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Response of Absolute and Specific Enzyme Activities in Clay Aridisol from Arid Monoculture Areas

Kurak Monokültür Alanlarından Alınan Killi Aridisol Topraklarda Mutlak ve Spesifik Enzim Aktivitelerinin Tepkisi

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Abstract: Soil enzyme activities play a key role in nutrient cycling, organic matter decomposition, and soil health, and are closely related to soil organic carbon (SOC) and microbial biomass carbon (Cmic). However, limited information is available on how absolute enzyme activities (AEA) and specific enzyme activities (SEA) respond to alkaline soil conditions. This study evaluated AEA and SEA together with CO_2-C, qCO_2, Cmic, Nmic, total C and N, and selected soil properties. Catalase (CAT) activity was highest in summer at Kocapınar (87.0 mL O_2 g^{-1} dry soil 5 min^{-1}), dehydrogenase (DHG) activity in spring at Yalıntepe (30 µg TPF g^{-1}), and urease activity in summer at Kocapınar (37 µg N g^{-1}). Cmic reached its maximum in spring at Yalıntepe (1224 mg C kg^{-1}). The Cmic:Corg ratio peaked in autumn (0.15), while the Cmic:Nmic ratio was highest in summer (69.54). Nmic (40 mg N kg^{-1}) and the Nmic:NT ratio (0.51) were highest in spring at Kocapınar. NH_4 content was highest in spring at Katran (101.29 mg kg^{-1}), whereas NO_3 content peaked in autumn (193.03 mg kg^{-1}). Microbial respiration (3.03 mg C kg^{-1} h^{-1}) and qCO_2 (1.83 mg CO_2-C g^{-1} Cmic h^{-1}) were highest in spring at Yalıntepe. Among SEA indices, CAT/C was highest in summer at Kocapınar (110), urease/C in summer at Yalıntepe (46), and DHG/C in winter at Yalıntepe (38). Overall, AEA and SEA proved effective indicators of soil quality in relation to microbial activity and environmental conditions.

Keywords: Specific enzyme activity, Absolute enzyme activity, Nutrient cycling, Microbial biomass carbon, Microbial biomass nitrogen

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Öz: Toprak enzim aktiviteleri; besin döngüsü, organik maddenin ayrışması ve toprak sağlığının sürdürülmesinde temel rol oynayarak toprak organik karbonu (SOC) ve mikrobiyal biyokütle karbonu (Cmic) ile yakından ilişkilidir. Ancak mutlak enzim aktiviteleri (AEA) ile spesifik enzim aktivitelerinin (SEA) alkali toprak koşullarına tepkisini inceleyen çalışmalar sınırlıdır. Bu araştırmada AEA ve SEA'nin yanı sıra CO_2-C, qCO_2, Cmic, Nmic, toplam C ve N ile çeşitli toprak özellikleri değerlendirilmiştir. Katalaz (CAT) aktivitesi yaz mevsiminde Kocapınar'da (87.0 mL O_2 g^{-1} kuru toprak 5 dk^{-1}), dehidrogenaz (DHG) aktivitesi ilkbaharda Yalıntepe'de (30 µg TPF g^{-1}) ve üreaz aktivitesi yazın Kocapınar'da (37 µg N g^{-1}) en yüksek düzeylere ulaşmıştır. Cmic ilkbaharda Yalıntepe'de (1224 mg C kg^{-1}) maksimum bulunurken, Cmic:Corg oranı sonbaharda (0.15) ve Cmic:Nmic oranı yazın (69.54) en yüksek değerleri göstermiştir. Nmic ve Nmic:NT oranı ilkbaharda Kocapınar'da sırasıyla 40 mg N kg^{-1} ve 0.51 olarak belirlenmiştir. NH_4 ilkbaharda Katran'da (101.29 mg kg^{-1}), NO_3 ise sonbaharda (193.03 mg kg^{-1}) en yüksek seviyededir. Mikrobiyal solunum (3.03 mg C kg^{-1} $saat^{-1}$) ve qCO_2 (1.83 mg CO_2-C g^{-1} Cmic $saat^{-1}$) ilkbaharda Yalıntepe'de maksimumdur. SEA sonuçları, CAT/C oranının yazın Kocapınar'da (110), üreaz/C oranının yazın Yalıntepe'de (46) ve DHG/C oranının kışın Yalıntepe'de (38) en yüksek olduğunu göstermiştir. Bulgular, AEA ve SEA'nin mikrobiyal aktivite ve çevresel faktörlerle bağlantılı olarak toprak kalitesinin değerlendirilmesinde güvenilir göstergeler olduğunu ortaya koymaktadır.

Anahtar Kelimeler: Spesifik enzim aktivitesi, Mutlak enzim aktivitesi, Besin döngüsü, Mikrobiyal biyokütle karbonu, Mikrobiyal biyokütle azotu

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INTRODUCTION

Plant species play a crucial role in ecosystem processes, influencing nutrient cycling, microbial biomass, soil structure, and enzyme activity (Thijs et al., 2016). These plant-soil-microbe interactions drive key biochemical and physical changes in the soil, including shifts in pH, moisture, temperature, and nutrient availability (Zhao et al., 2022). Such changes significantly impact microbial activity and biomass, thereby affecting soil functionality.

Seasonal variations strongly influence soil organic matter dynamics by regulating soil moisture, temperature, root activity, and plant biomass decomposition (Zhao et al., 2022). However, these effects vary depending on soil type, vegetation characteristics, land use, and management practices (Evers et al., 2018). The most sensitive indicators of these changes are soil health parameters, which govern biological productivity, water and air regulation, and overall ecosystem stability (Doran and Safley, 1997). Since soil health indices respond rapidly to environmental changes, metabolic activity is a key factor in assessing soil quality. Both biotic and abiotic components influence these indices, and while specific soil health assessment methods vary globally, several widely accepted ratio indices are used for evaluation.

Metabolic activity parameters provide a comprehensive assessment of soil health by integrating biotic and abiotic factors. For example, the microbial biomass carbon-to-organic carbon ratio (Cmic/Corg) reflects the microbial contribution to carbon cycling (Insam and Domsch, 1988). Soil respiration (CO_2-C) is an important indicator of microbial and root activity (Nielsen et al., 2002; Sakin, 2016), while the metabolic quotient (qCO_2) measures microbial respiration efficiency and stress responses (Maini et al., 2020; Sparling, 1997). Additionally, the microbial biomass carbon-to-nitrogen ratio (Cmic/Nmic) helps identify the dominant microbial groups—bacteria, fungi, or actinomycetes—within the microbial community, offering insight into microbial structure and function (Simfukwe et al., 2021). The organic carbon-to-total nitrogen ratio (Corg/Nt) serves as an index of organic matter decomposition and weight loss (Piotrowska-Długosz and Wilczewski, 2024).

Environmental factors, particularly soil temperature and moisture, regulate microbial cycles and enzyme activity. Microbial activity is significantly affected by extreme seasonal conditions, such as summer drought and winter freezing, which limit microbial functions. Conversely, wetting and thawing events enhance microbial growth and accelerate the decomposition of organic matter. Seasonal changes also influence carbon mineralization rates, microbial community composition, and soil nutrient availability (Li et al., 2023). However, microbial biomass fluctuations are not solely driven by climate; plant species and root system characteristics also contribute to these variations (Panikov, 1999). Studies indicate that seasonal shifts in microbial activity are more pronounced in organic-rich soils than in mineral soils, likely because active microbial communities are more sensitive to environmental changes (Viscarra Rossel et al., 2019). Compared to absolute enzyme activities of soil organic carbon (SOC) or microbial biomass carbon (MBC) provide a more standardized assessment of soil microbial functions, enabling accurate comparisons across different soil management practices. These metrics offer ecological insights into the metabolic state of microbial communities and extracellular enzyme activity variations (Raiesi and Beheshti, 2014). Since enzyme activities directly result from active microbial communities, they also serve as indicators of microbial physiological capacity and ecosystem stability (Waldrop et al., 2000).

Given the region's ecological importance and the limited understanding of microbial dynamics under semi-arid conditions, this study investigates the effects of dry monoculture systems and seasonal variability on key biochemical parameters and microbial health indicators — including enzyme activities (CAT, DHG, urease), carbon and nitrogen fractions (SOC, Nt, Cmic, Nmic and their ratios), microbial respiration (CO_2-C), and the metabolic quotient (qCO_2). We hypothesize that seasonal variation and monoculture-driven land use will significantly alter microbial activity and enzyme functions, with drier seasons suppressing microbial biomass and shifting community-level metabolic efficiency, as reflected by elevated qCO_2 values.

MATERIAL AND METHOD

Material

Study Area and Soil Sampling

This study was conducted between 2022 and 2023 in Katran, Kocapınar, and Yalıntepe villages in the Cizre district of Şırnak, Turkey, where traditional agriculture is practiced. Soil samples were collected from lentil fields in these villages at the following geographical coordinates: Katran village (X: 37.2511054, Y: 42.111194), Kocapınar village (X: 37.2929109, Y: 42.066452), and Yalıntepe village (X: 37.2773439, Y: 42.0508928). The study area is illustrated in Figure 1.

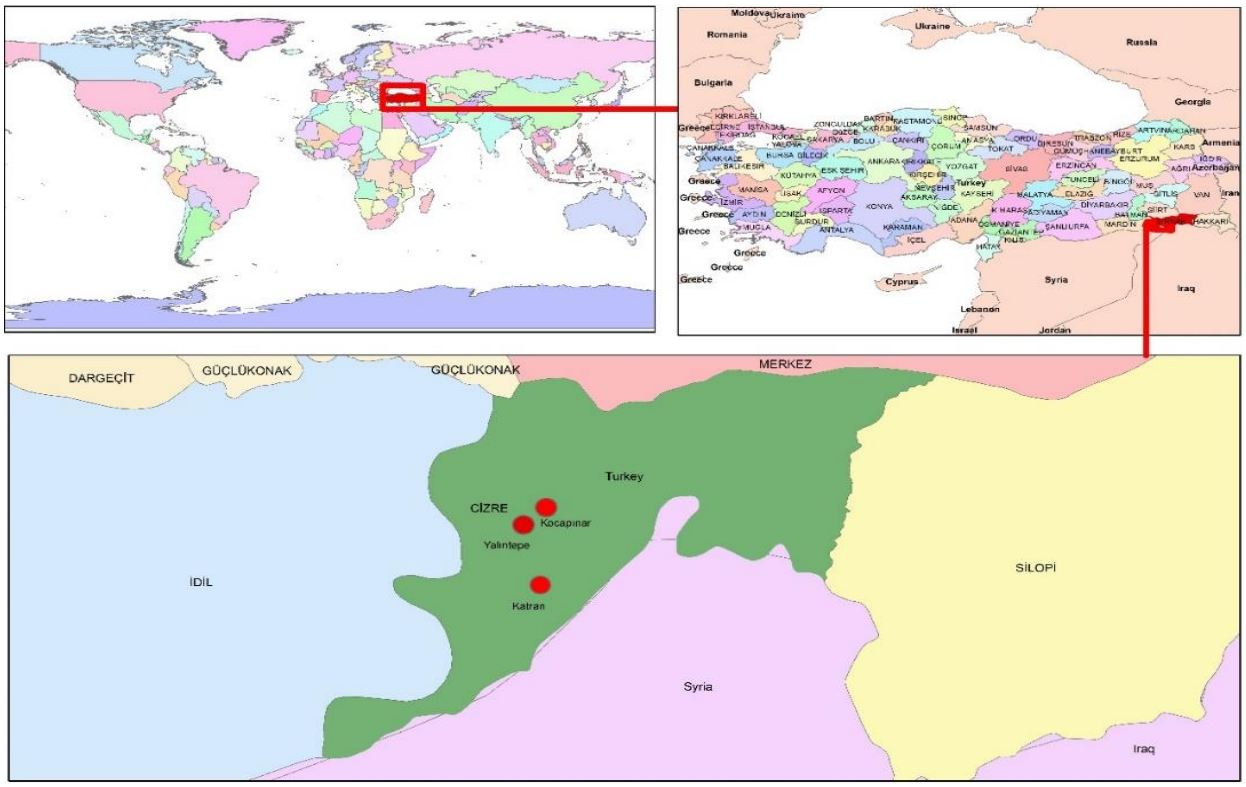

Figure 1. The geographical location of the experimental site at the country and city level.
Şekil 1. Deneme alanının ülke ve il (şehir) düzeyindeki coğrafi konumu.

Soil samples were collected from the cultivated soil of lentil (Lens culinaris Medik.) fields in these villages at a depth of 0–30 cm during spring, summer, autumn, and winter. After collection, the samples were sieved through a 2 mm sieve in the field, stored in plastic bags, and transported to the laboratory. Upon arrival, the samples were stored at 4 °C until further analysis. The general physicochemical properties of the soils are presented in Table 1.

Table 1. Physicochemical properties of soils in the study area.
Çizelge 1. Çalışma alanındaki toprakların fizikokimyasal özellikleri.

Location	pH (1:2.5 w/v)	EC (dS cm-1)(1:5 w/v)	Class texture	Lime (%)	C (%)
Katran	8.1	0.93	C	6.72	0.81
Yalıntepe	8.0	0.85	C	1.72	0.80
Kocapınar	8.0	0.70	C	5.51	0.80

Methods

Basic Soil Analyses

Soil texture was determined using the Bouyoucos hydrometer method (Gee and Bauder, 1986). Soil pH (1:2.5 w/v) and electrical conductivity (EC, 1:5 w/v) were measured in deionized water (Nelson and Sommers, 1982). SOC was analysed using the potassium dichromate wet oxidation method (Nelson and Sommers, 1982), while total nitrogen (Nt) was determined by the Kjeldahl method (Bremner and Mulvaney, 1982). Lime content was measured using the Scheibler calcimeter method (Allison and Moodie, 1965).

For mineral nitrogen (NH_4^+ and NO_3^-) analysis, soil samples were extracted with 2 M KCl solution (1:4 w/v) and shaken for 1 hour. The extracts were filtered through Whatman 602 filter paper and stored at -20 °C until analysis. Ammonium (NH_4^+) concentration was determined spectrophotometrically at 650 nm (Baethgen and Alley, 1989), and nitrate (NO_3^-) concentration was measured at 410 nm (Yang et al., 1998).

Soil Enzyme Analyses

DHG activity was determined by incubating soil samples with triphenyl tetrazolium chloride (TTC) at 25°C for 24 hours, followed by spectrophotometric measurement of triphenyl formazan (TPF) at 485 nm (Skujinš, 1973). Urease activity was measured spectrophotometrically at 578 nm after incubation at 37°C for 1 hour (Hoffmann and Teicher, 1961). Catalase activity was assessed by adding 10 mL phosphate buffer (pH 7) and 5 mL of 3% H_2O_2 substrate to 5 g of soil, and measuring the volume of O_2 released over 5 minutes at 20°C (Beck, 1971).

Soil Microbial Analyses

Microbial Biomass Carbon (Cmic) and Nitrogen (Nmic)

Microbial biomass carbon (Cmic) was determined using the fumigation-extraction method (Vance et al., 1987), calculated according to Equation 1:

$$\mathrm{MBC}\ (\mathrm{mg.\,C.\,kg^{-1}}) = (\mathrm{C_F} - \mathrm{C_{NF}})\mathrm{X}2.64 \qquad (1)$$

where:

C_F = organic C from fumigated soil

C_{NF} = organic C from non-fumigated soil

2.64 = proportionality factor for biomass C release

Microbial biomass nitrogen (Nmic) was determined using K_2SO_4 extraction (Amato and Ladd, 1988; Joergensen and Brookes, 1990), calculated using Equation 2:

$$MBN\ (mg.N.kg^{-1}) = (N_F - N_{NF})X0.45 \qquad (2)$$

where:

N_F = N extracted from fumigated soil

N_{NF} = N extracted from non-fumigated soil

0.45 = proportionality factor for biomass N release

Microbial Respiration

Microbial respiration was assessed by trapping CO_2 in 1 M NaOH (25 mL) and titrating with 0.5 M HCl (Anderson, 1982). Soil samples (50 g, oven-dry basis) were placed in 100 mL glass jars and incubated in 1 L airtight glass containers with 25 mL NaOH and 10 mL deionized water. After incubation, NaOH was mixed with 5 mL water and $BaCl_2$, then titrated with 0.5 M HCl using Equation 3:

$$CO_2 - C\ (mg\ C\ kg^{-1}\ h^{-1}) = \left(V_{blank} - V_{sample}\right)\ x\ M_{HCl}\ x\ 22/(W_{dry}\ x\ t) \quad (3)$$

where:

V_{blank}; Volume of HCl used for blank titration (mL), V_{sample}; Volume of HCl used for sample titration (mL), M; HCl molarity of HCl (0.5 M), 22; Equivalent weight of CO_2-C (mg meq^{-1}), W_{dry}; Oven-dry soil weight (kg), t; Incubation period (days translate to hour)

2.5 Microbial Metabolic Quotient (qCO_2)

The metabolic quotient (qCO_2) was calculated following Anderson and Domsch (1990) using Equation 4:

$$qCO_2 = \left(\frac{Basal\ respiration\ (mg\ CO_2 - C\ dry\ soil h^{-1}}{C_{mic}(mgC\ g^{-1}\ dry\ soil)}\right) \quad (4)$$

where qCO_2 is expressed in mg CO_2-C mg^{-1} Cmic h^{-1}.

The Cmic:Corg ratio was calculated following Anderson and Domsch (1989) and is widely used as an indicator of soil biological quality and microbial efficiency in soil carbon utilization with using Equation 5:

Cmic:Corg (%)= Cmic/Corg*100 (5)

Specific and Absolute Enzyme Activities

Specific enzyme activity (SEA) and absolute enzyme activity (AEA) were calculated based on García-Ruiz et al. (2008) and Kobierski et al. (2020) using Equations 6 and 7:

$$Specific\ enzyme\ activity\ (SEA\ per\ Cmic)\ =\ \frac{EA}{C_{mic}} \quad (6)$$

$$Absolute\ enzyme\ activity\ (AEA)\ =\ \frac{EA}{Wdry} \quad (7)$$

where EA is the enzyme activity (µg substrate g^{-1} dry soil h^{-1}), Cmic is the microbial biomass carbon (mg C g^{-1} dry soil), and SOC is the soil organic carbon (mg C g^{-1} dry soil).

Statistical Analysis

All data were analysed using SPSS 21. One-way ANOVA was performed, and significant differences between means were determined using Duncan's Multiple Range Test ($P \leq 0.05$). Pearson correlation coefficients were calculated to examine relationships between parameters. The final results were visualized using appropriate graphs and statistical plots (Lê et al., 2008).

RESULTS AND DISCUSSION

Results

Seasonal Variation of Soil Enzymes

The seasonal dynamics of catalase (CAT), dehydrogenase (DHG), and urease enzyme activities across different locations are presented in Figure 2. Enzyme activities varied significantly between locations and across seasons ($P < 0.05$).

During the summer, CAT activity was highest at Kocapınar (87.0 mL O_2 g^{-1} dry soil 5 min^{-1}) and lowest at Katran (15 mL O_2 g^{-1} dry soil 5 min^{-1}). DHG activity peaked in spring at Yalıntepe (30 µg TPF g^{-1} dry $soil^{-1}$) but dropped to its lowest in autumn at the same location (18 µg TPF g^{-1} dry $soil^{-1}$). Urease activity exhibited a similar trend, reaching its maximum (37 µg N g^{-1} dry $soil^{-1}$) in summer at Kocapınar and the lowest (6 µg N g^{-1} dry $soil^{-1}$) in autumn at Yalıntepe.

These findings suggest that seasonal temperature and moisture variations strongly influence enzymatic activities, with peaks generally occurring in warmer months, potentially due to enhanced microbial activity and organic matter decomposition. Conversely, lower temperatures and reduced substrate availability in autumn and winter may have contributed to the observed declines.

Seasonal Variation of Cmic, Cmic:Corg, Cmic:Nmic Contents

The seasonal variations in Cmic, Cmic:Corg, and Cmic:Nmic across different locations are illustrated in Figure 3. All three parameters showed significant differences between locations and seasons ($P < 0.05$).

Microbial biomass carbon (Cmic) content was highest in spring at Yalıntepe (1224 mg C kg^{-1}) and lowest in spring at Kocapınar (549 mg C kg^{-1}). The Cmic:Corg ratio, an indicator of microbial efficiency in utilizing soil organic carbon, was highest (0.15) at Yalıntepe in autumn and lowest (0.07) at Kocapınar in spring. The Cmic:Nmic ratio, which reflects microbial community composition and nitrogen availability, peaked in summer at Yalıntepe (69.54) and reached its lowest value (13.71) in spring at Kocapınar.

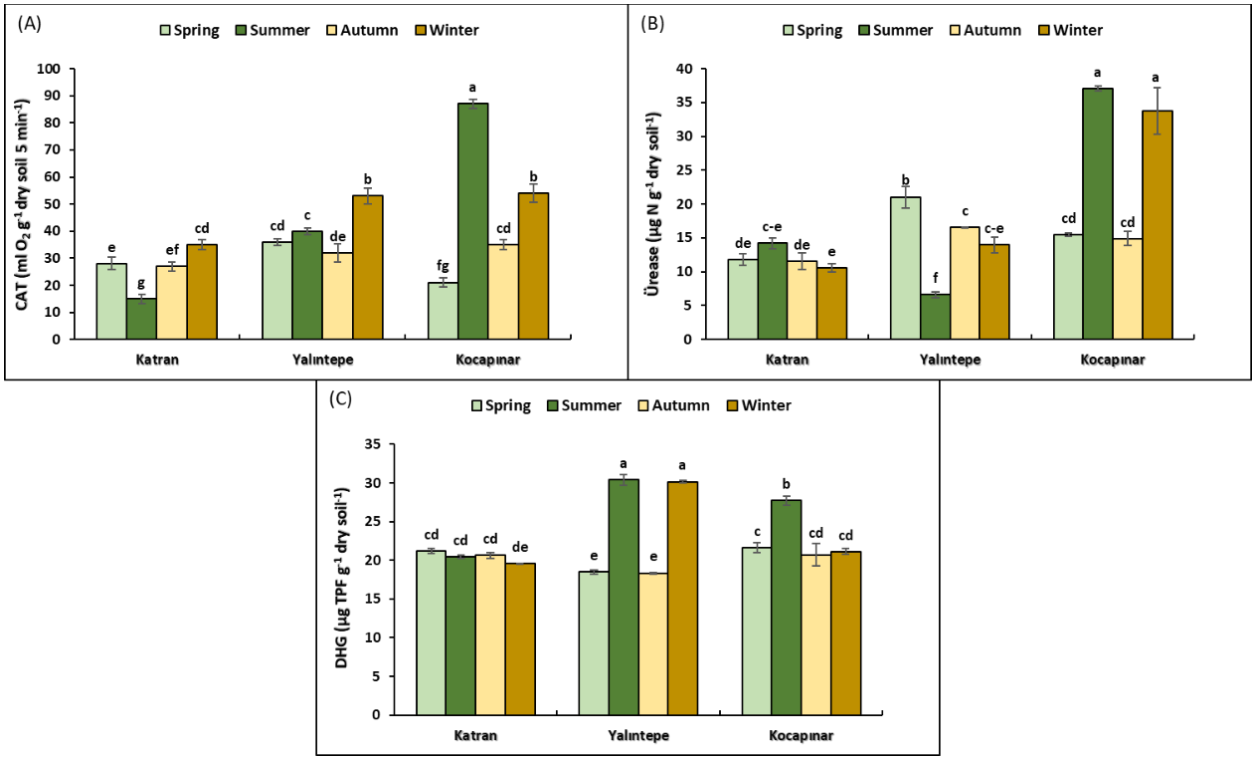

Figure 2. Catalase [CAT (a)], Urease (b), and Dehydrogenase [DHG (c)] activities across different seasons and locations (Mean ± S.E.). Different letters indicate significant differences ($P < 0.05$) among means within each variable.
Şekil 2. Farklı mevsimler ve lokasyonlar boyunca katalaz [CAT (a)], üreaz (b) ve dehidrogenaz [DHG (c)] aktiviteleri (Ortalama ± S.H.). Her bir değişken için ortalamalar arasındaki istatistiksel olarak anlamlı farklılıklar farklı harflerle gösterilmiştir (P < 0.05).

These results indicate that microbial biomass and efficiency fluctuate seasonally, likely due to temperature, moisture availability, and substrate supply. The higher Cmic content in spring suggests that microbial communities thrive when moisture availability and moderate temperatures favor microbial proliferation. In contrast, the decline in autumn and winter may be attributed to reduced microbial activity under lower temperatures and drier conditions. The high Cmic:Nmic ratio in summer suggests a shift in microbial

community structure, potentially favoring fungal dominance, which typically exhibits higher C:N ratios than bacteria.

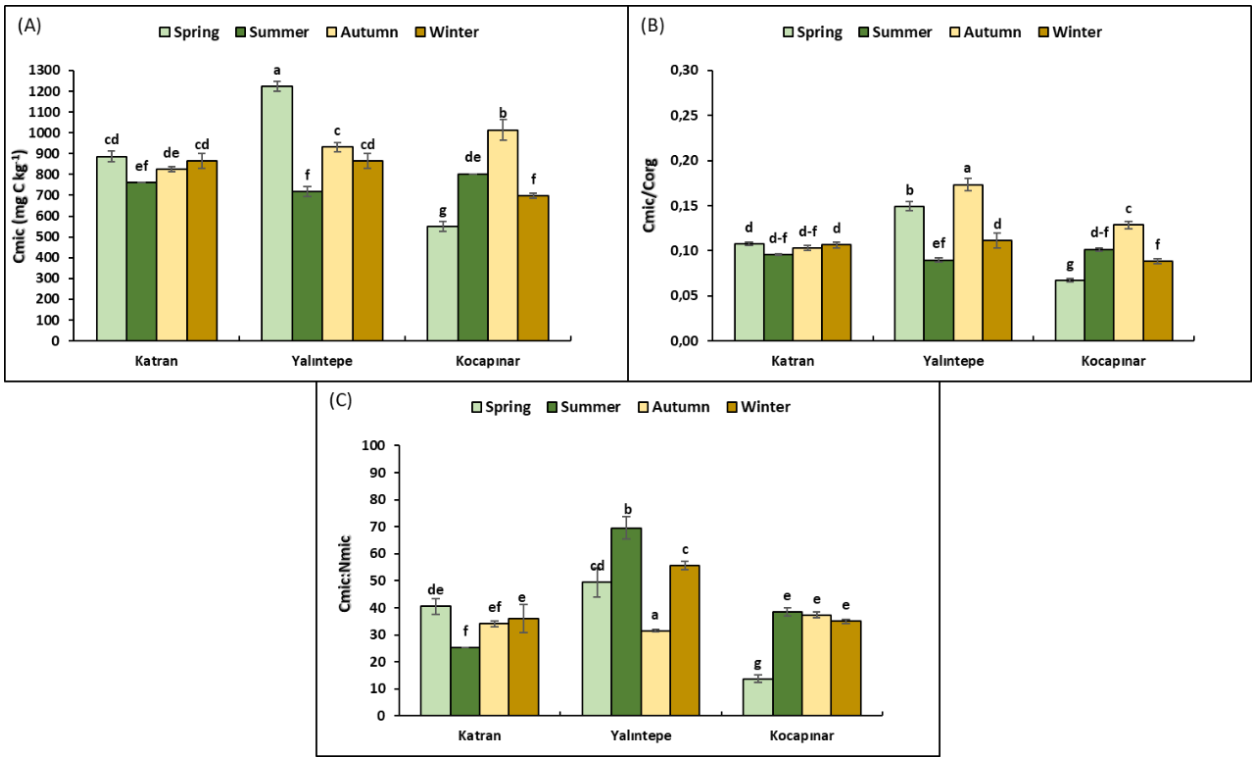

Figure 3. Cmic (a), Cmic/Corg (b) and Cmin:Nmic (c) effects of different seasons on soils of different land (Mean ± S.E.). Different letters on bars exhibit significant difference between means within each variable.
Şekil 3. Farklı arazi kullanımına sahip topraklarda, farklı mevsimlerin Cmic (a), Cmic/Corg (b) ve Cmin:Nmic (c) üzerine etkileri (Ortalama ± S.H.). Her bir değişken için ortalamalar arasındaki istatistiksel olarak anlamlı farklılıklar sütunlar üzerindeki farklı harflerle gösterilmiştir.

Seasonal Variation of Nmic, Nmic:NT, NH_4 and NO_3 Contents

The seasonal and locational variations in microbial biomass nitrogen (Nmic), the Nmic:NT ratio, ammonium (NH_4^+), and nitrate (NO_3^-) contents are presented in Figure 4. Significant differences ($P < 0.05$) were observed in all parameters across seasons and locations.

Nmic content was highest in spring at Kocapınar (40 mg N kg^{-1}) and lowest in summer at Yalıntepe (10 mg N kg^{-1}). The Nmic:NT ratio, which reflects the proportion of microbial nitrogen relative to NT, was highest in spring at Kocapınar (0.51) and lowest in summer at Yalıntepe (0.014). NH_4^+ content peaked in spring at Katran (101.29 mg kg^{-1}) and was lowest in summer at Kocapınar (16.94 mg kg^{-1}). Similarly, NO_3^- content reached its highest level in autumn at Katran (193.03 mg kg^{-1}) and its lowest in autumn at Kocapınar (89.78 mg kg^{-1}).

These findings suggest that spring conditions favor microbial nitrogen accumulation, likely due to increased plant root activity and microbial mineralization. In contrast, summer depletion of Nmic and NH_4^+ may result from higher microbial respiration rates, increased nitrogen uptake by plants, and volatilization losses under warmer temperatures. The elevated NO_3^- content in autumn suggests a possible accumulation due to reduced plant uptake and enhanced nitrification processes during this period.

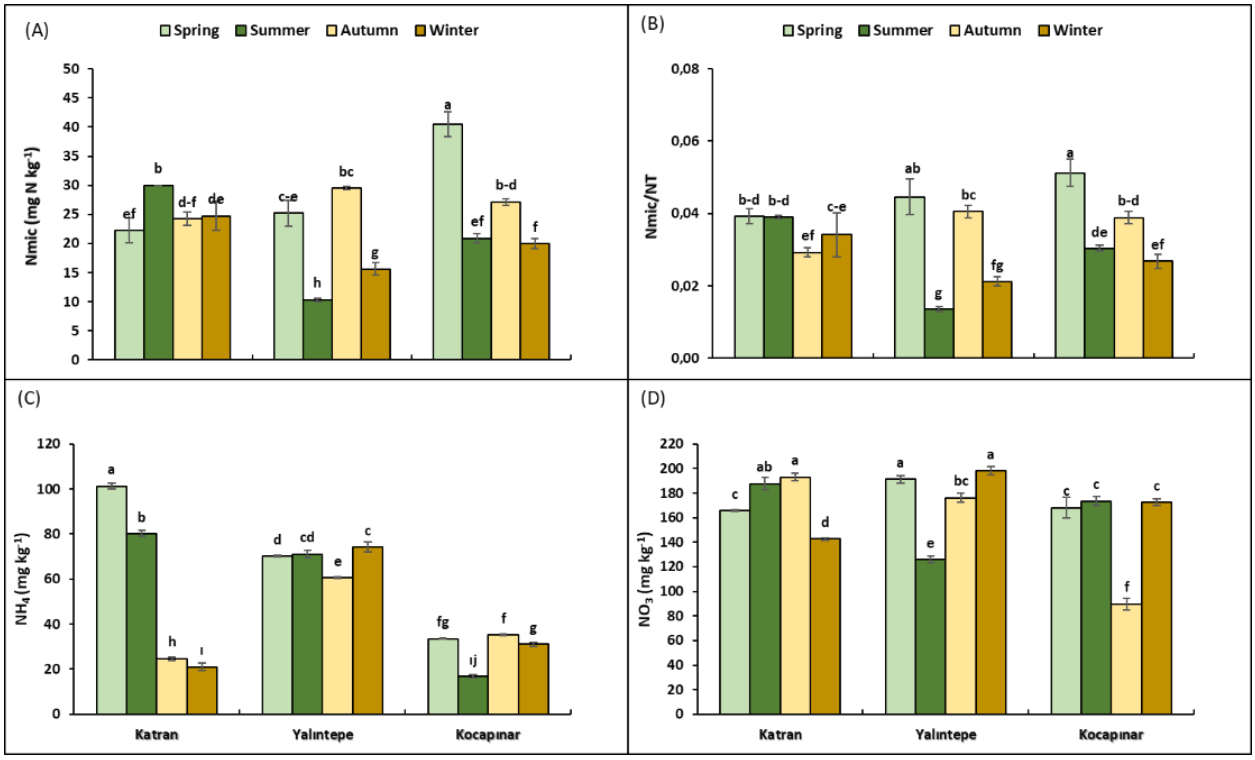

Figure 4. Nmic (a), Nmic/NT (b), Ammonium NH_4 (c) and Nitrate [NO_3 (d)] effects of different seasons on soils of different land (Mean ± S.E.). Different letters on bars exhibit significant difference between means within each variable.
Şekil 4. Farklı arazi kullanımına sahip topraklarda, farklı mevsimlerin Nmic (a), Nmic/NT (b), Amonyum NH_4 (c) ve Nitrat [NO_3 (d)] üzerine etkileri (Ortalama ± S.H.). Her bir değişken için ortalamalar arasındaki istatistiksel olarak anlamlı farklılıklar sütunlar üzerindeki farklı harflerle gösterilmiştir.

Seasonal Variation of %C and %N and C:N Ratios

The seasonal and locational variations in total carbon (%C), total nitrogen (%N), and C:N ratios are illustrated in Figure 5. While %N content and C:N ratios exhibited significant variations across seasons and locations ($P < 0.05$), %C content remained statistically unchanged ($P > 0.05$).

The highest %N content was recorded in summer at Yalıntepe (0.08%), whereas the lowest was observed in spring at Katran (0.056%). The C:N ratio, a key indicator of organic matter decomposition and nitrogen availability, was highest in spring at Katran (14.66) and lowest in autumn at Katran (9.7).

These results indicate that seasonal changes influence nitrogen cycling more strongly than carbon levels, likely due to microbial decomposition, plant nitrogen uptake, and leaching losses. The higher C:N ratio in spring may reflect an accumulation of undecomposed organic matter, while the lower autumn C:N ratio suggests enhanced microbial decomposition and nitrogen mineralization during this period.

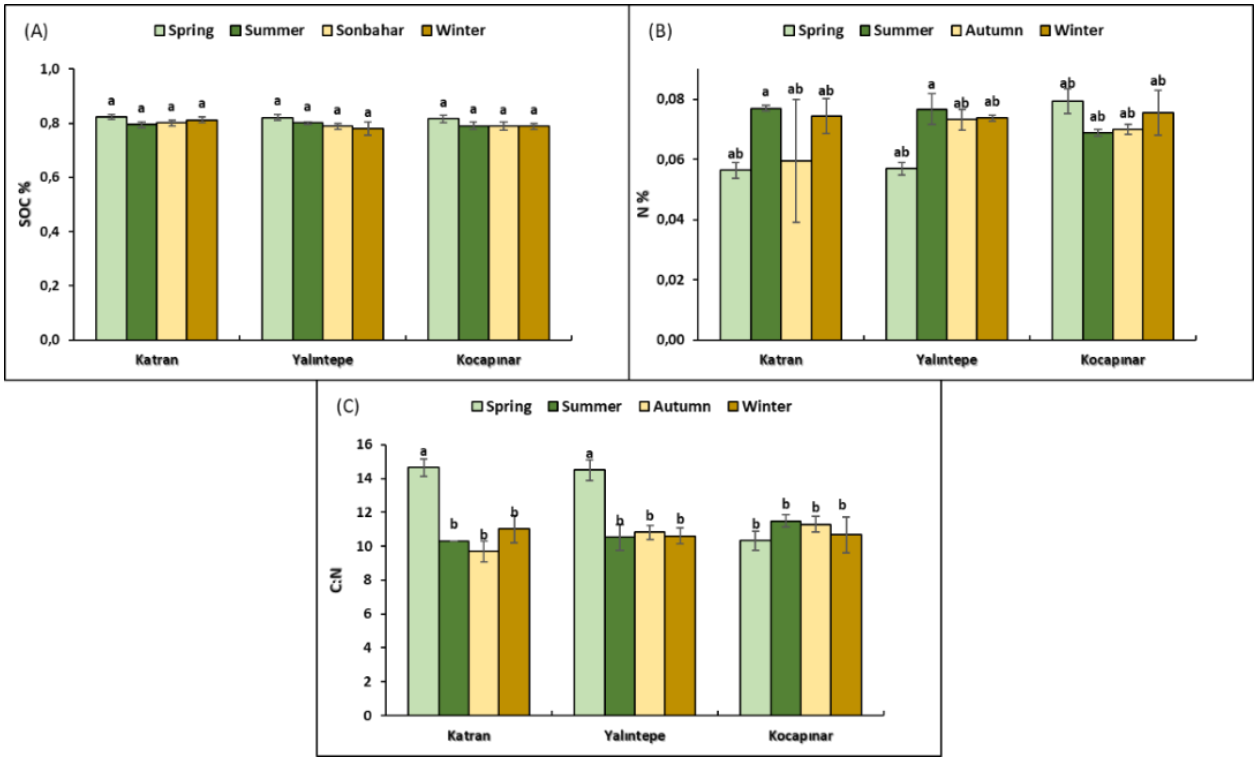

Figure 5. SOC% (a), N% (b) and C:N (c) effects of different seasons on soils of different land (Mean ± S.E.). Different letters on bars exhibit significant difference between means within each variable.
Şekil 5. Farklı arazi kullanımına sahip topraklarda, farklı mevsimlerin SOC% (a), N% (b) ve C:N (c) üzerine etkileri (Ortalama ± S.H.). Her bir değişken için ortalamalar arasındaki istatistiksel olarak anlamlı farklılıklar sütunlar üzerindeki farklı harflerle gösterilmiştir.

Seasonal Variation of Microbial Respiration (CO_2-C) and Microbial Metabolic Quotient (qCO_2) of Soil Samples

The seasonal variations in CO_2-C (microbial respiration) and qCO_2 (metabolic quotient) are presented in Figure 6. Both parameters exhibited significant differences ($P < 0.05$) between locations and seasons. The highest CO_2-C values were recorded in spring at Yalıntepe (3.03 mg CO_2-C kg^{-1} h^{-1}) and the lowest in autumn at Kocapınar (1.36 mg CO_2-C kg^{-1} h^{-1}). The qCO_2 values, an indicator of microbial efficiency and stress, were highest in spring at Yalıntepe (1.83 mg CO_2-C g^{-1} Cmic h^{-1}) and lowest in autumn at Kocapınar (0.16 mg CO_2-C g^{-1} Cmic h^{-1}).

These results suggest that microbial activity is highest in spring, coinciding with optimal soil moisture and moderate temperatures, while it declines in autumn and winter, likely due to reduced microbial substrate availability and lower temperatures. The lower qCO_2 values in autumn indicate a more efficient microbial community, whereas the higher values in spring may reflect stress conditions or a higher proportion of active microbial biomass, leading to increased respiration rates.

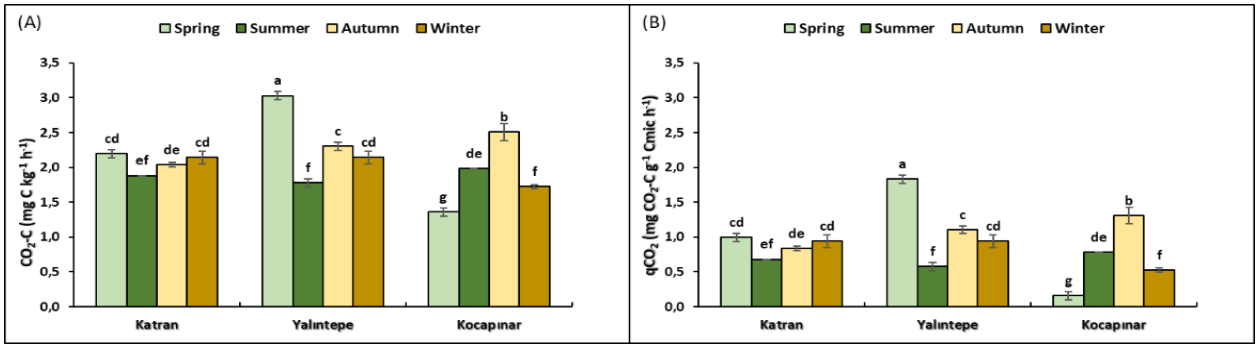

Figure 6. CO_2-C (a), and qCO_2 (b) effects of different seasons on soils of different land (Mean ± S.E.). Different letters on bars exhibit significant difference between means within each variable.
Şekil 6. Farklı arazi kullanımına sahip topraklarda, farklı mevsimlerin CO_2-C (a) ve qCO_2 (b) üzerine etkileri (Ortalama ± S.H.). Her bir değişken için ortalamalar arasındaki istatistiksel olarak anlamlı farklılıklar sütunlar üzerindeki farklı harflerle gösterilmiştir.

Seasonal Changes in Absolute and Specific Enzyme Activities

The seasonal variations in absolute and specific enzyme activities are presented in Figure 7. Both enzyme activity types varied significantly across seasons and locations ($P < 0.05$).

For absolute enzyme activities per unit SOC, the CAT/C ratio was highest in summer at Kocapınar (110 units) and lowest in summer at Katran (18 units). The Urease/C ratio was highest in summer at Yalıntepe (46 units) and lowest in winter at Katran (13 units). The DHG/C ratio was highest in winter at Yalıntepe (38 units) and lowest in spring at Yalıntepe (22 units).

These findings indicate that enzyme activities are highly responsive to seasonal conditions, with higher activity in warm, biologically active periods (spring and summer) and lower activity in colder seasons. The variations across locations may also reflect differences in soil organic matter content, microbial biomass, and environmental stressors.

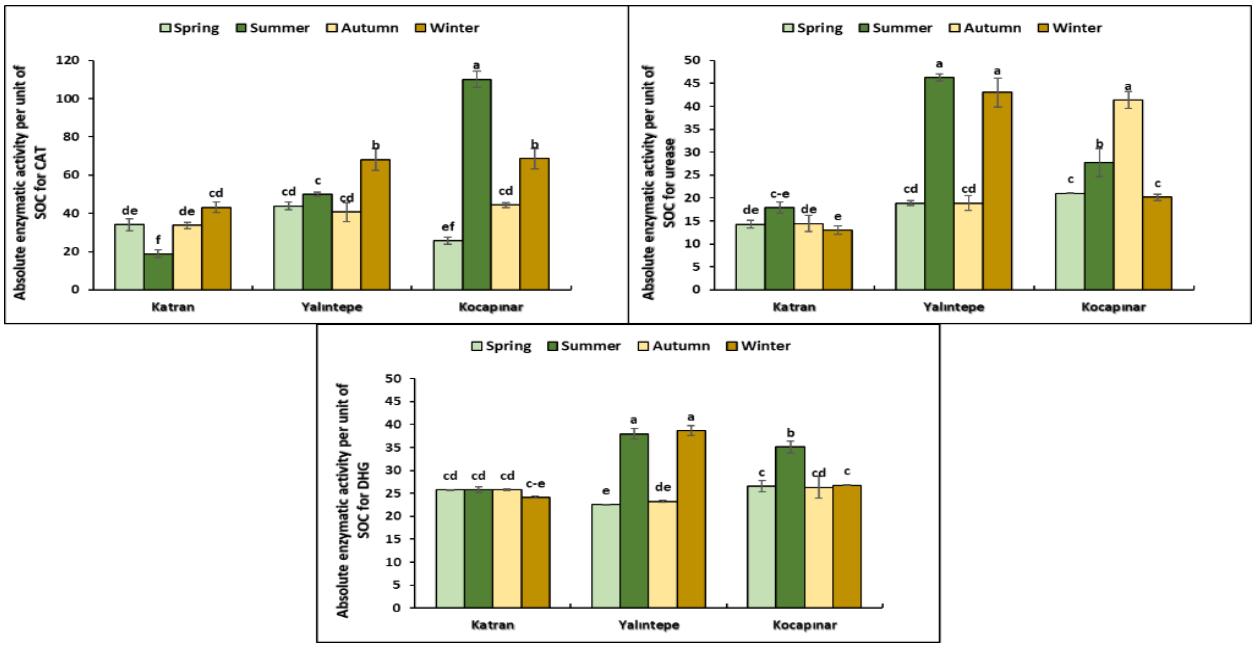

Figure 7. Effects of different seasons on soils absolute enzymatic per unit of different land (Mean ± S.E.). Different letters on bars exhibit significant difference between means within each variable.
Şekil 7. Farklı arazi kullanımına sahip topraklarda, farklı mevsimlerin birim toprak başına mutlak enzim aktiviteleri üzerindeki etkileri (Ortalama ± S.H.). Her bir değişken için ortalamalar arasındaki istatistiksel olarak anlamlı farklılıklar, sütunlar üzerindeki farklı harflerle gösterilmiştir.

For specific enzyme activities per unit Cmic (EA/Cmic), the CAT/Cmic ratio was highest in summer at Kocapınar (0.1 unit) and lowest in summer at Katran (0.02 unit). The Urease/Cmic ratio was highest in spring at Yalıntepe (0.5 units) and lowest in autumn at Yalıntepe (0.002 units). The DHG/Cmic ratio was highest in summer at Yalıntepe (0.04 units) and lowest in spring at Yalıntepe (0.015 units) (Figure 8).

These results suggest that specific enzyme activities per unit microbial biomass fluctuate in response to seasonal changes in microbial composition and activity levels. The high Urease/Cmic and DHG/Cmic ratios in spring and summer indicate enhanced microbial metabolic activity, while the lower values in autumn and winter suggest a reduced microbial capacity for enzymatic reactions under less favorable conditions.

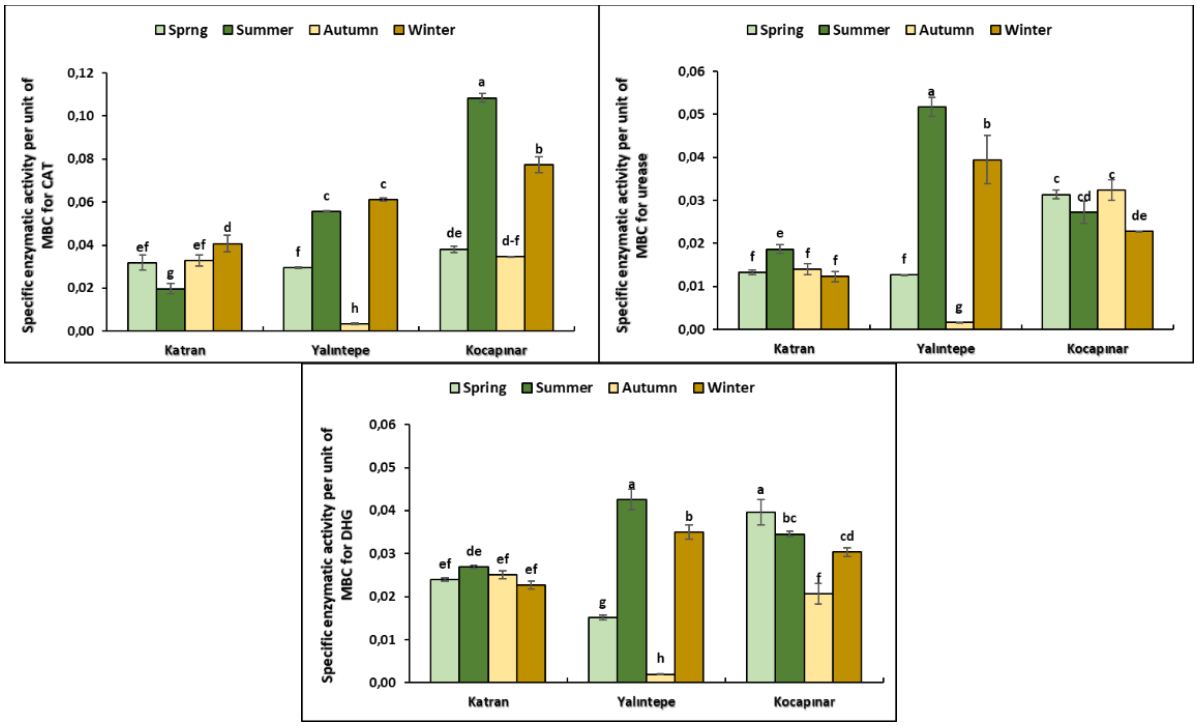

Figure 8. Effects of different seasons on soils of specific enzymatic per unit of MBC for DHG of different land (Mean ± S.E.). Different letters on bars exhibit significant difference between means within each variable.

Şekil 8. Farklı arazi kullanımına sahip topraklarda, farklı mevsimlerin mikrobiyal biyokütle karbonu (MBC) birimi başına spesifik dehidrogenaz (DHG) aktivitesi üzerindeki etkileri (Ortalama ± S.H.). Her bir değişken için ortalamalar arasındaki istatistiksel olarak anlamlı farklılıklar sütunlar üzerindeki farklı harflerle gösterilmiştir.

Discussion

Seasonal Variation of Soil Enzyme Activities

Soil enzymes are key biological indicators of soil quality, as they reflect microbial activity and are influenced by soil physicochemical properties, temperature, moisture, and vegetation type (Li et al., 2022). Seasonal fluctuations and land-use changes significantly impact soil enzymatic activity, as different environmental conditions promote or suppress microbial metabolic processes (Kong et al., 2023).

The high catalase (CAT) activity in summer at Kocapınar suggests that the soil in this region may be rich in organic matter and microbial biomass, which supports catalase production. This could be attributed to favorable moisture and temperature conditions, that enhance microbial responses to oxidative stress.

Conversely, low CAT activity at Katran may indicate reduced microbial activity due to limitations in nutrient availability, pH conditions, or organic matter content (Ning et al., 2024).

The highest DHG activity recorded in spring at Yalıntepe indicates high microbial respiration, as dehydrogenase enzymes are directly linked to microbial metabolic processes. Spring conditions, with optimal moisture and temperature, are likely to stimulate microbial growth, thereby enhancing dehydrogenase activity (Ning et al., 2024). In contrast, the lowest DHG activity in autumn at Yalıntepe may be due to decreased temperatures and soil moisture, which limit microbial metabolism (Kong et al., 2023).

Similarly, high urease activity in summer at Kocapınar suggests optimal conditions for microbial urease production and urea decomposition. This indicates high nitrogen availability and microbial biomass (Nmic), supported by organic matter decomposition and warm temperatures (Chen et al., 2024). Conversely, low urease activity at Yalıntepe in summer is likely due to low microbial biomass (Nmic) and moisture content, both of which regulate urease synthesis. Soil moisture and temperature fluctuations significantly influence urease activity, reflecting the seasonal dynamics of microbial nitrogen cycling (Kong et al., 2023).

Seasonal Variation of Cmic, Cmic:Corg, and Cmic:Nmic

The high Cmic content in spring at Yalıntepe is likely due to favorable soil temperature and moisture, which stimulate microbial growth and organic matter decomposition. Cooler wetter conditions in spring generally promote greater microbial biomass accumulation, leading to higher Cmic content. In contrast, lower Cmic values at Kocapınar in spring may result from drier conditions, which limit microbial activity (Evangelou and Giourga, 2024; Khan and Joergensen, 2006).

The Cmic:Corg ratio, an important indicator of microbial efficiency in utilizing organic carbon, peaked in autumn at Yalıntepe (0.173) and was lowest in spring at Kocapınar (0.07). The higher autumn ratio suggests that microbial decomposition of plant residues accelerated with rainfall, whereas the lower spring ratio indicates a slower microbial response to early seasonal changes. Previous studies have shown that microbial activity is typically slower in colder months, resulting in lower microbial turnover rates (Davidson and Janssens, 2006; Swift et al., 1979).

The high Cmic:Nmic ratio in summer at Yalıntepe reflects a seasonal shift in microbial metabolism, with higher temperatures (25–30°C) enhancing organic matter decomposition and increasing microbial carbon storage. However, microbial nitrogen remains relatively limited in summer, leading to a higher Cmic:Nmic ratio (Just et al., 2023). The low Cmic:Nmic ratio in spring at Kocapınar suggests that increased nitrogen availability in spring promotes microbial growth, allowing microbes to maintain a more balanced carbon-to-nitrogen ratio (Liu et al., 2024; Zhong et al., 2023).

Seasonal Variation of Nmic, Nmic:NT, NH_4^+, and NO_3^- Contents

The highest Nmic content in spring at Kocapınar reflects increased microbial nitrogen incorporation, likely due to higher organic matter decomposition and reduced plant nitrogen uptake at the beginning of the growing season (Chen et al., 2024; Evangelou and Giourga, 2024). In contrast, low Nmic values in summer at Yalıntepe may result from high plant nitrogen demand and microbial competition for nitrogen, leading to lower microbial nitrogen storage.

The Nmic:NT ratio was highest in spring at Kocapınar, indicating active nitrogen mineralization as microbial populations incorporate nitrogen into their biomass (Schimel and Bennett, 2004). In summer, plant nitrogen uptake intensifies, leading to reduced microbial nitrogen assimilation and a lower Nmic:NT ratio (Nieder and Benbi, 2008).

The highest NH_4^+ content in spring at Katran suggests that microbial ammonification was enhanced by rising temperatures and moisture, promoting organic nitrogen decomposition (Schimel and Bennett, 2004). In contrast, low NH_4^+ levels in summer at Kocapınar result from increased microbial nitrification and plant uptake, which deplete available ammonium (von Wirén et al., 2001).

Seasonal Variation of %C, %N, and C:N Ratios

Soil total nitrogen (%N) and C:N ratios varied significantly across seasons, while total carbon (%C) remained relatively stable. The highest %N content in summer at Yalıntepe suggests that plant activity and

microbial nitrogen cycling were at their peak. The lowest %N content in spring at Katran may result from slower microbial nitrogen turnover under lower temperatures (Cameron et al., 2013; Powlson, 1993).

The C:N ratio was highest in spring at Katran, indicating lower microbial decomposition rates and higher carbon retention (Beese, 1986). In contrast, the lowest C:N ratio in autumn reflects accelerated organic matter breakdown, facilitated by higher microbial activity and nitrogen mineralization (Siebielec et al., 2020).

Seasonal Variation of Microbial Respiration (CO_2-C) and Metabolic Quotient (qCO_2)

Microbial respiration (CO_2-C) varied significantly with seasons, with the highest values in spring at Yalıntepe due to increased microbial activity under optimal moisture and temperature conditions (Yu et al., 2015). The lowest respiration rate in autumn at Kocapınar may result from reduced microbial substrate availability and colder temperatures (Yan et al., 2020).

The metabolic quotient (qCO_2) was highest in spring at Yalıntepe, suggesting a high microbial energy demand, possibly due to stressor rapid microbial growth (Winding et al., 2005). The lowest qCO_2 in autumn at Kocapınar suggests higher microbial efficiency, as microbial communities become more adapted to stable environmental conditions (Anderson and Domsch, 1990).

Seasonal Changes in Absolute and Specific Enzymatic Activities

Enzyme activities normalized by SOC exhibited spatial and temporal variability, with catalase (CAT/C) and urease (Urease/C) activities peaking in summer at Kocapınar and Yalıntepe, respectively. Dehydrogenase activity per unit SOC (DHG/C) was most pronounced in winter at Yalıntepe. These patterns appear to reflect differences in land management, microbial activity levels, and seasonal climatic influences (de Medeiros et al., 2015; Raiesi and Beheshti, 2014).

When enzymatic activity was assessed relative to microbial biomass carbon (EA/Cmic), distinct seasonal trends emerged. Elevated CAT/Cmic in summer at Kocapınar and peak urease activity per Cmic unit in spring at Yalıntepe suggest that microbial enzymatic performance is closely aligned with environmental conditions and microbial physiological states (Bastida et al., 2008). Compared to absolute enzyme activity, EA/Cmic provides a more nuanced view of microbial functionality, highlighting the dynamic response of microbial communities to changes in soil conditions, biomass turnover, and nutrient availability (Nannipieri et al., 2002; Waldrop et al., 2000).

CONCLUSIONS

This study highlights the critical role of absolute and specific enzyme activities—including catalase, dehydrogenase, and urease—as key indicators of soil quality, microbial activity, and nutrient cycling. These enzymes are closely linked to SOC and microbial biomass carbon (Cmic), both of which regulate organic matter decomposition, microbial respiration, and soil fertility. The findings demonstrate that seasonal and locational variations significantly influence enzymatic activity, with temperature, moisture availability, and land use playing pivotal roles in microbial metabolic processes.

The study also underscores the importance of specific enzyme activities per unit SOC and Cmic as valuable tools for assessing microbial efficiency and ecosystem functionality under varying environmental conditions. The observed variations in enzyme activity suggest that these biochemical markers can serve as early warning indicators of soil degradation or improvement, helping guide sustainable land management and agricultural practices.

Despite the strong correlations identified between enzyme activities and soil biochemical properties, further research is needed to fully understand the mechanisms governing enzymatic responses to alkaline soil conditions. Future studies should focus on long-term monitoring, the influence of land use changes, and the interactions between microbial communities and enzymatic functions. Understanding these relationships will be crucial for enhancing soil fertility, improving carbon sequestration, and promoting resilient agricultural systems in semi-arid and alkaline soil environments.

In conclusion, this research reinforces the fundamental role of soil enzyme activities in maintaining ecosystem stability, underscoring their potential as reliable, sensitive, and practical indicators of soil health and environmental sustainability.

CONFLICT OF INTEREST

The authors declare that there is no conflict of interest.

DECLARATION OF AUTHOR CONTRIBUTION

The authors contributed equally to each stage of the study.

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