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Effects of long-term conventional and conservational tillage systems on biochemical soil health indicators in the Mediterranean region

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ABSTRACT

Improved soil health is essential to sustain agricultural production. Therefore, understanding the effects of management on soil health is crucial to implement new agricultural practices. This study aimed to assess the effects of long-term tillage systems on biochemical indicators of a Typic Haploxerert soil under winter wheat-soybean-corn rotation in the Mediterranean region of Turkey. The experiment consisted of two conventional (CT), three reduced (RT), no-tillage (NT), and a strategic tillage practice. The biochemical indicators were total nitrogen (TN), total carbon (TC), soil organic carbon (SOC), microbial biomass carbon (MBC), potential mineralizable nitrogen (PMN), microbial quotient (qM), beta-glucosidase enzyme activity (BGA), and carbon sequestration (Cs) potential. The SOC significantly decreased with the increased tillage intensity, while the tillage had a little effect on PMN, with its highest concentration (78.2 mg kg⁻¹) occurring in the NT. The qM was the only indicator found to be higher under CT than RT and similar to the NT. The BGA peaked in NT which was 460.2 and 536.3% higher than that of the CT. The results showed that SOC, MBC, PMN, BGA and Cs were enhanced with the NT and RT systems which favor sustainability of agricultural production.

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Microbial biomass carbon; beta glucosidase enzyme activity; total nitrogen; C sequestration; tillage

Introduction

Soil health, known also as soil quality, is the potential of soils to sustain productivity, plant and animal health, and environmental quality, and is significantly affected by the management decisions (Karlen et al. 2019). The sustainability of agricultural practices can be assessed by measuring the changes in soil health indicators, which provides information about how the soil is functioning with respect to a particular management practices (Silveira and Kohmann 2020). Biogeochemical properties of soils are significantly affected by the management practices, in particular, tillage, altering quantity, and quality of soil microorganisms. For example, changes in bulk density, water-holding capacity, pore size distribution, aggregation, soil organic matter (SOM), and nutrient accumulation due to the tillage led to the domination of different microorganisms in soils (Schjønning et al. 2011;

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Mathew et al. 2012). Therefore, the biochemical properties have been used as the indicators of soil health in assessing the impacts of agricultural practices (Çelik et al. 2019; Nouri et al. 2019) as they respond faster to the changes in soil environment and management (van Es and Karlen 2019). The biochemical components of soils are responsible for the vital ecosystem functions such as decomposition and mineralization of SOM, biogeochemical cycles, synthesis of humic substances, breakdown of xenobiotics, and nitrogen (N) fixation (García-Orenes et al. 2010). For example, beta glucosidase enzyme plays a key role in the C cycle involving degradation of cellulose, the most abundant compound of plant organic matter synthesized by photosynthesis (Lammirato et al. 2010). Thus, beta-glucosidase enzyme activity (BGA) is considered an important indicator for the changes in the SOM status and degradation (Günel et al. 2018). Potential mineralizable N (PMN) is another indicator of soil health providing information on plant-available N in terms of agricultural productivity and environmental quality (Moebius-Clune et al. 2016).

Tillage is the main driver of changes in soil microclimate and soil microbial communities. The extent of soil inversion, and the amount of residues left on soil surface after tillage are the main criteria used to describe a tillage system. Moldboard plow in the conventional tillage (CT) completely turns over the first 20–30 cm of soil surface and degrades soil health with the adverse impacts on soil microorganisms (Nunes et al. 2020). Conservation tillage practices such as no-tillage (NT), and reduced tillage (RT) have been adopted as the best management practices to improve soil health since they reduce soil erosion, prevent the loss of nutrients and SOM (Zuber and Villamil 2016), sustain productivity and mitigate climate change (Lal 2018). Long-term conservational tillage practices result in higher SOM content (Busari et al. 2015), stable soil aggregates (Çelik et al. 2019), and better habitat for soil organisms (Henneron et al. 2015). However, some studies about long-term conservation practices showed that cropping under reduced soil disturbance may adversely influence the physical and chemical properties as well as biological activity of soils (Hungria et al. 2009; Çelik et al. 2019).

Negative environmental impacts associated with the CT systems have led to the increased adoption of conservation practices, which improved soil properties, provided savings of time, energy, and water and protected soils against water and wind erosion (Muñoz et al. 2007; Liu and Wiatrak 2011). Despite the demonstrated advantages of conservation tillage systems in most parts of the world, moldboard plow, the most common equipment of the CT systems, is still commonly used for the seedbed preparation and weed control in the arable lands. Moreover, the practice of open burning of crop residues before tillage is still common although it has been legally prohibited (Korucu et al. 2009).

The assessment of positive and negative impacts of tillage systems on soil properties is essential to devising the best tillage practices to improve soil health and to sustain the ecosystem goods and services (Nunes et al. 2020). In this study, soil health was assessed using the MBC, BGA, total C and N, C/N ratio, PMN, SOC, microbial quotient (qM), and C sequestration (Cs) as a function of the conventional and conversional tillage systems. This study aimed to answer the following questions: i) Does soil-tillage methods influence soil health? (ii) Which of the soil health indicators are more sensitive to soil-tillage methods? (iii) What is the current status of soil biochemical indicators under long-term tillage practices? and iv.) What would be the impacts of strategic tillage on soil biochemical properties compared to the NT?

Material and methods

Experimental site and design

The study was carried out in agricultural experimental station of Cukurova University established in southern Turkey (37°00'54" N, 35°21'27" E at 32 m asl). The prevailing Mediterranean climate of the study area (Csa, according to Koppen-Geiger classification) had a mean annual temperature of 19.2 °C, total annual rainfall of 639 mm about 75% of which falls during winter and spring months, and total annual evapotranspiration of 1557 mm. The soil-tillage experiment was established on a fine, smectitic, active, mesic Typic Haploxerert soil with surface texture of 50% clay, 32% silt, and 18%

sand contents. Mean pH, electrical conductivity, bulk density, total organic carbon and calcium carbonate content of soils formed over old terraces of Seyhan river were 7.82, 0.15 dS m⁻¹, 1.31 Mg m⁻³, 8.76 g kg⁻¹ and 244.0 g kg⁻¹ in a soil depth of 0–30 cm, respectively (Celik et al. 2011).

The experiment initially consisted of the six tillage treatments in triplicate (Table 1). The tillage plots were 12 m in width and 40 m in length (three replicates per treatment) with a 4 m buffer zone between the plots where the treatments were distributed following a completely randomized block design (Table 1). After 9 years of cropping under NT, half of the NT plots (240 m²) were ploughed with a moldboard plow only once in November 2015 to create a strategic tillage (ST) system. The two CT treatments were the most representative practices of the study area.

Winter wheat and second crops in the NT, RT, and CT systems received the same amount of fertilizers, based on the soil analysis and the nutrient recommendations for the cultivars. Fertilizers were applied in the seedbeds at rates of 172 kg N ha⁻¹ and 55 kg P₂O₅ ha⁻¹ for wheat, 250 kg N ha⁻¹ and 60 kg P₂O₅ ha⁻¹ for corn and 120 kg N ha⁻¹ and 40 kg P₂O₅ ha⁻¹ for soybean. Wheat was sown in the first week of November and harvested in the first week of June. Weed management in the NT and ST systems were carried out using a non-selective herbicide (500 g ha⁻¹ glyphosate) two weeks prior to sowing. Corn and soybean were sprinkler-irrigated once in every 13-day (6 times) during the growing period, and no irrigation water was applied to wheat (Çelik et al. 2020).

Soil analysis

Disturbed soil samples were collected from the top layers (0–10 cm) of each treatment plot between cropped lines after the harvest of wheat in June 2017. BGA was determined following the colorimetric estimation of the p-nitrophenol (Tabatabai 1994). The content of soil MBC (mg C kg⁻¹ soil) was

Table 1. Summary of tillage methods and equipment used in the long-term experiment (Çelik et al. 2019).

Tillage method	Soil tillage for winter wheat	Soil tillage for second crop maize and soybean
Conventional tillage with stubbles (CT-1)	<ul style="list-style-type: none"> ● Second crop residues chopped ● Moldboard plow (30–33 cm)^a ● Disc harrow (13–15 cm, 2 passes) ● Floating (two passes) ● Drill (4 cm) 	<ul style="list-style-type: none"> ● Wheat stubbles chopped ● Heavy tandem disc harrow (18–20 cm) ● Disc harrow (13–15 cm, 2 passes) ● Floating (two passes) ● Planter (8 cm)
Conventional tillage with stubbles burned (CT-2)	<ul style="list-style-type: none"> ● Second crop residues burned ● Moldboard plow (30–33 cm) ● Disc harrow (13–15 cm, 2 passes) ● Floating (2 passes) ● Drill (4 cm) 	<ul style="list-style-type: none"> ● Wheat stubbles burned ● Chisel plow (35–38 cm) ● Disc harrow (13–15 cm, 2 passes) ● Floating (two passes) ● Planter (8 cm)
Heavy disc harrow reduced tillage (RT-1)	<ul style="list-style-type: none"> ● Second crop residues chopped ● Heavy tandem disc harrow (18–20 cm, 2 passes) ● Floating (two passes) ● Drill (4 cm) 	<ul style="list-style-type: none"> ● Wheat stubbles chopped ● Rotary tiller (13–15 cm) ● Floating (two passes) ● Planter (8 cm)
Rototiller reduced tillage (RT-2)	<ul style="list-style-type: none"> ● Second crop residues chopped ● Rotary tiller (13–15 cm) ● Floating (two passes) ● Drill (4 cm) 	<ul style="list-style-type: none"> ● Wheat stubbles chopped ● Rotary tiller (13–15 cm) ● Floating (two passes) ● Planter (8 cm)
Heavy disc harrow zero soil tillage (RT-3)	<ul style="list-style-type: none"> ● Second crop residues chopped ● Heavy tandem disc harrow (18–20 cm) ● Floating (two passes) ● Drill (4 cm) 	<ul style="list-style-type: none"> ● Wheat stubbles chopped ● Herbicide treatment ● No-till planter (8 cm)
No-till or zero tillage (NT)	<ul style="list-style-type: none"> ● Second crop residues chopped ● Herbicide treatment ● No-till drill (4 cm) 	<ul style="list-style-type: none"> ● Wheat stubbles chopped ● Herbicide treatment ● No-till planter (8 cm)
Strategic tillage (ST)*	<ul style="list-style-type: none"> ● Second crop residues chopped ● Herbicide treatment ● No-till drill (4 cm) 	<ul style="list-style-type: none"> ● Wheat stubbles chopped ● Herbicide treatment ● No-till planter (8 cm)

^aFigures in parenthesis are average working depth of the equipment; *This treatment was continued as NT from 2006 until November 2015, tilled with moldboard plow only once in November 2015 and has remained as NT.

determined using the substrate-induced respiration (SIR) method (Anderson and Domsch 1978). The SOC content was calculated by subtracting the soil inorganic C measured by a volumetric calcimeter from the total C determined by the dry combustion. Total soil C (TC) and N (TN) contents were measured using a Costech Elemental Combustion System 4010 (Costech Analytical Technologies Inc. Valencia, CA, USA). The soil inorganic C content was determined using a Scheibler calcimeter, where carbonates were decomposed in a closed line by excess hydrochloric acid, and the degassing CO₂ was determined volumetrically (Allison and Moodie 1965).

The qM describes the SOC availability and was calculated as the ratio of MBC to SOC (Anderson and Domsch 1990). Nitrogen (N), tied up in the complex organic residues which is converted into plant available N forms in incubation under the controlled conditions of temperature, humidity, ventilation, and time, is defined as potentially mineralizable N (Mahal et al. 2019). Potential soil N mineralization was measured using laboratory incubations (Drinkwater et al. 1997). The soil Cs potential under the tillage systems was calculated on an equal mass basis for a soil depth of 0–10 cm using the SOC content and soil bulk density values (Mishra et al. 2010).

$$C_s = (C_m \times D_b) \times D \times A \quad (1)$$

where C_s is the SOC stock (kg m⁻²), C_m is the C concentration (kg kg⁻¹), D_b is the soil bulk density (g cm⁻³), D is the thickness of soil horizon, and A is the area (ha:10⁴ m²). Bulk density was determined using a soil core (its length and diameter: 5 cm) collected from three depths. The soil samples of known volumes were weighed, oven-dried at 105°C for 24 h to a constant weight and weighed to calculate bulk density (Blake and Hartge 1986). Penetration resistance (PR) was determined by a hand-pushed electronic Penetrologger (Eijkelkamp 06.15.SA) as described by American Society of Agricultural Engineers (ASAE 1994). The readings were recorded at 10 mm intervals. The undisturbed soil samples were capillary saturated and equilibrated to -1/3 bar matric potential to determine the moisture content at field capacity (FC) (Klute 1986). Soil moisture at -15 bar matric potential (permanent wilting point, PWP) was determined in disturbed soil samples (Klute 1986). Plant available water (PAW) content was the difference between volumetric water content at FC and PWP.

Statistical data analysis

Statistically significant differences among the tillage systems in each measured soil biochemical health indicator were determined using a variance analysis (ANOVA). Duncan's multiple range test at 95% probability was used as post-hoc where ANOVA indicated significant differences among the tillage treatments. Correlations between the soil indicators were carried out using Spearman's correlation matrix analysis. All the statistical analyses were performed with SPSS version 21.0 (SPSS Inc. USA).

Results and discussion

Total nitrogen and carbon, soil organic carbon and C/N ratio under different tillage systems

The mean TN, TC, SOC values, and C/N ratio under different tillage systems are presented in Table 2. The tillage systems had a significant effect on TN, TC, and C/N ratio. The TN accumulation was enhanced with the decreased tillage intensity after 11 years, and the highest TN level was obtained with NT (0.15%). In contrast, the lowest TN level (0.11%) occurred with CT2, where stubbles were burned and seedbed was prepared using a moldboard plow. The TN level of NT was 25.0 and 36.4% higher than that of CT1 and CT2, respectively.

Similarly, the TC level increased with the decreased tillage intensity. The highest TC (4.7%) obtained with RT3 followed by RT2 (4.7%), and NT (4.6%) (Table 2). The higher calcium carbonate levels of the surface layers under the RT treatments (Çelik et al. 2019) were associated with the slightly higher TC of the RT compared to NT treatments. The C/N ratio increased with the decreased

Table 2. Effects of tillage treatments on soil biochemical health indicators for a soil depth of 0–10 cm ($n = 3$; $p < 0.05$).

Tillage	TN (%)	TC (%)	SOC (%)	C/N ratio
CT1	0.12 ± 0.00 ^{cd}	4.23 ± 0.07 ^d	1.16 ± 0.04 ^d	9.94 ± 0.39 ^d
CT2	0.11 ± 0.01 ^d	4.33 ± 0.06 ^{cd}	1.21 ± 0.04 ^d	10.70 ± 0.31 ^{cd}
RT1	0.12 ± 0.01 ^{cd}	4.29 ± 0.12 ^d	1.41 ± 0.08 ^c	11.36 ± 0.37 ^{bc}
RT2	0.14 ± 0.00 ^{ab}	4.66 ± 0.06 ^{ab}	1.70 ± 0.05 ^b	12.21 ± 0.41 ^{ab}
RT3	0.14 ± 0.00 ^{ab}	4.71 ± 0.06 ^a	1.72 ± 0.06 ^b	12.01 ± 0.45 ^{ab}
NT	0.15 ± 0.01 ^a	4.57 ± 0.09 ^{abc}	1.94 ± 0.10 ^a	12.72 ± 0.52 ^a
ST	0.13 ± 0.00 ^{bc}	4.43 ± 0.08 ^{bcd}	1.49 ± 0.04 ^c	11.42 ± 0.20 ^{bc}

CT1: conventional tillage with residue incorporated, CT2: conventional tillage with residues burned, RT1: reduced tillage with heavy tandem disc harrow, RT2: reduced tillage with rotary tiller, RT3: reduced tillage with heavy tandem disc harrow followed by no-tillage for the second crop, NT: no-tillage, direct planting, ST: strategic tillage, and the mean values with the different superscript letters in the same column are significantly different according to Duncan test at $p < 0.05$.

tillage intensity. The lowest C/N ratio was obtained with CT1 (9.94) and CT2 (10.70), while the highest C/N ratio was obtained with RT2 (12.21) and NT (12.72).

The differences in the SOC contents among the tillage systems were clearly pronounced after 11 years in that the SOC content was significantly lower under CT1 and CT2 systems. The highest SOC content (1.9%) occurred with NT, followed by RT3 (1.7%) and RT2 (1.7%), while the lowest SOC content was obtained with CT1 (1.2%) and CT2 (1.2%) (Table 2). The C loss in agricultural lands was mainly attributed to the post-harvest removal of crop residues and the increased decomposition and mineralization rates of SOC (Lal 2004). Open burning of the crop residues after the harvest, and the intensive tillage operations before planting in CT2 caused the lowest SOC content. In a meta-analysis, Nunes et al. (2020) reported that SOC contents under chisel plowing and NT were 1.18 and 1.45 times higher than those under the moldboard plowing. Tillage was reported to enrich the microbial activity feeding on SOM by breaking down crop residues into smaller pieces, increasing their surface areas and incorporating into the soil (Lupwayi et al. 2004).

One-time soil disturbance after 9 years (ST) reduced TN, SOC, and C/N relative to NT at a ratio of 13.3, 23.2, and 10.2%, respectively. Our results revealed that the accumulated SOC level under the NT system was sensitive to when NT was not continued, and hence, quickly reduced. In contrast to the decrease in SOC, Hungria et al. (2009) reported no detectable change in SOC content with the tillage of NT plots every 3 years.

Potential mineralizable nitrogen under different tillage systems

PMN is N tied up in the complex organic residues, can be easily transformed into the plant available forms, is the main N source for plants and was reported to have a positive relationship with crop yields (Gardner and Drinkwater 2009; Mahal et al. 2019). It is prone to mineralization and can assist in optimizing the N fertilization management (Franzluebbers 2016). The mean PMN values for the tillage systems are shown in Figure 1(a). A significant increase in PMN occurred with the decreased tillage intensity. PMN was estimated at 56.78 mg kg⁻¹ in CT2 (where the stubbles were burned, while a moldboard plow fully inverted the soil), 73.94 mg kg⁻¹ in RT3 (where soil tillage was significantly reduced) and 78.20 mg kg⁻¹ in NT (Figure 1(a)). The mean PMN value was 31.1 and 37.7% higher under the NT than CT1 and CT2 treatments, respectively. The tillage effects on PMN were reported to vary depending on soil type, climate, crop rotation, and management practices. The PMN level was reported to be about 23% higher with NT than the chisel or moldboard plow for an average soil depth of 15 cm (Mahal et al. 2019). Zuber and Villamil (2016) reported that the disk tillage disturbed soil more than did the conservation tillage since the disks inverted the soil as with the moldboard plow. The tillage depth of disks was set to 18–20 cm in RT1 and RT3. However, PMN was significantly higher in RT3 than RT1 which was similar to CT1 and CT2 (Figure 1(a)). The rotary tiller was used in

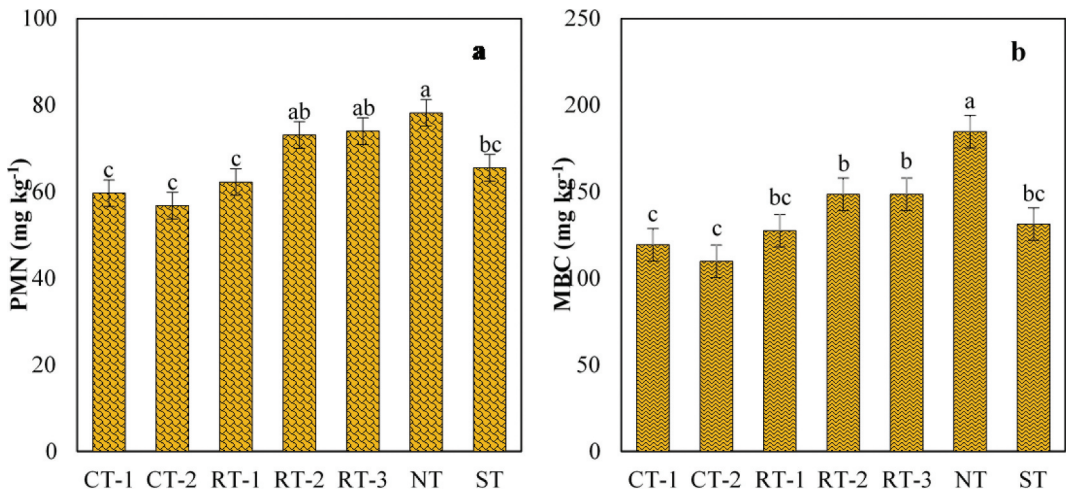


Figure 1. Potential mineralizable nitrogen content (PMN, mg kg⁻¹) (a) and microbial biomass carbon (MBC, mg kg⁻¹) (b) under different soil-tillage treatments. Lower case letters indicate significant differences ($p < 0.05$). Error bars indicate standard error. CT-1: Conventional tillage with stubbles, CT-2: Conventional tillage with stubbles burned, RT-1: Heavy disc harrow reduced tillage, RT-2: Rototiller reduced tillage, RT-3: Heavy disc harrow zero soil tillage, NT: No-till or zero soil tillage, ST: Strategic tillage.

seedbed preparation of the second crop for RT1, while NT planter was used in RT3 for the second crop. The use of rotary tiller in RT1 decreased PMN by 15.8% relative to RT3.

Microbial biomass carbon under different tillage systems

Soil MBC and TN play an important role in the stability of soil aggregates, and the C and N cycles (Zuber and Villamil 2016). The MBC pool, despite its small size, constitutes an important labile fraction of the SOM pool and is indicative of both SOM mineralization and plant nutrients (Zhang et al. 2017). MBC is an indicator of early changes in SOM, and thus, quickly responds to the changing environment and management practices in the agricultural lands. The increased accumulation of plant residues, namely, the increased addition of plant organic C to the soil surface layers with the decreased soil tillage increased MBC under NT and RT when compared to the CT treatments (Figure 1 (b)). The higher MBC storage under NT and RT created a better environmental condition for the microbial growth and activities Zuber and Villamil (2016).

The MBC contents of 119.36 and 109.81 mg kg⁻¹ under CT1 and CT2 rose to 127.50, 148.46, 148.53, 184.65, and 131.16 mg kg⁻¹ with RT1, RT2, RT3, NT, and STI, respectively (Figure 1(b)). Given all the RT systems, the use of heavy disc harrow (for winter wheat) and rotary tiller (for second crop) in RT1 slightly decreased the MBC content compared to rotary tiller (for winter wheat and the second crop) in RT2 and heavy disc harrow (for winter wheat) and NT (second crop) in RT3. Zuber and Villamil (2016) stated that the MBC content was similar under the disking and moldboard plow treatments. Our results for the disking in RT1 supported the findings by Zuber and Villamil (2016), but the MBC level was significantly higher under RT3 than CT1 and CT2 (Figure 1(b)).

The MBC content was 54.7 and 68.2% higher with NT than CT1 and CT2. Similarly, MBC concentrations and enzyme activities were found to be higher with NT owing to the increased amount of crop residues left on the soil surface but to decrease with the burning of residues (Ajwa et al. 1999). The increased MBC content under NT compared to CT was reported as 74% for 14 years of NT (Hungria et al. 2009) and 103% for 16 years of NT (Balota et al. 2003). Nunes et al. (2020) indicated 1.19 and 1.66-fold increases in MBC with chisel plow and NT when compared to moldboard in the topsoil. The lowest MBC level occurred with CT2. No significant difference in MBC was reported by Jokela and Nair (2016).

The increased MBC in NT, RT2, and RT3 was probably related to the changed structure of soil microbial community. In a similar case, Hungria et al. (2009) stated that changes in metabolic activity and soil microbial biomass correlated with changes in soil microbial community structure, and thus, recommended the characterization of soil biodiversity in addition to the measurements of soil microbial biomass.

Carbon sequestration under different tillage systems

The decreased soil disturbance stimulated the accumulation of soil microbial biomass and improved Cs in the fine-textured experimental soil under the conservational tillage practices (Table 3). Wiesmeier et al. (2014) also indicated higher SOC storage of fine-textured soils, particularly in cropland soils. After 11 years, NT on average increased the soil Cs by 75.1 and 92.2% relative to CT1 and CT2, respectively. The lack of crop residues from the preceding period may explain the lower Cs under CT1 and CT2. The adoption of NT added 834.1 kg C ha⁻¹ to the soil each year relative to CT2. Lal (2004) stated that a sequestration rate of 0.4–0.8 Pg C y⁻¹ can be achieved with the adoption of NT. Similarly, Álvaro-Fuentes et al. (2008) indicated a significant reduction in CO₂ released to the atmosphere with the implementation of conservation tillage in the Mediterranean region, where SOM content of agricultural lands is low and heavily depends on incorporation of crop residues.

The residue retention in RT increased the C input into the soil, and thus, increased the soil Cs (Table 3). The positive impacts of RT on the crop residue retention, and Cs were reported in many studies (Murphy 2015; Li et al. 2020). However, Li et al. (2020) reported only 10 and 6% increases under NT and RT relative to CT and suggested that the residue retention in addition to the conservation tillage (NT or RT) further increased the SOC stocks. The increase in Cs under RT was mainly restricted to the surface layer or plough layer, and no significant differences were reported between RT and CT when greater soil depths were investigated (Piccoli et al. 2016). The inconsistencies and questions on the influence of RT on soil Cs are mainly related to the different sampling depths as the quantification of the entire soil profile may not indicate any significant Cs advantage for RT (Nunes et al. 2020).

Microbial quotient under different tillage systems

Microbial quotient is a useful measure of soil microbial activity and was reported to be a more sensitive index than SOC alone (Sparling 1992). Although some studies reported significantly higher

Table 3. Carbon sequestration (kg C ha⁻¹ soil) and microbial quotient (%) as a function of soil-tillage treatments for a soil depth of 0–10 cm ($n = 3$; $p < 0.05$).

Tillage	C sequestration (kg C ha ⁻¹)	Microbial quotient (%)
CT1	10,922.8 ± 380.9 ^e	1.39 ± 0.09 ^a
CT2	9946.1 ± 251.3 ^f	1.48 ± 0.05 ^a
RT1	15,007.7 ± 307.9 ^d	1.13 ± 0.05 ^b
RT2	17,190.3 ± 306.9 ^{bc}	1.17 ± 0.05 ^b
RT3	18,284.2 ± 400.4 ^{ab}	1.09 ± 0.04 ^b
NT	19,121.2 ± 518.3 ^a	1.33 ± 0.05 ^a
ST	16,696.0 ± 405.8 ^c	1.06 ± 0.03 ^b

CT1: conventional tillage with residue incorporated, CT2: conventional tillage with residues burned, RT1: reduced tillage with heavy tandem disc harrow, RT2: reduced tillage with rotary tiller, RT3: reduced tillage with heavy tandem disc harrow followed by no-tillage for the second crop, NT: no-tillage, direct planting, ST: strategic tillage, and the mean values with the different superscript letters in the same column are significantly different according to Duncan test at $p < 0.05$.

qM under the NT than CT systems CT (Balota et al. 2004), this study found no significant difference in qM between NT and CT. The mean qM value was significantly lower under RT and ST than NT and CT (Table 3). These results can be attributed to the differences in the environmental characteristics and management practices such as the duration of tillage practices implemented, soil texture, crop rotation, and the amount of residues returned to soil (Nunes et al. 2020).

The changes in qM indicate the additions of SOM, the changes in microbial C, C losses, and organic C stabilization in soil mineral fractions (Sparling 1992). In contrast to the higher C losses indicated by the lower MBC and SOC contents in CT, the mean qM value was significantly higher under CT than RT (Table 3). The higher qM values pointed to the more active soil microorganisms under NT and CT. Similar qM values under NT and CT in the long-term experiments were attributed to the activation of microorganisms in NT as with tilled plots even with a larger microbial community (Zuber and Villamil 2016).

Beta-glycosidase enzyme activity under different tillage systems

Beta-glycosidase enzyme as the most abundant organic compound in the terrestrial biosphere involves the terminal process of the cellulose (β -1,4-D-glucose polymer) degradation and plays an important role in the C cycle (Eriksson et al. 1990). Greater substrate availability after wheat harvest stimulated the production of BGA. The lowest BGA ($22.25 \text{ mg PNP kg}^{-1} \text{ h}^{-1}$) was obtained in CT2, while the highest activity ($141.57 \text{ mg PNP kg}^{-1} \text{ h}^{-1}$) was recorded under NT (Figure 2). The increased BGA under NT can be attributed to the increased microbial substrate utilization due to the SOC availability, and the slower decomposition rate of fresh organic inputs (Guo et al. 2016). Therefore, the mean BGA value was 560.2 and 636.3% higher under NT than CT1 and CT2, respectively. The decreased BGA in ploughed soils had a negative impact on soil microbial communities by decreasing the surface area for stabilization and the substrate loss due to the increased mineralization of SOM (Lützow et al. 2006). In contrast to the CT systems, the chisel and other equipment used in RT slightly disturbed the surface soil and kept higher amounts of residues on the soil surface. Therefore, the conservational tillage systems have been considered to sustain and improve the soil health.

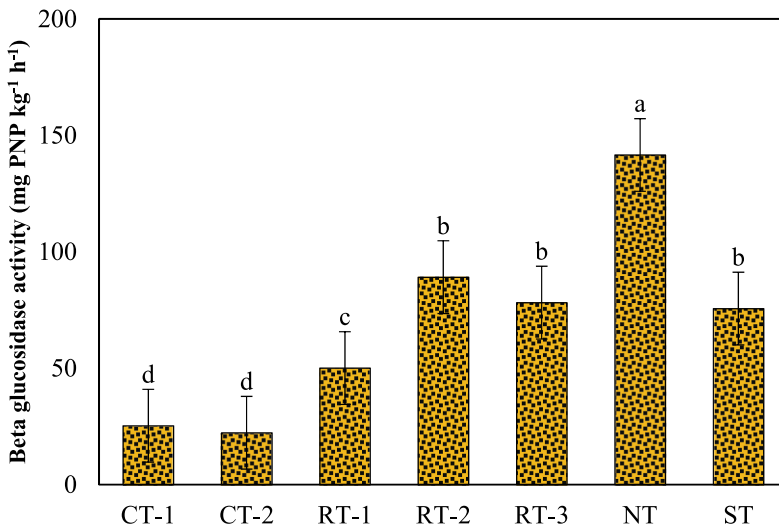


Figure 2. Beta-glucosidase enzyme activity ($\text{mg PNP kg}^{-1} \text{ h}^{-1}$) under different soil-tillage treatments. Lower case letters indicate significant differences ($p < 0.05$). Error bars indicate standard error. CT-1: Conventional tillage with stubbles, CT-2: Conventional tillage with stubbles burned, RT-1: Heavy disc harrow reduced tillage, RT-2: Rototiller reduced tillage, RT-3: Heavy disc harrow zero soil tillage, NT: No-till or zero soil tillage, ST: Strategic tillage.

Mean BGA value under RT1 (where heavy tandem disc was used in winter wheat, with rotary tiller in second crop) was significantly higher than the BGA value in CT1 and CT2 but lower than RT2, RT3, ST1, and NT. Our results were supported by Zuber and Villamil (2016), who indicated that the impact of disking on microbial biomass was not different from that of a moldboard plow. BGA in ST1 indicated a sharp decrease by 46.6% compared to NT in 2 years after the one-time moldboard ploughing of the NT plots. The decreases in TN, TC, PMN, MBC, Cs and BGA under ST1 relative to NT emphasized their importance as indicators to assess the soil health changes by presenting their susceptibility to short-term changes in the tillage practices. Monitoring of the changes via these indicators may guide farmers' decisions to implement new and best management practices in their lands (Nunes et al. 2020).

Correlations between soil properties

All the soil properties were subjected to the correlation analysis to determine the direction and strength of linear relationships under the tillage practices (Tables 4–Tables 6). Significant positive relationships ($p < 0.01$) of aggregate stability with SOC ($r = 0.70$), C/N ratio ($r = 0.53$), PMN ($r = 0.49$) and BGA ($r = 0.80$) indicated that the improved aggregate stability provided a safer habitat for microorganisms and improved the performance of the indicators of soil health (Table 4). The effects of the aggregate formation and stability on the decomposition and mineralization of SOC have been reported by Tiemann and Grandy (2015). Ananyeva et al. (2013) reported that aggregation in fine texture soils caused the formation of larger pores, enhanced the connection between pores, facilitated the movement of water and gases, created a habitat for soil organisms and promoted microbial activity. SOC is used to glue mineral particles in the aggregation process which in turn causes the physical protection of SOM against the microbial attack (Rabot et al. 2018).

Soil water content has a significant impact on the populations and activities of soil microorganisms, and the biochemical processes (Subhani et al. 2001). In this study, significant negative correlations were obtained between plant available water (PAW) content and soil biochemical properties. In contrast to PAW, most biochemical properties had a positive correlation with water content at field capacity and permanent wilting point (PWP) (Table 4).

In contrast to non-significant correlations detected between the biochemical properties and total porosity, Kizilkaya and Dengiz (2010) reported a positive relationship between extracellular enzyme activities and total porosity. The PMN represents the active fraction of soil organic N that is released during the decomposition of SOM (Curtin and Campbell 2008). The PMN had a strong correlation ($p > 0.01$) with aggregate stability ($r = 0.49$), penetration resistance ($r = 0.35$), permanent wilting point ($r = 0.50$) and PAW ($r = -0.42$).

Since the increased bulk density decreases the available water content of soils, its higher values indicates inadequate aeration (Logsdon and Karlen 2004). Bulk density had a significant correlation

Table 4. Spearman's correlation test between soil biochemical and physical indicators.

	AS	BD	PR	FC	PWP	PAW	MiP	MaP	TP
TN	0.55**	0.19	0.40**	0.23	0.57**	-0.47**	0.23	-0.20	-0.04
TC	0.31*	0.35**	0.43**	0.05	0.39**	-0.43**	0.05	-0.32**	-0.37**
SOC	0.70**	0.30*	0.47**	0.42**	0.74**	-0.49**	0.42**	-0.31*	-0.01
C/N	0.53**	0.29*	0.31*	0.47**	0.59**	-0.26*	0.47**	-0.29*	0.06
PMN	0.49**	0.12	0.35**	0.20	0.50**	-0.42**	0.20	-0.16	-0.03
MBC	0.65**	0.22	0.42**	0.50**	0.63**	-0.28*	0.50**	-0.29*	0.09
BGA	0.80**	0.25*	0.53**	0.45**	0.74**	-0.46**	0.45**	-0.22	0.14
Cs	0.78**	0.46**	0.51**	0.44**	0.73**	-0.47**	0.44**	-0.34**	-0.03
qM	-0.18	-0.09	-0.06	0.09	-0.10	0.21	0.09	-0.05	0.01

TN: total nitrogen; TC: total carbon; SOC: soil organic carbon; PMN: potential mineralizable nitrogen; MBC: microbial biomass carbon; BGA: Beta-glucosidase enzyme activity; Cs: carbon sequestration; qM: microbial quotient; AS: aggregate stability; BD: bulk density; PR: penetration resistance; FC: field capacity; PWP: permanent wilting point; PAW: available water content; MiP: microporosity; MaP: macroporosity; TP: total porosity; ** and * refer to significance levels of $p < 0.01$ and $p < 0.05$, respectively.

with SOC ($r = 0.30$), C/N ratio ($r = 0.29$) and BGA ($r = 0.25$). However, biochemical properties such as PMN and MBC did not have a significant correlation with bulk density, while BGA was poorly correlated with bulk density. Soil compaction, indicated by penetration resistance and bulk density, was reported to decrease the microbial biomass and enzymatic activity due to the decreased soil biodiversity (Nawaz et al. 2013). However, positive correlations of penetration resistance were detected with SOC ($r = 0.47$), C/N ratio ($r = 0.31$), PMN ($r = 0.35$), MBC ($r = 0.42$) and BGA ($r = 0.53$) (Table 4). Li et al. (2002) recorded the highest correlations between the soil microbial population and bulk density during the six fully expanded leaf and anthesis growth stages of maize and concluded that the metabolic activity of the microbial population rose with the continuity of exudate releases by the plant roots. In addition, the amount of carbohydrates translocated from leaves to roots was reported to fall during the end of growth stages (Keith et al. 1986). Therefore, the lack of correlation between bulk density and microbial properties, or contradictory relationship between penetration resistance and the biochemical properties can be associated with the time of soil sampling, which was carried out after the harvest of winter wheat in June, when the plant roots were not active and not releasing organic substances.

The significant positive correlations of BGA, an indicator of C mineralization potential, were found with electrical conductivity ($r = 0.42$), SOC ($r = 0.74$); plant available P ($r = 0.69$) and K ($r = 0.55$), while its negative correlation was detected with calcium carbonate content ($r = -0.53$) (Table 5). Similarly, Maurya et al. (2012) indicated that BGA was a useful soil health indicator by virtue of its having a strong positive correlation with many soil fertility parameters such as available N, P, and soil health indicators such as MBC. A significant positive correlation between SOC and BGA was reported earlier and attributed to the active role of beta-glucosidase in the mineralization of carbohydrates (Wick et al. 2002).

Enzyme activities are determined under optimal pH values in laboratory, and the effect of soil pH on enzyme activities is related to the amount of enzyme available in soil (Aon and Colaneri 2001). Contrary to our expectation to have a correlation of soil pH with BGA and the other biochemical

Table 5. Spearman's correlation test between soil biochemical and chemical indicators.

	pH	EC	Calcium carbonate	P	K
TN	-0.07	0.40**	-0.31*	0.58**	0.54**
TC	-0.19	0.21	0.35**	0.57**	0.40**
SOC	-0.21	0.50**	-0.46**	0.74**	0.65**
C/N	-0.29*	0.40**	-0.44**	0.57**	0.47**
PMN	-0.07	0.38**	-0.27*	0.51**	0.49**
MBC	-0.19	0.29*	-0.44**	0.53**	0.37**
BGA	-0.20	0.42**	-0.53**	0.69**	0.55**
Cs	-0.15	0.50**	-0.35**	0.83**	0.76**
qM	-0.10	-0.27*	-0.01	-0.36**	-0.50**

TN: total nitrogen; TC: total carbon; SOC: soil organic carbon; PMN: potential mineralizable nitrogen; MBC: microbial biomass carbon; BGA: Beta-glucosidase enzyme activity; Cs: carbon sequestration; qM: microbial quotient; EC: electrical conductivity; P: available phosphorus; K: available potassium; ** and * refer to significance levels of $p < 0.01$ and $p < 0.05$, respectively.

Table 6. Correlation matrix between soil biochemical indicators.

	TN	TC	SOC	C/N	PMN	MBC	BGA	Cstock
TC	0.61**							
SOC	0.82**	0.67**						
C/N	0.17	0.37**	0.70**					
PMN	0.97**	0.59**	0.77**	0.12				
MBC	0.59**	0.32**	0.66**	0.40**	0.59**			
BGA	0.59**	0.33**	0.74**	0.55**	0.53**	0.74**		
Cs	0.64**	0.50**	0.75**	0.52**	0.56**	0.59**	0.73**	
qM	-0.17	-0.22	-0.21	-0.16	-0.09	0.32**	-0.11	-0.52**

TN: total nitrogen; TC: total carbon; SOC: soil organic carbon; PMN: potential mineralizable nitrogen; MBC: microbial biomass carbon; BGA: Beta-glucosidase enzyme activity; Cs: carbon sequestration; qM: microbial quotient; ** and * refer to significance levels of $p < 0.01$ and $p < 0.05$, respectively.

properties, soil pH, ranging from 7.68 (RT1) to 7.83 (CT1) (data not presented), did not have any significant correlation with the soil biochemical properties, except for the negative correlation with C/N ratio (Table 5). Aon and Colaneri (2001) indicated that as long as pH remained constant, the other soil variables exerted a higher control over the biochemical and microbial activities.

The biochemical properties were significantly correlated with each other except for qM (Table 6). The significant positive correlation between BGA and MBC ($r = 0.74$, $p < 0.01$) found in this study was in close agreement with the findings by León et al. (2017) that higher BGA under NT than CT was associated with a greater microbial population to catalyze carbohydrates in SOM. Since crop residues are the C source for soil microbial community (Madejón et al. 2009), increased crop residues elevated MBC in the soil surface. This stimulating effect of increased plant organic matter was evidenced by the high correlation of SOC with all the soil biochemical properties measured (except for qM) (Table 6). The high correlations of SOC with the biochemical properties were related to microbial population under different conditions (Madejón et al. 2009; Guo et al. 2016; Zuber and Villamil 2016). Beta-glucosidase enzyme related to the C and N cycles in that BGA was positively correlated with the C/N ratio. The BGA was also correlated positively with SOC, TN, and PMN ($r = 0.74$, 0.59 , and 0.53 , respectively, $p < 0.01$) (Table 6). Ajwa et al. (1999) attributed to the correlation between TN and BGA to the long-term N fertilization. The high correlations among SOC, MBC, and BGA indicated that enzymes were not stabilized in SOM. However, Trasar-Cepeda et al. (2008) attributed the lack of correlation between SOC and MBC to the stabilization of enzymes by the soil components. PMN was positively correlated with most of the biochemical properties. Similar to our finding, Dessureault-Rompré et al. (2010) reported a significant positive correlation between PMN and SOC due to the closely coupled C and N cycles (Mallory and Griffin 2007).

Conclusions

Our results verified the importance of conservation tillage systems to the agricultural soil health in the Mediterranean region of Turkey, where CT practices are still being commonly used. The indicators of total nitrogen, soil organic carbon, carbon sequestration, potential mineralizable nitrogen, and beta-glucosidase enzyme activity proved to be sensitive and useful for the assessment of soil health. The results of 11-year-old tillage experiment revealed that the CT systems negatively impacted the soil health indicators of the Typic Haploxerert soil. Among all tillage systems, the NT exerted the most positive effect on soil health of agricultural ecosystems, and their sustainability. The conservation tillage is an effective system to increase soil C sequestration, and thus, to mitigate the elevated atmospheric CO₂ concentration. Also, our results can be extended to the adoption of best management practices to reverse the degradation and desertification of the other hotspot ecosystems in the Mediterranean region.

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