



OPEN Soil respiration, microbial biomass, and stoichiometry within riparian buffers and adjacent land use

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Terrestrial ecosystems exhibit varied land uses as a result of both anthropogenic activities and natural processes. These variations in land use alter plant composition, soil characteristics, topography, and management practices, and hence lead to significant differences in soil microbial communities and their properties. This study evaluates the impact of distinct land use types (riparian, forest, pasture) on soil microbial biomass and microbial stoichiometric indices under uniform climatic and pedological conditions within a micro-basin in the Eastern Mediterranean region. The microbial biomass C (C_{mic}) in the riparian area was 2.5 and 4 times lower than in the meadow and forest areas, respectively. Additionally, the riparian zone's microbial quotient ($qMic$) was 0.5 times higher than the forest and meadow areas. Microbial stoichiometric indices, particularly $qMic$ and metabolic quotient (qCO_2), across all land uses, indicated that soils within this micro-basin were healthy and exhibited no signs of stress. The study further corroborated that land use exerts significant effects on soil microbial communities, with microbial biomass and activities largely influenced by soil organic matter. Notably, the C_{mic}/N_{mic} ratio remained within the range of 10–12 across all land uses, illustrating a fungal dominance in the microbial biomass. These findings underscore the role of land use patterns in altering soil properties, thereby influencing microbial biomass, microbial respiration, and stoichiometry in soils under similar environmental conditions.

Keywords Land use, Microbial biomass, Stoichiometry, Riparian, Soil microbial activity, Forest

A comprehensive understanding of soil properties within areas delineated by land use capabilities is essential for sustainable environmental management. In Turkey, as in other regions, land degradation is occurring at an alarming rate due to inappropriate land use practices¹. Land use change and degradation lead to intense erosion. Kara, et al.² reported that 86% of Turkey is vulnerable to varying degrees of erosion. Erosion predominantly affects the topsoil, where soil microbial communities are most active, significantly impacting microbial populations and their functional activities³.

Soils serve as the foundation for primary production, host diverse microbial communities that drive material cycling, and provide vital nutrients and hydrological reservoirs essential for the survival of higher organisms⁴. Therefore, soils in sensitive areas must be protected from changes such as erosion, inappropriate forestry practices, and other anthropogenic impacts. Over the last 30 years, the increasing demand for agricultural products has triggered the conversion of forests to agricultural land, with land use change increasing at least fivefold worldwide (from ~ 3 to ~ 15 million km²)⁵.

Topography, climate variations, soil properties, aboveground vegetation diversity, and land use are the main factors controlling the microbial properties and dynamics of soils⁶. Ecosystem health and continuity depend on carbon and nutrient flow mediated by microbial organisms in the soil–plant–atmosphere cycle⁶. However, anthropogenic effects and climate variability impact microbial population and diversity⁷. Microorganisms are sensitive to the aboveground litter composition, root structure, and root enzyme ratios of vegetation, i.e., to changes in land use⁸. So, the substrate quality of the organic matter entering the soil plays an important role in the diversity and populations of microorganisms as it regulates the microbial decomposition rate⁹.

Recently, there has been growing interest in assessing the role of soil microbial biomass and basal respiration in ecosystem functioning, particularly their influence on soil quality following land use changes¹⁰. Therefore, microbial biomass, basal respiration, and their stoichiometric properties serve as early warning indicators,

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exhibiting rapid and heterogeneous responses to environmental disturbances, making them invaluable for ecosystem monitoring and management^{11,12}. Soil microorganisms are essential for terrestrial carbon and nutrient cycling, ecosystem stability, and climate regulation, as they drive organic matter decomposition and regulate nutrient dynamics, thereby enhancing soil organic carbon storage¹³. Notably, microbial biomass carbon contributes approximately 1–3% of the total soil organic carbon pool, underscoring its ecological significance¹⁴. Unfortunately, disruptions beyond microbial adaptability can lead to the collapse of essential ecosystem functions, such as nutrient cycling and soil structure maintenance, threatening overall ecological balance and resilience^{11,15}.

Riparian areas differ from other regions in terms of soil and hydrological properties, yet few studies have focused on the microbial properties, soil respiration, and stoichiometric indices of soils in riparian areas¹⁶. Positioned between agricultural lands, pastures, meadows, and streams, riparian zones play a crucial role in protecting stream and lake shores from erosion by acting as buffers that filter sediments, nutrients, and pesticides¹⁷. To perform these functions effectively, riparian soils must support distinct microbial biomass and activity, reflecting characteristics of both terrestrial land uses and aquatic systems¹⁸. Studies on soil microbial biomass carbon in different land-use systems have been conducted by many researchers worldwide^{18–20}. However, most research has focused on forests and their conversion to agricultural land^{12,21}, with limited studies on adjacent forests, pasture, and riparian areas in natural water production basins.

The Mediterranean basin, which is rich in biodiversity but highly sensitive to global warming and fires, is particularly fragile due to frequent land use transformation/change through deforestation, land degradation, and hydrological disruption²². These changes and transformations significantly impact microbial community compositions and activities²³, with variations in soil biodiversity, substrate quality, and quantity, microbial carbon productivity, and stoichiometric limitations depending on soil type^{24,25}.

Land use patterns play a crucial role in shaping microbial activity within the Mediterranean region's micro-basins, a particularly fragile ecosystem vulnerable to the impacts of global warming²¹. Although numerous studies have examined the physical, chemical, and microbial properties of soils developed under different tree species^{16,19,21}, seasons^{20,21}, and land uses^{12–14,17,18}, comprehensive research on soils within adjacent land use patterns at the micro-catchment scale remains limited. Therefore, to ensure the sustainability of ecosystem health, it is crucial to understand the soil physicochemical and microbial properties in various land uses, particularly within drinking-water collection basins^{26,27}. In this context, this study focuses on the interactions between microbial biomass and respiration as influenced by organic matter availability and other soil health indices. Moreover, more in-depth studies to increase understanding of how land use modifications impact soil health, microbial functions, and ecosystem stability, may provide valuable implications for sustainable land and water management practices. Therefore, this research was conducted to evaluate the effects of land use on soil microbial respiration, microbial biomass, and stoichiometric properties within the Meydan Pond micro-basin in the Andırın District of Kahramanmaraş. It aims to deepen our understanding of microbes' regulatory roles within various ecosystems and the mechanisms underlying soil microbial responses. Additionally, it provides a scientific foundation for enhancing watershed management practices, protecting vegetation, and preserving adjacent zonal ecosystems.

Material and method

Study sites

This study was conducted within a 50 km distance from Kahramanmaraş Province, located in the Northeast Mediterranean Sea mountainous region of Turkey. Except for the different land uses in each case, the sampling area has similar topographic, geological, and climatic conditions characterized by the presence of a typical karstic topography (Fig. 1). This mountain range varies from the Upper Cretaceous to Upper Miocene and a Mosezoic limestone succession; it is derived from the Taurus System²⁸. The elevation ranges from 1200 to 1350 m, with steep slopes (50%). The forest canopy in the research area consists primarily of mixed coniferous tree species, aged 80–100 years, including black pine (*Pinus nigra* Arn.), Taurus fir (*Abies cilicica* subsp. *cilicica*), Lebanon cedar (*Cedrus libani* A. Rich.). The pasture area is typically populated by species from the genera *Trifolium* sp., *Cynodon* sp., *Astragalus* sp., *Salvia* sp., and others²⁹. In the riparian zone, *Phragmites australis* is present.

The region has a hot-dry summer and cool-wet winter climate type, Mediterranean climate, (Csa) according to the Köppen-Geiger climate classification (www.koepen-geiger.vu-wien.ac.at/). Weather data, including long-term (at least 30 years 1990–2020) averages for monthly air temperature and precipitation in Andırın district, were obtained from the Turkish State Meteorological Service (www.mgm.gov.tr). The annual mean temperature is 13.0 °C, with a high of 30 °C in August and a low of –2.5 °C in February. Annual precipitation is 729 mm, with about 80% occurring during the winter and early spring seasons. The main soil types are generally brown forest soils, classified as Alfisols by the U. S. Soil Taxonomy³⁰. The rugged topography consists of andesitic and limestone bedrock. Table 1 provides a detailed characterization of the study site.

Experimental design, soil sampling, and pretreatment

This study was conducted in the forest, pasture, and riparian areas of the Baskonus Plateau Meydan Pond micro-basin in May 2019. Five different sampling points were chosen randomly from each land use type (forest, pasture, and riparian) at the 0–10 cm depth, a total of fifteen soil samples for this study. A maximum depth of 10 cm was chosen for soil sampling because of the most intense microbial activity and the significant limiting effect of riparian areas beyond 10 cm depth due to the presence of water tables and rocks. These areas are designated as protected zones and are generally restricted from human activity, minimizing anthropogenic influence. Human access is typically limited to recreational use.

For microbial analysis, the litter, root debris, and stones were removed from all soil samples, then sieved through a <2 mm mesh. Distilled water was added to until 50–60% moisture content, and the samples were

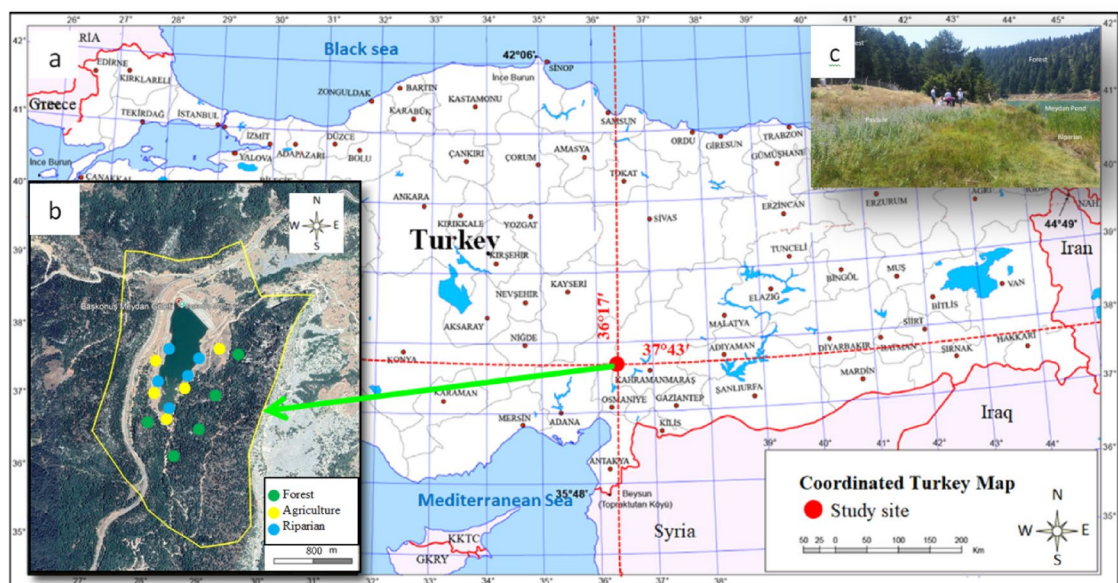


Fig. 1. Locations of the study site (a), sampling points (b) and field study (c). The map was created using Google Earth Pro Maps (version 7.3.6.9796) and ArcGIS (version 10.4.1, <https://www.esri.com/en-us/arcgis/products/arcgis-pro>).

Environmental factors	Land-use types		
	Forest	Pasture	Riparian
Longitude	36° 35' 15"	36° 35' 25"	36° 35' 20"
Latitude	37° 34' 25"	37° 34' 45"	37° 34' 47"
Altitude mean (m)	1250	1210	1200
Slope degree mean (%)	25	10	5
Vegetation type	Black Pine-Fir	Pasture vegetation	Rush bed
Stand canopy (%)	70–100	70–100	40–70
Soil texture	Sandy loam	Sandy clay loam	Sandy loam
pH class	7.66	7.56	7.50

Table 1. Study site characteristics.

stored in a portable coolant below +4 °C until promptly transported to the laboratory for storage at +4 °C before analysis for the maintains microbial activity. Physicochemical properties and erodibility indices were determined by Babur, et al.³¹.

Microbial Biomass C (C_{mic}) and Microbial Biomass N (N_{mic})

The chloroform-fumigation-extraction (CFE) method was performed to determine soil microbial biomass carbon (C_{mic}) and biomass nitrogen (N_{mic})³². For CFE, each soil sample was divided into 2 portions of 30 g, 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 vacuum desiccator in the dark incubator, while the second portion of samples was used as a control without fumigation. 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 min at 200 rpm on a reciprocating oscillator and supernatants were filtered through a Whatman No. 42 filter paper and stored in below +4 °C before titration. Total organic carbon in both fumigated and non-fumigated extracts was estimated as specified by the Walkley Black method^{33,34}. Briefly, 8 ml aliquot of K_2SO_4 extracts after oxidation with 0.4N $K_2Cr_2O_7$ and back-titration with ferrous ammonium sulfate. C_{mic} was calculated from the difference in extractable organic C between fumigated and non-fumigated soil samples using the formula as follows in Eq. 1^{32,34}.

$$\text{Microbial Biomass C } (C_{mic}) = 2.64 * E_C \quad (1)$$

where E_C refers to the difference in extractable organic C between the treatments; 2.64 is the proportionality factor for biomass C released by fumigation extraction.

The microbial quotient (C_{mic}/C_{org} or $qMic$) was calculated by expressing microbial C as a percentage of total soil organic C.

The Kjeldahl digestion-distillation-titration method was used to determine the total N in the extracted K_2SO_4 solution. Briefly, a mixture was prepared by adding 15 ml of the extract solution + 0.4 ml of 0.2 M $CuSO_4$ + to promote organic matter degradation and then 10 ml of high purity (97%) H_2SO_4 and placed in the digestion unit. This mixture was digested at 380 °C for at least 3 h (until all organic compounds were decomposed). The solution was made up to 250 ml with deionized water. A 50 ml subsample was taken and distilled in the distillation unit with 10 M NaOH (strong base). The distillate was collected in an indicator solution mixed with boric acid and the solution was titrated. N_{mic} was calculated from the difference in extractable N between fumigated and non-fumigated soil samples by using the following Eq. ^{23,35,36}:

$$\text{Microbial Biomass N (} N_{mic} \text{)} = FN/0.54 \quad (2)$$

where FN is the difference between N extracted from fumigated and non-fumigated samples and 0.54 is the fraction of microbial biomass N released by the fumigation extraction process.

Microbial respiration (MR)

Microbial respiration was estimated by the sodium hydroxide (NaOH) trap method (absorption method) to measure carbon dioxide (CO_2)³⁷. Briefly, 30 g of oven-dried equivalent weight moist soils were weighed and transferred to separate 500 mL capacity airtight beakers. Small beakers filled with 0.1 M NaOH were placed in the center of the bottom of the jar without soil underneath to capture the CO_2 released as a result of microbial respiration in the beakers containing the soil sample. The jars were hermetically sealed and incubated at 25 °C for 7 days. After 1 week of incubation, the small beaker containing the CO_2 absorbed in the NaOH solution was removed and titrated with 0.05 M HCl after adding $BaCl_2$. An empty jar without soil was used as the control. Then, MR was calculated by dividing the amount of CO_2 released during the incubation period by the dry mass of the soil. Metabolic quotient qCO_2 , an important parameter that can be used to predict soil quality and stress response, was calculated as the ratio between MR ratios ($mg\ CO_2\text{-C}\ g\ C_{mic}^{-1}\ h^{-1}$) and C_{mic} ^{35,38}.

Statistical analyses

The data were assessed for normality and homogeneity of variance using skewness, kurtosis, and the Shapiro–Wilk test. Parameters that deviated from a normal distribution were log-transformed (log10) before conducting variance analysis (ANOVA). Once the data met the assumptions for parametric testing, a one-way ANOVA was performed to evaluate the effects of land use type on all measured parameters, followed by Duncan's multiple range tests ($\alpha=0.05$) for post-hoc comparisons. Pearson's linear correlation analysis was conducted to investigate the relationships between microbial soil properties and stoichiometric parameters across different land uses. Principal component analysis (PCA) was performed using RStudio software, based on the correlation matrix of soil properties with sufficient replicate data across different land use types. This approach allowed for the visualization of overall similarities and differences among the soil properties of various land uses. PCA was used to calculate the differences between data groups, employing one or more discriminant functions to optimally distinguish among the identified groups. Statistical analyses were conducted using the SPSS Inc. (Statistical Package for Social Sciences, Chicago, IL, USA), RStudio, and figures were generated with the Rstudio and XLSTAT software package for Windows.

Results and discussion

Changes in microbial biomass C (C_{mic}), N (N_{mic}), and microbial respiration (MR) under different land-use

This study revealed significant differences in microbial properties and stoichiometric parameters across land use types (Fig. 2 and Table 2). Soil microbial biomass decreased from forest to pasture and riparian ecosystems, reflecting a transition from well-developed to sparser vegetation. Forest ecosystems supported more active microbial communities compared to pasture and riparian zones.

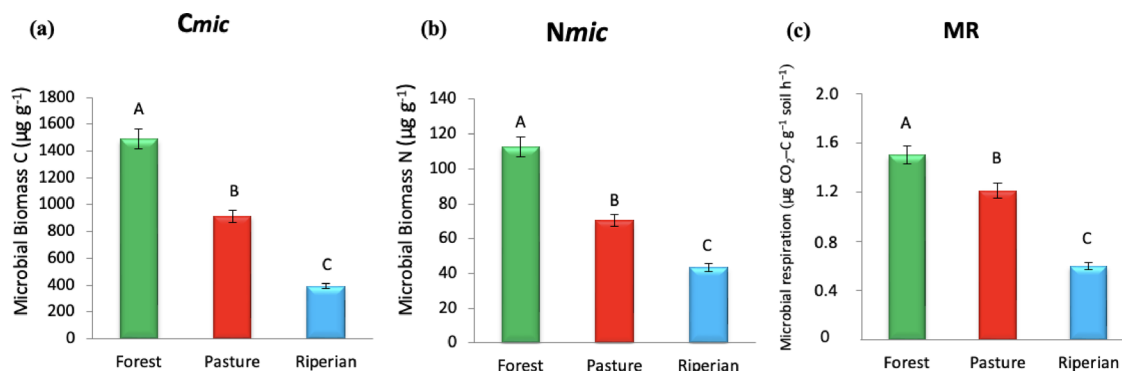


Fig. 2. Changes in microbial biomass C (a), microbial biomass N (b), and microbial respiration (MR) (c) with afforestation in three land-use types. Different letters above the bars indicate significant differences at $p < 0.05$ among the land use types (Error bars show \pm standard errors).

	C_{mic}	N_{mic}	MR	$qMic$	qCO_2	C_{mic}/N_{mic}	N_{mic}/TN
Land-use type	***	***	***	*	ns	**	***
F-values	556	80.0	39.1	6.28	2.97	10.73	22.7

Table 2. F statistics of the one-way ANOVA for the effects of land-use types on soil microbial properties and microbial stoichiometric indices. C_{mic} carbon in the microbial biomass ($\mu\text{g g}^{-1}$), N_{mic} nitrogen in the microbial biomass ($\mu\text{g g}^{-1}$), MR microbial basal respiration ($\mu\text{g CO}_2\text{-C g}^{-1}\text{ h}^{-1}$), qCO_2 microbial metabolic quotient ($\text{mg CO}_2\text{-C g C}_{mic}^{-1}\text{ h}^{-1}$) $qMic$ Microbial quotient. a No significant differences are indicated by ns. * Significant at $p \leq 0.05$; ** Significant at $p \leq 0.01$, *** Significant at $p \leq 0.001$.

The C_{mic} and N_{mic} varied significantly across land-use types ($p < 0.001$) with forest soil being significantly higher than both pasture and riparian soils (Fig. 2; Table 2). Briefly, the soil C_{mic} in the topsoil (0–10 cm), the forest soils had $1487 \mu\text{g g}^{-1} C_{mic}$, 63% and 280% higher than those under the pasture, $913 \mu\text{g g}^{-1}$, and the riparian soils, $391 \mu\text{g g}^{-1}$ (Table 2 and Fig. 2a). Determination of soil C_{mic} is one of the main factors in most soil monitoring studies and programs³⁹. Previous studies conducted in different forest soils have reported microbial biomass C ranging from 470 to $1600 \mu\text{g g}^{-1}$ in beech forests, 586 to $790 \mu\text{g g}^{-1}$ in high-altitude (1750 m) cedar forests, and 251 to $838 \mu\text{g g}^{-1}$ in pine forests^{12,13,40,41}. In addition, C_{mic} concentrations of soils in the rainy season were found to be $1330.00 \mu\text{g g}^{-1}$ in mixed forest ecosystems, $1222.98 \mu\text{g g}^{-1}$ in pastures, $380.33 \mu\text{g g}^{-1}$ in afforestation areas, and $284.30 \mu\text{g g}^{-1}$ in agricultural areas^{12,21}. In our study, the mean C_{mic} values were found within these previously reported ranges. These results suggest that the high nutrient availability in mixed coniferous forests, driven by substantial litter input and composition, creates more favorable conditions for microbial decomposers. Chen, et al.⁴² stated in their study that the amount and type of organic matter caused significant differences in the C_{mic} content of soils. In addition, Vidyanaagar⁴³ found that there were significant differences in the C_{mic} content of topsoil (0–10 cm) developed under different land use types.

This study revealed that the average N_{mic} levels in soil samples varied significantly across different land use types. N_{mic} represents the total nitrogen content of soil microorganisms and plays a crucial role in evaluating the contribution of soil microorganisms to the nitrogen cycle, as well as in monitoring soil fertility, health, and quality¹¹. In this study, the differences soil N_{mic} were statistically significant ($p < 0.001$) with forest soil containing significantly more N_{mic} than both pasture and riparian soils (Table 2). For example, the forest vegetation had $122.6 \mu\text{g g}^{-1} N_{mic}$ which was 74% and 183% higher than those under pasture, $70.4 \mu\text{g g}^{-1}$, and riparian soils, $43.3 \mu\text{g g}^{-1}$ (Fig. 2b). Forest soils exhibited higher N_{mic} contents compared to other land uses, which can be attributed to the high levels of organic carbon and total nitrogen contributed by the rapid decomposition of cedar needles in the areas¹². Díaz-Raviña, et al.⁴⁴ reported that N_{mic} content in topsoil under different tree species ranges between 42 and $191 \mu\text{g g}^{-1}$, Arunachalam and Arunachalam⁴⁵ found higher N_{mic} content ranging between 172 and $331 \mu\text{g g}^{-1}$ in soils from humid forests in Northeast India. Other studies have found N_{mic} values ranging from 42 to $142 \mu\text{g g}^{-1}$ across various forest ecosystems^{21,46}. Vidyanaagar⁴³ observed N_{mic} values of $123.30 \mu\text{g g}^{-1}$ in mixed forest ecosystems, $107.85 \mu\text{g g}^{-1}$ in pastures, $47.85 \mu\text{g g}^{-1}$ in afforested areas, and $42.83 \mu\text{g g}^{-1}$ in agricultural lands. These findings indicate that N_{mic} content is highest in forested areas, while it is lowest in agricultural and other land-use types. The lignin and nitrogen content in organic material serves as indices of litter resistance to decomposition and substrate quality⁴⁷. The differences in C_{mic} and N_{mic} of soils developed under different land uses may be due to the variability in soil type. Although the soils are derived from similar parent material, they have different soil properties due to different plant biodiversity and land use^{7,19}. For example, forest soils have higher organic matter inputs than pastures and riparian areas⁴³.

Soil microorganisms metabolize organic carbon sources such as plant residues and dead organisms to obtain energy, resulting in the release of carbon dioxide (CO_2)^{35,48}. This represents microbial respiration. MR plays a critical role in maintaining the carbon cycle within the soil ecosystem, facilitating the decomposition of organic matter, and contributing to soil fertility^{12,49}. Soils of this study further showed that soil microbial respiration (MR) rates were significantly altered by land use impacts (Fig. 2c). MR in riparian soils was lower than those under both forest and pasture ($p < 0.001$; Table 2) where forest soils exhibited the highest MR of $1.5 \mu\text{g CO}_2\text{-C g}^{-1}\text{ h}^{-1}$, which was followed by pasture soils with $1.21 \mu\text{g CO}_2\text{-C g}^{-1}\text{ h}^{-1}$ MR, and riparian soils with $0.6 \mu\text{g CO}_2\text{-C g}^{-1}\text{ h}^{-1}$, respectively. In this study, as a result of distinct land use among microbial respiration trends, forest > pasture > riparian areas are ranked. Some researchers have noted that increased organic matter or optimal climatic conditions enhance microbial activity, which is positively correlated with MR^{50,51}. Similarly, in our study, it is estimated that the high OC and C_{mic} amounts in the soils in forest areas are due to the high tendency of microorganisms in the soil to produce energy by breaking down organic matter. VM and Marcos ML⁴⁶ determined in their study on oak forest soils that the amount of MR in different seasons varied between 0.30 and $1.20 \mu\text{g CO}_2\text{-C g}^{-1}\text{ h}^{-1}$. Chen, et al.⁵² reported that MS values of soils in mixed pine forests and pastures varied between 0.17–0.65 and 0.11–0.84 $\mu\text{g CO}_2\text{-C g}^{-1}\text{ h}^{-1}$, respectively. However, Bolat⁵³ found the highest MR value in fir forest soils with $1.79 \mu\text{g CO}_2\text{-C g}^{-1}\text{ h}^{-1}$. In cases where soils carry stress conditions, the MR value will be higher as microbial organisms will produce more CO_2 to provide the energy they need.

The pattern of microbial stoichiometry in the three land-use

The findings from this study also showed a significantly altered pattern of microbial stoichiometry impacted by different land uses (Tables 2 and 3). In this study, the pattern of microbial stoichiometry includes all soil $qMic$, qCO_2 , C_{mic}/N_{mic} , and N_{mic}/TN indices.

Characteristics	Land-use types		
	Forest	Pasture	Riparian
q_{Mic}	$2.31 \pm 0.06^{\text{B}}$	$2.06 \pm 0.19^{\text{B}}$	$2.90 \pm 0.63^{\text{A}}$
q_{CO_2}	$1.00 \pm 0.07^{\text{B}}$	$1.32 \pm 0.14^{\text{A,B}}$	$1.54 \pm 0.59^{\text{A}}$
$C_{\text{mic}}/N_{\text{mic}}$	$12.21 \pm 1.29^{\text{A}}$	$13.14 \pm 1.57^{\text{A}}$	$9.18 \pm 1.35^{\text{B}}$
N_{mic}/TN	$4.02 \pm 0.41^{\text{A}}$	$2.10 \pm 0.06^{\text{B}}$	$4.95 \pm 1.11^{\text{A}}$

Table 3. Selected soil microbial stoichiometric indices \pm standard deviation derived from different land use types at the topsoil (0–10 cm). Data are means \pm standard deviation ($n = 5$). Different letters indicate significant differences among the Land use types at $p < 0.05$.

The results of this study indicate significant differences in q_{Mic} in the topsoil (0–10 cm) across the three land uses: forest, pasture, and riparian ($p < 0.05$; Table 2). Notably, the forest soils had significantly lower q_{Mic} (2.31) where the riparian soils had the highest q_{Mic} (2.90). A similar difference was also observed between pasture soils and riparian soils where pasture soils had significantly lower q_{Mic} (2.06) than riparian soils. However, although differences between forest and pasture soils were not significant, forest soils had slightly higher q_{Mic} compared to those of pasture soils. The q_{Mic} ($C_{\text{mic}}/C_{\text{org}}$) ratio is a crucial parameter for assessing soil microbial activity, fertility, and overall health, and it can also serve as a tool for monitoring the effectiveness of soil management practices⁸. In addition, the q_{Mic} ratio is widely recognized as an effective microbial characteristic for monitoring spatial and temporal changes in soils¹¹. Those like Jenkinson and Ladd⁵⁴ suggested that a q_{Mic} ratio of 2.0 is a balance threshold for soils. The mean q_{Mic} percentage values in our study were above this threshold, reflecting the efficient use of organic substrates by microbial biomass across all land uses. According to Anderson⁴⁸, a low q_{Mic} ratio results from excessive carbon being utilized for respiration, with minimal microbial biomass production, which is indicative of environmental stress conditions. Low q_{Mic} ratios suggest that only a small fraction of organic carbon is accessible to soil microorganisms, indicating poor substrate quality⁵⁵, whereas a high ratio is often considered a sign of carbon immobilization⁴³. The values obtained align with previous studies, where the q_{Mic} ratio was arranged from a minimum of 0.27 to a maximum of 9.9^{43,44,48,56,57}.

The mean q_{CO_2} values and standard deviations for soils across different land uses are presented in Table 3. In contrast to q_{Mic} , q_{CO_2} did not show a significant variability ($p > 0.05$) across different land-use types (Table 2). The q_{CO_2} of riparian soils ($1.54 \text{ mg CO}_2\text{-C g C}_{\text{mic}}^{-1} \text{ h}^{-1}$) was higher than those of both pasture ($1.32 \text{ mg CO}_2\text{-C g C}_{\text{mic}}^{-1} \text{ h}^{-1}$) and forest ($1.00 \text{ mg CO}_2\text{-C g C}_{\text{mic}}^{-1} \text{ h}^{-1}$) soils by 14.3% and 35.1%. However, differences were not always statistically significant indicating that great variability in data may lead higher standard deviations (~ 0.59) and in turn kept p -values higher than 0.05 significant level. The metabolic quotient (q_{CO_2}) represents one of the key indicators of the functioning and health of soil ecosystems. To assess the stress conditions within an ecosystem, it is essential to determine q_{CO_2} values alongside other indicators, such as the microbial quotient ($q_{\text{Mic}} = C_{\text{mic}}/C_{\text{org}}$)^{12,49}. According to the ANOVA test, the q_{CO_2} values for soils from different land uses were not significantly different ($p = 0.09$; Table 2). However, post hoc analysis revealed that the land uses formed distinct groups based on their q_{CO_2} values, with pasture soils showing similarity to both forest and riparian soils in terms of their q_{CO_2} averages (Table 3). This may be attributed to the relatively close and small differences in q_{CO_2} average values across the land uses. Babur, et al.¹² found that the average q_{CO_2} amount of soils varied between 1.55 and 2.63 $\text{mg CO}_2\text{-C g}^{-1} \text{ C}_{\text{mic}} \text{ h}^{-1}$ in different forest ecosystems. Again, in the same study, it was stated that the average q_{CO_2} amount increased to 4.99 $\text{mg CO}_2\text{-C g}^{-1} \text{ C}_{\text{mic}} \text{ h}^{-1}$ in the winter season when the microbial population was stressed with the decrease in air temperature and had to respire more to convert frozen organic carbon into nutrient energy. Some researchers have determined that this value varies between 0.74 and 162 $\text{mg CO}_2\text{-C g}^{-1} \text{ C}_{\text{mic}} \text{ h}^{-1}$ ^{146,49,53}.

When comparing the average $C_{\text{mic}}/N_{\text{mic}}$ ratios of soils across different land uses during the sampling time, the highest average ratio was observed in pasture areas (13.14), while the lowest was found in riparian areas (9.18). Statistical analysis also showed that the $C_{\text{mic}}/N_{\text{mic}}$ ratio in forest and pasture was significantly higher than in riparian soils ($p \leq 0.01$, Tables 2 and 3), indicating more fungal dominance in forest and pasture than riparian soils which eventually may have more bacteria domination. The $C_{\text{mic}}/N_{\text{mic}}$ ratio is a valuable indicator for determining the dominant type, structure, and status of the microorganisms that constitute the microbial biomass in the study areas⁵⁸. According to Jenkinson and Ladd⁵⁴, a $C_{\text{mic}}/N_{\text{mic}}$ ratio of approximately 10–12 suggests that the microbial biomass is predominantly composed of fungi, whereas a ratio between 3 and 5 indicates bacterial dominance. Based on these threshold values, the $C_{\text{mic}}/N_{\text{mic}}$ ratios obtained from the soils in our study indicate that the microbial community in our research fields is primarily composed of fungal species. Bauhus and Khanna⁵⁶ emphasized that fungi play a more critical role than bacteria in the initial decomposition of litter, and thus, a higher $C_{\text{mic}}/N_{\text{mic}}$ ratio is expected. Other studies have reported that the $C_{\text{mic}}/N_{\text{mic}}$ ratio can range from 3.0 to 17.3, depending on land use and tree species^{12,13,53,59}.

The average N_{mic}/TN ratios of soils across different land uses in the study areas were highest in riparian areas (4.95%) and lowest in pasture areas (2.10%). In forests, this value was found to be 4.02 (Table 3). The N_{mic}/TN values differed significantly among soils from different land uses ($p \leq 0.001$, Table 2). The mean and standard deviation values of the N_{mic}/TN ratios for soils from different land uses are presented in Table 3. However, differences between forest and riparian were not statistically significant ($p > 0.05$). Soil N_{mic}/TN ratio, like other stoichiometric indices, is an important biological indicator used to evaluate soil microbial activity, nitrogen cycle, soil fertility, and general health⁵⁷. It has been shown that a high N_{mic}/TN ratio in soils does not significantly impact the availability of nitrogen (N). In environments with elevated N_{mic}/TN ratios, organic compounds can

be more readily utilized, leading to regular decomposition of organic matter and a subsequent decrease in its content⁶⁰. According to Bauhus, et al.⁵⁵, both low and very high N_{mic}/TN ratios may indicate limitations in substrate availability. This is because the retention of released nitrogen by microbial organisms can reduce the amount of nitrogen available for plant uptake. The low N_{mic}/TN ratio observed in pasture areas may be attributed to a low microbial biomass N content, increased nitrogen uptake by plants or nitrogen loss from the environment and subsequent accumulation in the riparian zone due to leaching. The riparian areas are adjacent to pasture lands and contain substantial pond water, which could contribute to this effect. Additionally, the low N_{mic}/TN ratio could be explained by the fixation of free nitrogen in the soil by microbial organisms. The N_{mic}/TN values observed in this study (0.5–12.40%) are consistent with findings from previous research^{12,43,49,53,56}.

Relationship between microbial properties and stoichiometric indices

Pearson's correlation analysis showed that soil MR had a significant positive correlation with C_{mic} , N_{mic} , and C_{mic}/N_{mic} ($p < 0.05$). About 92% of MR was explained by C_{mic} whereas N_{mic} explained 82% of MR (Table 4). A similar relationship was observed between C_{mic} and N_{mic} ($p < 0.05$) where these properties explained about 96% variability. Another important positive correlation was observed between $qMic$ and N_{mic}/TN ($p < 0.05$) where both $qMic$ explained about 76% variability in N_{mic}/TN . Also, the negative correlation between C_{mic} (58%) and N_{mic} (61%) with qCO_2 was statistically significant ($p < 0.01$). Finally, A significant negative correlation was found between C_{mic}/N_{mic} and N_{mic}/TN ($p < 0.05$ with $r = 74\%$) (Table 4).

The chemical and biochemical properties of soils interact in ways that can either enhance or hinder each other⁸. Since microbial stoichiometric indices are influenced by these properties, strong correlations between them are expected. For example, Sharma et al.⁶¹ reported a significant relationship between microbial biomass C and microbial biomass N ($r = 0.869$, $p < 0.0005$), a finding supported by Babur et al.²¹ ($r = 0.580$, $p < 0.05$). Our study corroborates these observations. Microbial biomass and activity are generally highest in topsoil, where organic material is abundant⁵⁸. This is largely due to favorable climatic conditions and the higher nutrient availability in these layers²¹. However, as microbial populations increase, so does competition for nutrients, leading to elevated microbial activity and respiration^{12,28}. This increased metabolic demand can cause higher qCO_2 levels, reflecting a stressed environment. Moreover, microorganisms must compete with plant roots for limited resources, which further exacerbates energy expenditure and soil stress²⁸.

The principal component analysis (PCA; Fig. 3) exhibits the relationships between microbial communities and their stoichiometric indices across three land uses where the first two principal components explain 57.5% and 22.8% of the total variability, respectively. The first Dim (Dim1) explains about 57.5% of the variability in the data, likely indicating a gradient of microbial activity and carbon utilization efficiency which perhaps led by a positive correlation with qCO_2 , $qMic$, and N_{mic}/TN , and negative correlations with C_{mic}/N_{mic} , MR, C_{mic} , and N_{mic} . The second Dim (Dim 2) explains about 22.8% variability in data, perhaps reflecting a gradient of nitrogen availability and a positive correlation with N_{mic} and N_{mic}/TN and countered by C_{mic}/N_{mic} and qCO_2 . Perhaps due to environmental stressors, riparian soils appear to have higher qCO_2 , $qMic$, and N_{mic}/TN and clustered on the positive side of the Dim 1. However, pasture soils clustered on the negative side of Dim 1 and Dim 2, having high C_{mic}/N_{mic} . PCA also shows a higher response of C_{mic} and N_{mic} to the forest. These patterns suggest that Dim 1 (PC1) captures ecological shifts in microbial efficiency and activity, while Dim 2 (PC2) reflects nitrogen dynamics. The principal component analysis (PCA) results (Fig. 3) also shows 80.3% of the total variation along with the two principal components. Changes in land use had a significant impact on the ability to characterize soil microbial properties and stoichiometric indices⁶². These indices driven by riparian areas appear to be qCO_2 , $qMic$, and N_{mic}/TN where $qMic$ is the highest affected property. In addition, C_{mic}/N_{mic} was the main driver of pastureland. Those of C_{mic} , N_{mic} , C/N, OC, and MR are drivers of the forest. The negative correlation of qCO_2 with C_{mic} and N_{mic} played a critical role in differentiating the microbial properties of forests in comparison to riparian areas. On the other hand, a distinct association was observed between C_{mic}/N_{mic} and N_{mic}/TN , which differentiates soils from pasture areas and riparian zones.

Conclusions

The results of this study demonstrate that land use significantly influences soil microbial properties and stoichiometric indices. In tree-based systems, the increased organic matter input enhances soil organic carbon, promoting better soil health and productivity, which in turn supports greater microbial diversity. Forest soils exhibited the highest microbial biomass carbon and the lowest microbial coefficient, whereas riparian areas had

	MR	C_{mic}	N_{mic}	$qMic$	qCO_2	C_{mic}/N_{mic}	N_{mic}/TN
MR	1						
C_{mic}	,914 (**)	1					
N_{mic}	,823 (**)	,956 (**)	1				
$qMic$				1			
qCO_2		-,577 (*)	-,614 (*)		1		
C_{mic}/N_{mic}	,724 (**)	,579 (*)				1	
N_{mic}/TN				,759 (**)		-,737 (**)	1

Table 4. Pearson's correlation analysis for microbial properties and microbial stoichiometric indices

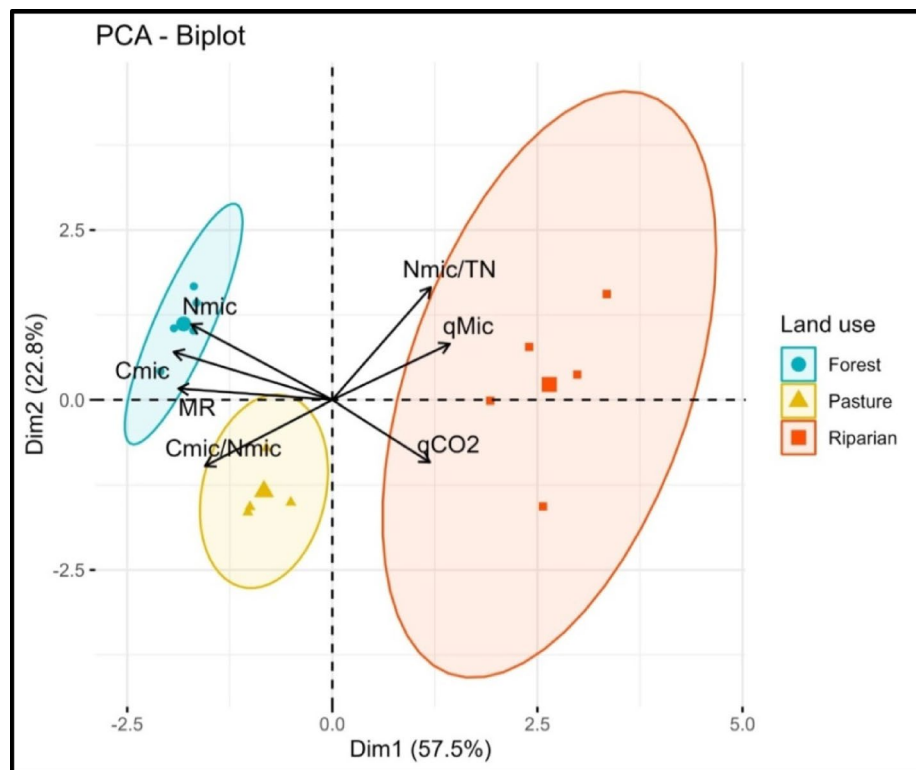


Fig. 3. Principle component analysis of microbial properties and stoichiometric indices.

the lowest values. This can be attributed to the fact that organic carbon in riparian zones is often washed into rivers and lakes, leading to nutrient-poor conditions. The low SOC and MBC in riparian areas, coupled with the lack of organic matter inputs and frequent flooding, result in poor aeration, which likely reduces soil fertility and microbial activity. When humidity, temperature, and nutrient levels are adequate, microbial biomass and activity are expected to increase. However, in riparian zones, excessive soil water and nutrient leaching contribute to diminished microbial communities and activity. These study results emphasize the need for strategic land management practices to increase soil organic matter, mitigate nutrient leaching, and enhance soil health and microbial activity in riparian zones in the Mediterranean basin. In addition, the results obtained indicate the need to improve management practices, especially in watersheds, preserve forest vegetation, and protect and enhance adjacent regional ecosystems.

Data availability

All data generated or analyzed during this study are included in this published article (and its Supplementary Information files).

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E.B.: Conceptualization, Investigation, Methodology, Data curation, Software, Visualization, Writing—Original, Draft, Writing—review and editing. E.O.: Investigation, Normal analysis, Methodology, Data collection, Software, Writing—Original Draft, Writing—review and editing. O.S.U.: Software, Validation, Methodology, Visualization, Software, Writing—Original, Draft, Writing—review and editing.

Declarations

Competing interests

The authors declare no conflicts of interest.

Additional information

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