt, and clay were calculated for both afforested and control areas (Table 3). It was determined that there were positive correlations between TH and dust, clay, pH, EC, SOC, TN, $CaCO_3$, C_{mic} , and DBH ($p \leq 0.001$). The highest positive correlations with TH were shown by clay, $CaCO_3$, DBH, and C_{mic} , respectively. Again, the highest positive correlation values were found between DBH and $CaCO_3$, C_{mic} , MR, and Clay, from largest to smallest, respectively. Here, it was determined that microbial parameters had a positive effect on plant growth parameters at least at the significance level of 0.05. In addition, all of the physicochemical properties of the soils, except sand and C/N ratio, showed positive correlations with plant growth parameters. Other interesting positive correlation values were found between C_{mic} , SOC and TN. On the other hand, the highest negative correlations were found between plant growth parameters and sand and C/N (Figure 3a).

In addition, when the relationships of soil properties in control areas belonging to different bedrocks were examined according to Pearson correlation, it was determined that sand, C/N, and qCO_2 values had a significant negative relationship with clay, silt, pH, EC, $CaCO_3$, C_{mic} , MR, $qMic$. Except for qCO_2 , the microbial indices of the soil showed the highest positive correlation with clay, dust, and $CaCO_3$ among the physicochemical properties ($p < 0.001$) (Figure 3b).

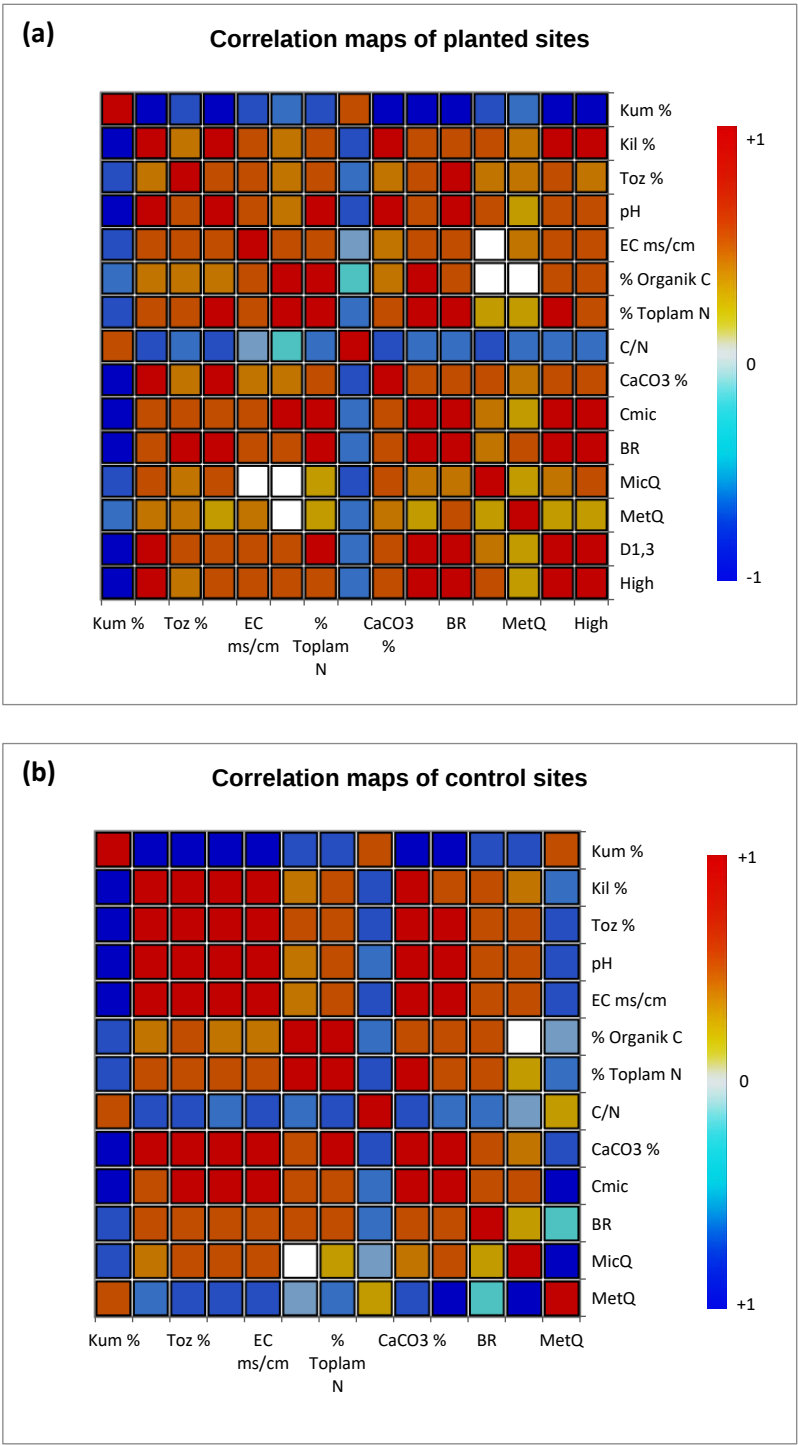


Figure 3. Correlation maps of soil samples of different parent material planted (a) and control (b) sites according to correlation coefficient matrix (R-values) for physical, chemical, and microbiological characteristics of soils in different land uses.

Discriminant analysis (DA) was able to distinguish between soils formed from limestone and andesite bedrocks in terms of physicochemical, microbiological properties, and plant growth parameters (Figure 4). The soil variables contributing to the classification and the values of the three canonical functions are shown in Table 4. Evaluations were made by accepting significant values above 0.3 [48]. Eigenvalues for the first 2 axes in all soil samples were 1.027 and 0.841, respectively; and explained 61.53% and 24.25% of the total variance (Figure 4a). Correlation coefficients show that DA axis 1 has the strongest correlation with q_{Mic} , C/N, qCO_2 , TH, DBH, and clay, respectively. DA

axis 2 is seen to be mainly related to SOC, TN, C_{mic}, and MR, respectively, with the strongest correlation (Figure 4).

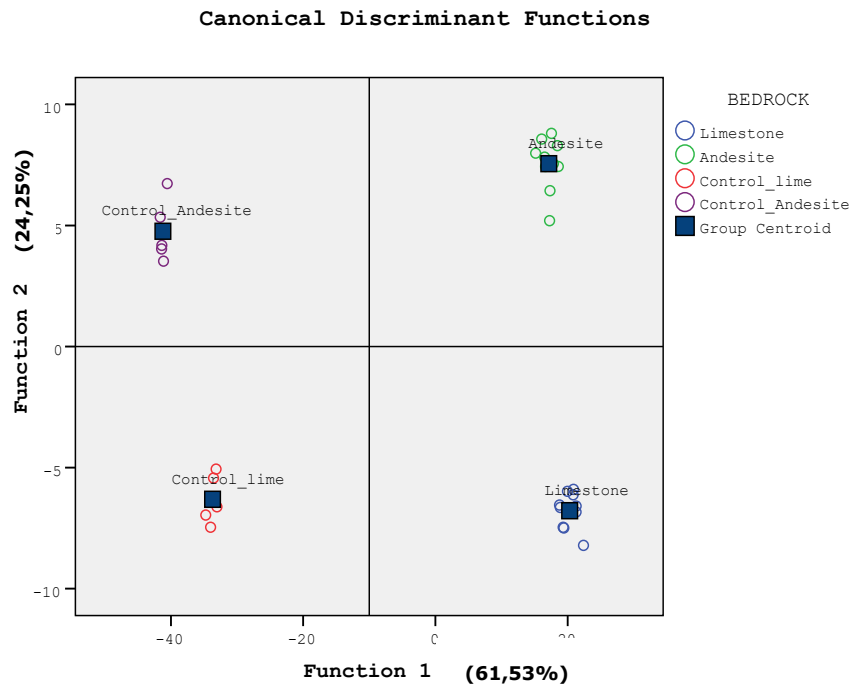


Figure 4. Scatter plot of the first two discriminant function values in soil samples.

Table 4. Standardized Canonical Discriminant Function Coefficients of soil properties and plant growth characteristics.

Soil variables	Function		
	1	2	3
TH	1,195	0,330	0,384
DBH	1,233	-0,133	-0,510
Sand	-0,613	0,900	-0,258
Clay	-0,906	0,000	-0,483
pH	0,262	0,103	0,779
EC	-1,798	-0,646	-1,695
SOC	17,718	2,887	17,085
TN	0,936	-0,066	-2,746
C/N	-0,512	-0,197	-1,344
CaCO ₃	-0,030	-0,258	0,436
C _{mic}	-14,303	-2,517	-11,114
MR	-0,434	0,412	0,607
<i>q</i> Mic(Cmic/Corg)	7,158	0,590	5,012
<i>q</i> CO ₂	0,958	0,069	0,004

4. Discussion

In this study, it was revealed that physicochemical and microbiological soil properties as well as plant growth parameters were significantly affected by the type of parent material. Limestone soils formed fine-grained soils with significantly lower sand and higher silt and clay contents. Some studies indicated that soils developed from limestone bedrock had lower sand and higher clay and silt contents than other bedrock types such as granite and acroscopic [4], and andesite [1]. Similarly, [3] determined that the grain sizes of soils from granite, granodiorite, and schist bedrocks were different.

The parent material difference also influenced the chemical properties of soils, such as pH, EC, SOC, TN, and CaCO_3 , in both control and afforestation sites. Previous studies have reported significant variations in soil pH values due to differences in parent material [1,3,4]. The EC values of soils from both parent materials at the study sites were below the critical salinity level of 0.20 dS cm^{-1} . SOC and TN content differed significantly depending on the parent material and were higher in afforestation sites compared to control sites. The increase in carbon and nitrogen sources in soils after afforestation affects the quality and quantity of organic material, including litter production [49], root exudates [50], and microbial community structure and activity [15]. Plant residues such as leaves, cones, and branches contribute to increase SOC and TN levels in the ecosystem [32,51,52]. Moreover, factors like higher clay content, tree presence [53,54], root exudates, and plant residues [15] promote soil formation. While the effect of bedrock on soil properties was observed in control sites, afforestation, particularly with Scots pine, appears to have significantly contributed to the increases in SOC and TN. The concentration of SOC and TN in the study area may be lower than in mature forests, as only fifteen years have passed since afforestation. It may take a longer period for litter to accumulate on the soil surface and integrate into the soil. Typically, SOC and TN concentrations are higher in the topsoil layer (0-20 cm) due to litter accumulation [4,32]. As expected, CaCO_3 content was significantly higher in soils with limestone parent material, attributed to the inherent lime content of limestone. There was a notable decrease in CaCO_3 content after afforestation, likely due to plant influence on soil formation. It has been reported that afforestation with different species can reduce the amount of lime in the soil, particularly in the topsoil (Babur, 2019). Limestone soils are characterized by alkaline properties and texture variations between sand (sandstone) and clay (shale) [55].

The chemical properties of soils such as pH, SOC, and TN and microbial properties of C_{mic} , N_{mic} , MR, and other indices are important sensitive indicators affected by several factors such as seasonality, tree species, bedrock properties, and soil properties [16,56]. The available C_{mic} in soils directly indicates the microbial biomass pool in the soil. The study results illustrated that the parent material significantly affects the C_{mic} concentrations in soils. Mahia et al. [47] stated that schist was significantly different from granite parent material in C_{mic} , while Babur (2019) reported that soils developed from limestone parent material had higher C_{mic} than andesite. In addition, afforestation of bare land contributes to the nutrient cycle of the ecosystem, ecosystem productivity, soil quality, and the increase in the amount of soluble and storable organic C in the soil with the input of organic matter from litter and dead roots [57,58]. Increased organic carbon in soil also affects the biological properties of soils [59]. Soil C_{mic} is generally dependent on soil organic matter as a substrate [24]. In addition, exudates brought to the soil by plant roots can increase C_{mic} accumulation, especially in topsoil [60]. In this article, it was confirmed that the increased organic matter input after afforestation also significantly increased the C_{mic} amount.

Microbial respiration is the most important microbial factor in monitoring decomposition [35,46]. Soil MR indicates the oxidation status of the soil by microbes and is considered one of the most important and sensitive indicators of the carbon cycle in soils [61,62]. Babur et al. [32], in their study conducted in four different seasons, determined that m respiration in winter was $8.96 \mu\text{g CO}_2\text{-C g}^{-1} \text{ h}^{-1}$, which was higher than in summer ($6.19 \mu\text{g CO}_2\text{-C g}^{-1} \text{ h}^{-1}$) and autumn ($6.85 \mu\text{g CO}_2\text{-C g}^{-1} \text{ h}^{-1}$). In addition, in the same study, it was determined that there were significant differences in C_{mic} and MR values of different tree species. Similarly, it was stated that the optimum soil temperature and moisture content, which accelerate microbial activities in different tree plantations, were in rainy seasons [63].

The q_{Mic} values of all soil samples were found to be within the range suggested by Anderson and Domsch [64]. q_{Mic} can be used to quickly estimate the change in organic C [65] and to compare soil quality parameters in ecosystems with different organic matter [59]. The q_{Mic} ratio is determined by using the relationship between microbial biomass carbon and soil organic carbon [66]. The q_{Mic} ratio of andesite soils of the study areas was lower than that of limestone. This provided more suitable conditions for the growth of soil microorganisms in the limestone bedrock. Although the average q_{Mic} values increased after afforestation, this value remained low for fertile lands and did not reach equilibrium 15 years after afforestation. The q_{Mic} value is estimated by microbial activity, i.e., soil basal respiration and organic carbon cycling within an ecosystem [35,67]. Higher MR and C_{mic} amounts in limestone soils indicated that it had more microbial activity, proving faster decomposition.

The q_{CO_2} is often used to describe the soil stress response of an ecosystem [67]. Environmental stresses, nutrient scarcity, and substrate resistance to decomposition can increase q_{CO_2} [68]. On the other hand, the productivity of the area can be increased by using natural vegetation in stressed areas [69]. Although Anderson [67] reported that q_{CO_2} values in neutral soils ranged from 0.5 to 2.0 $\mu g\ CO_2-C\ g^{-1}C_{mic}\ h^{-1}$, in this study, the average q_{CO_2} values in areas afforested with andesite and limestone bedrocks were found to be 3.37 to 4.06 $\mu g\ CO_2-C\ g^{-1}C_{mic}\ h^{-1}$, respectively. This may cause microbial respiration to increase, decomposition to slow down and energy to be consumed in it. Low q_{CO_2} rate in soils provides positive information about efficient carbon use for that soil, low environmental stress levels, usable nutrients being free in the area, microbial activity, and soil quality [22].

The q_{CO_2} and OC mineralization rates in soil constitute the basis of the carbon cycle in terrestrial ecosystems [10,70]. SOC content negatively correlated with the metabolic coefficient in both parent materials. Some studies have noted that OC and q_{CO_2} in different forest soils are closely related [1,60]. As a result of the correlation analysis, it was determined that basal respiration was positively correlated with soil organic carbon in both parent materials. Similar results were found with Cheng and Xia [71].

Differences in parent material and soil properties showed significant differences in plant growth parameters such as tree height and diameter at breast height. In particular, Scots pines on limestone bedrock were 20% and 25% larger in height and diameter than on andesite bedrock, respectively. There are many studies on the importance of the effects of habitat properties on plant yield and growth characteristics [72–74]. Microbiological, physical, and chemical soil properties can improve as the age of the afforested area. Afforestation and rehabilitation practices in forest management contribute to the health and productivity of soils and support the restoration of soil microbial activities.

Available C fractions are one of the most important environmental factors affecting soil microbial activity [22,75]. Analyses performed on soil samples, particularly microbial biomass, MR, and other indices, may be responsible for the separation of different parent materials and control areas with DA because the parent material is an important factor in soil formation, and tall plants also provide more living space and substrate for microbial communities in soils with their aboveground and belowground biomass and waste. Improvement of microorganisms' living environments can also be understood by the increase in q_{Mic} value and decrease in q_{CO_2} value [10,76]. In fact, from our research, it was determined that there was an increase in q_{Mic} amount and a decrease in q_{CO_2} amount in soils on both bedrocks after afforestation. These results provide strong evidence that afforestation on different bedrocks results in different soil properties and microbial communities compared to bare lands.

5. Conclusion

In this study, it was determined that the physicochemical and microbiological properties of soils formed from different parent materials were different and this significantly affected the growth parameters of the planted seedlings. It was determined that the fertility of soils in terrestrial ecosystems was significantly affected primarily by the type of parent material and then by the

presence of vegetation on it. The fact that limestone parent rock can be easily decomposed compared to andesite and that it creates deep and fertile soil allowed Scots pine trees to grow better on limestone parent rocks. It was shown that afforestation contributed to the increase in organic C and N, especially in the upper soils, in bare lands, and this contributed to the increase in microbial biomass and activity in the soils. Soils from limestone parent rocks afforested with Scots pine were energetically more efficient (had a lower $q\text{CO}_2$) with more microbial biomass and a higher $q\text{Mic}$ ($C_{\text{mic}}/C_{\text{org}}$) compared to andesite soils. This study emphasized how important parent material is in terrestrial ecosystems and that it directly affects soil microbial activity. In particular, afforestation provides substrate and C to the ecosystem via tree litter, thus increasing soil microbial communities and activities. Since the measured microbial parameters are sensitive to changes in the ecosystem, they may have the potential to be used as indicators of forest management on soil organic matter quality. In order to ensure data continuity in such areas and to verify the data of afforestation sites in arid and semiarid areas, more comprehensive studies are required. In afforestation to be made on andesite bedrocks, more care should be taken, and maintenance, fertilization, and even irrigation during periods of water stress can contribute to the growth of seedlings and environmental fertility. In the afforestation of arid and semiarid areas, attention should be paid to the selection of local species and close origins. In addition, it would be more appropriate to make afforestation using mixed species with leaves and conifers to increase the ecological sensitivity limit. Thus, ecosystem diversity, nutrient cycle, and soil C input will be supported.

Author Contributions: For research articles with several authors, a short paragraph specifying their individual contributions must be provided. The following statements should be used “Conceptualization, E.B. and E.B.; methodology, E.B.; software, E.B.; validation, E.B.; formal analysis, E.B.; investigation, E.B.; resources, E.B.; data curation, E.B.; writing—original draft preparation, E.B.; writing—review and editing, E.B.; visualization, E.B.; supervision, E.B.; project administration, E.B.; funding acquisition, E.B. All authors have read and agreed to the published version of the manuscript.” Please turn to the [CRediT taxonomy](#) for the term explanation. Authorship must be limited to those who have contributed substantially to the work reported.

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