



The impact of tillage systems and crop residues on microbial mass and soil structure stability indices

Afsaneh Alinejadian-Bidabadi¹, Abbas Maleki² and Mahtab Roshaniyan¹

¹ Lorestan University, Faculty of Agriculture, Soil Science Dept., Khoramabad, Iran. ² Lorestan University, Faculty of Agriculture, Water Engineering Dept., Khoramabad, Iran.

Abstract

Aim of study: This research investigated the effects of management practices, including plant residues and tillage practices, on soil stability indices, microbial biomass carbon, and the number of bacteria.

Area of study: Northern Khorasan Province, Iran.

Material and methods: This study explored the effects of the three year-old tillage systems of conventional tillage (CT), minimum tillage (MT), and no-tillage (NT) at three levels 0, 40, and 70% of plant residues on soil physical and microbiological properties for a rotation of three years (wheat, canola, and wheat). Variables measured in this study included the whole soil stability index, the normalized stability index, the percentage of aggregate destruction (PAD), the number of bacteria, and microbial biomass carbon.

Main results: Management practices could affect variables, such as soil structure stability as well as the number of bacteria. The results also showed that soils of higher stability were more resistant to soil degradation. In addition, by reducing tillage and adding plant residues, the PAD index decreased significantly. NT and MT practices improved soil structure stability indices and significantly increased the number of bacteria as well as microbial biomass carbon in contrast to CT, what could be attributed to the increased soil organic matter.

Research highlights: Reduced tillage practices showed the potential for enhancing soil physical quality only through improving aggregate stability. Therefore, NT with 70% residue retention was found to be suitable to improve soil sustainability indices and increased soil microbial population.

Additional key words: improving aggregate stability; microbial biomass carbon; normalized stability index; plant residues; whole soil stability index.

Abbreviations used: CT (conventional tillage); DL (disruption level); MT (minimum tillage); MWD (mean weight diameter); NT (no-tillage); NSI (normalized stability index); PAD (percentage of aggregate destruction); SOC (soil organic carbon); SOM (soil organic matter); WSA (water-stable aggregation); WSSI (whole soil stability index).

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Correspondence should be addressed to Afsaneh Alinejadian Bidabadi: alinezhadian.a@lu.ac.ir; alinejadian@yahoo.com

Introduction

Soil aggregation withstands soil fertility by reducing erosion and improving soil aeration, as well water infiltration and retention. In addition, soil aggregation prevents soil organic matter (SOM) from being mineralized by physically reducing the accessibility of organic compounds for microorganisms, extracellular enzymes, and oxygen (Spohn & Giani, 2010).

The study of aggregates is one way to quantify whether management practices improve natural cha-

racteristics and agricultural capacity of soil. Soil aggregate and structure stability is the result of the interaction among many agents, such as the environment, management practices, crop, inherent soil features, and soil biological and non-biological processes (Bronick & Lal, 2005). The size of soil aggregate and stability are also used to characterize soil structure because those indicators are correlated with various soil functions, including carbon sequestration and gas exchange through physical protection of SOM (Rabot *et al.*, 2018). Other studies show the stability index

is higher under reduced tillage conditions than under conventional tillage (CT) conditions in spring and summer (Eynard *et al.*, 2004). Land use and associated management practices, such as crop sequencing, fertilization, soil conditioning, drainage, and irrigation are the major and most direct ways of affecting soil structure and properties by their impact on destruction forces and aggregate-creating processes (Lehrsch *et al.*, 2012). Wright *et al.* (2007) reported that the use of the minimum tillage (MT) system improves the content of soil organic carbon (SOC) and increases soil microbial population. In other regions of the world, no-tillage (NT) and MT practices have shown to improve soil quality and crop productivity (Norton *et al.*, 2012). In most agricultural systems, crop residues are the main substrate for soil microorganisms (Govaerts *et al.*, 2009), and their mineralization rate depends on the placement and existence of favorable conditions (Helgason *et al.*, 2014).

The sustainability of tillage systems has received considerable attention in recent years. In addition, with an increase in the adoption of conservation-oriented tillage practices, they are considered more sustainable than CT. Soil management involving reduced tillage has often been reported to improve soil structure (Oyedele *et al.*, 1999). MT operations for example, have positive influence on soil physical, chemical and biological properties as macro-pore structure, aggregate stability, nutrients availability, and the diversity of microbial populations while reducing soil disturbance (Heidari *et al.*, 2016). Soil aggregates have been found to be more stable in reduced tillage than in conventional moldboard tillage (Pagliai *et al.*, 2004). Although the effects of tillage operations on soil structure has been well documented, past research mainly focused on aggregate stability (Emami *et al.*, 2012). In addition, soil aggregate distribution and stability measurements have been suggested as soil quality indices. However, aggregate stability is often measured in terms of a specific aggregate size class that is not a measurement of whole soil structure (Six *et al.*, 2000).

Due to the importance of soil in Iran's food production and its key role in natural ecosystems, it is necessary to pay more attention to soil management issues in achieving sustainable development. Therefore, it is necessary to study the effective features of the maintenance and enhancement of soil structure stability indices resulting from various management practices. In arid and semi-arid regions, such as Northern Khorasan Province (Iran), because of limited rainfall and the decomposition of SOM, the careful management of organic matter and soil properties, such as soil biota and physical indices, as well as conservation of water, are very important. In addition, due to the high rate of cultivation in this region, the conservation of the structure and properties of the soil is of high significance. Therefore, to promote the preservation of soil and water resources in agricul-

tural systems in arid and semi-arid regions, conservation tillage operations (NT and MT) are necessary, in that they can contribute to avoiding soil degradation by compaction.

The key hypothesis was that in the three year-old tillage systems for a rotation use of three years (wheat, canola, and wheat), NT and MT practices cause shifts in the soil bacterial community as well as in the microbial biomass carbon to improve soil structure stability indices relative to CT practices. Based on this introduction, in this research the effects of different tillage methods on soil stability indices, microbial biomass carbon, and the number of bacteria were investigated.

Material and methods

Experimental area

The present research was conducted in Northern Khorasan Province (Iran). This province has the geographic coordinates of (37°28'N, 57°19'E), is 1070 m above the sea level, and has a temperate warm semi-arid climate. The slope of the studied site was 0-2%. The region of the study has warm summers and relatively cold winters. Based on a 44-year statistical period, the average annual rainfall at the meteorological station of the region is reported to be 259.7 mm, with the mean annual temperature of 13.7 °C.

Experimental design and treatments

This study used a factorial experiment with a randomized complete block design and three replications. The experimental treatments of this study consisted of different soil tillage methods: (1) CT (moldboard plowing, disc, leveling, furrowing, and seed drilling), (2) MT (chisel plowing, furrowing, and seed drilling), and (3) NT (seed drilling), which were considered together as the first factor. In addition, the management of plant residues, as the second factor, included no residue, 40% of plant residues, and 70% of plant residues. In the direct drilling method (NT), tillage operations were not done before seeding, and cultivation was done with a one-time tillage operation. The area of each plot for the tillage systems (first factor) was 30 m in width × 50 m in length, and the area of each individual plot for the plant residues (second factor) was 10 m in width × 50 m in length. The total area of the experiment (27 plots) was 13500 m². In this study, 9 treatments were used, which included CT + no plant residues (CT0), CT + 40% of plant residues (CT40), CT + 70% of plant residues (CT70), no tillage + 0% of plant residues (NT0), NT + 40% of plant residues (NT40), NT + 70% of crop residues (NT70), MT + 0% of plant residues (MT0),

MT + 40% of plant residues (MT40), and MT + 70% of plant residues (MT70), with each treatment having had three replicates.

The sources of nitrogen, phosphorous and potash were urea, triple superphosphate and sulphate of potash, respectively. Based on the soil testing results, the amounts of the applied fertilizer for all plots were 230-145-40 NPK. In addition, trifluralin as a herbicide for canola and 2, 4-Dichlorophenoxy acetic acid (2, 4-D) as a herbicide for wheat were applied 2 L/ha and 1.5 L/ha, respectively.

This study explored the effects of the three year-old tillage systems of CT, NT, and MT with the plant residues of 0%, 40%, and 70% on soil structure stability indices, the number of bacteria and microbial biomass carbon for a rotation of three years wheat (*Triticum aestivum* L.), canola (*Brassica napus* L.), and wheat.

Soil sample collection and experimental materials

Surface soil layers (0-20 cm) were randomly sampled from 10 points in each treatment plot with an auger and thoroughly mixed into one composite sample. For this purpose, a total of 27 composite samples (9 treatments × 3 replicates = 27 samples) were collected in sterile plastic bags and divided into two parts, for physical and microbial analysis, respectively. To manage plant residues in different tillage treatments, some plots received any residues (0%), whereas, in the other plots, the plant residues (40% or 70%) were incorporated into the first 10 cm of soil uniformly by tilling using a cultivator.

Measurement of soil physical and microbial properties

In the end, after the third year, the soil samples were collected from the soil surface (0-20 cm), and soil structural indices, *i.e.* the whole soil stability index (WSSI) and normalized stability index (NSI), the number of bacteria, as well as microbial biomass carbon were measured. The properties of the initial soil are listed in Table 1. In this study, soil texture was determined based on the hydrometer method (Bouyoucos, 1936, 1962; Beverwijk, 1967). In addition, the soil saturated paste extract (ECe) and the pH of the saturated soil paste were determined based on the study of Smith & Doran (1996), and the amount of SOC, as an index of the SOM, was determined based on the study of Nelson & Sommers (1982).

WSSI, aggregate distribution, and water-stable aggregation (WSA) were measured in dry-sieved aggregates in four aggregate size classes (9.5–2, 2–1, 1–0.25, and 0.25–0.053 mm). The dry sieving practice consisted of placing soil atop a screen with the size equal to that of the largest

Table 1. Physicochemical properties of the initial soil in the experiment

Properties	Value
Sand (%)	48
Silt (%)	41
Clay (%)	11
Textural class	Loam
pH _{paste}	7.3
ECe (dS/m)	0.65
OC (%)	0.67
TNV (%)	25
Total N (%)	0.32
Available P (mg/kg)	7.30
Available K (mg/kg)	250

ECe: electrical conductivity of a saturated soil extract. OC: organic carbon. TNV: total neutralizing value (percentage of equivalent CaCO₃).

aggregates in size, tapping the sides at least 50 times with the palm to make the soil pass through the screen, collecting the soil passing through the screen on a piece of kraft paper, and pouring it onto a screen equal to the smallest aggregates in size, followed by tapping. Each aggregate of the size classes was collected individually from the largest to the smallest ones. The weight of the aggregates in each size class was measured and used to calculate the proportion of dry-sieved the aggregates in each size class (Pai) to the whole soil (Eq. [1]).

The soil on top of the 9.5 mm screen and below the 0.053 mm screen was collected and weighed as part of the summed total weight (WT) (Nichols & Toro, 2011). Pai was achieved as follows:

$$Pa_i = \left[\frac{W_A - [(W_C/W_O) \times W_A]}{W_T} \right] \quad [1]$$

where, W_A is the weight of the total material in each size class i , W_C represents the weight of the coarse material measured during wet sieving for size i , W_O is the weight of the aggregates placed on the sieve before wet sieving for size i .

WSA was measured in four subsamples from each aggregate size class according to the modified Kemper & Rosenau (1986)'s method. In brief, the aggregates (4 g for 9.5–2 and 2–1 mm aggregates, 2 g for 1–0.25 mm aggregates, and 1 g for 0.25–0.053 mm aggregates) were placed onto screens 1/4 of the smallest size and capillary rewetted for 10 min. Stable aggregates were separated by mechanical wet sieving for 5 min using a piece of apparatus introduced by Kemper & Rosenau (1986). The material collected on the sieve was washed gently into weighing boats, dried at 70 °C, and weighed. The coarse material, including sand, roots, and particulate organic matter, was

removed by dispersing the aggregates in 0.5% sodium hexametaphosphate, which was shaken periodically over a 5-min period. Next, pressurized water and rubber policemen were used to push the disrupted aggregates through a screen matching the smallest aggregates in the size class. The material remaining on the screen was collected, dried at 70 °C, weighed, and subtracted from the weight of the aggregates collected after wet sieving. The formula used to calculate WSA for each size class (Nichols & Toro, 2011) is as follows:

$$WSA_i = [(W_A - W_C)/W_O] \times 100 \quad [2]$$

In addition, the dry aggregate size distribution and WSA calculated above were included in the formula for the WSSI (Nichols & Toro, 2011) as follows:

$$WSSI = \left[\sum_{i=1}^n (I) \times (P_{ai} \times ((WSA_i) \div 100)) \right] \div n \quad [3]$$

where, n represents the number of the aggregate size classes, i equals n and decreases by an increase of 1 from the largest to the smallest aggregate size classes, I is each size class (Nichols & Toro, 2011). In addition, the NSI is the measure of soil stability based on comparing the aggregate distribution before and after the disruption proposed by Six *et al.* (2000). Briefly, samples from each treatment were applied into three groups before wet sieving. In one group, air-dried soil samples were rewetted by capillary overnight before wet sieving. In another group, soil samples quickly were wet-sieved without rewetting (slaked), and the final group was provided for a maximum disruption to measure the coarse material (*i.e.* sand and organic matter). Maximum disruption used forced water to destroy aggregates and wash them through stacked sieves. The capillary-rewetted and slaked samples were wet-sieved for 2 min through three sieves (2000, 250, and 53 µm) individually by moving the material that went through the sieve onto the next smaller sieve. The water-stable aggregates on sieves in both the capillary-rewetted and slaked treatments were collected, dried at 70°C, weighed, and corrected for the coarse material (Six *et al.*, 2000).

The formula for the calculation of the disruption level of a size class upon slaking (DLSi) is as follows:

$$DLS_i = \{[(P_{i0} - S_{i0}) - (P_i - S_i)] - [(P_{i0} - S_{i0}) - (P_i - S_i)]\} / 2 \times [1 / (P_{i0} - S_{i0})] \quad [4]$$

where, P_{i0} stands for the proportion of the total sample weight in size class I before disruption (*i.e.* rewetted), P_i represents the proportion of total sample weight in size class I after disruption (*i.e.* slaked), S_{i0} is the proportion of sand with size I in the aggregates of size I (aggregate-sized sand) before disruption, and S_i is the proportion of sand with size I in the aggregates of size I after disruption. All proportions have been expressed on a soil weight basis (g fraction g⁻¹ soil). Size classes used in

this study included I = 1: < 53 µm, I = 2: 53-250 µm, I = 3: 250-2000 µm, and I = 4: > 2000 µm. The factor before the multiplication sign in Eq. [4] calculates the level of disruption caused by slaking and corrects the proportions of aggregate size classes for the aggregate-sized sand content. Besides, this factor ensures that only weight losses are used in calculating the index; in other words, this factor will be 0 if there is a weight gain. The factor after the multiplication sign normalizes the weight loss to the maximum value for that size class. The aggregate-sized sand content could be used for sand correction. The whole soil disruption level (DL) was calculated as follows:

$$DL = 1/n \sum_i^n [(n + 1) - I] \times DLS_i \quad [5]$$

DL is the weighted sum of disruption for each size class I because a weight loss in a smaller size class is more indicative of soil instability than a weight loss in a larger size class.

Weighting factors for disruption in different aggregate size classes were assigned arbitrarily by ranking the aggregate size classes from 1 to 4. The weighting factors are not based on arithmetic or geometric means because the use of mean indices will be questionable if the aggregate distribution is skewed (Stirk, 1958), which is often the case. The maximum disruption level for each size class I [DLSi (max)] was calculated by the following formula:

$$DLS_i (max) = [(P_{i0} - P_p) + |(P_{i0} - P_p)|] / 2 \times [1 / (P_{i0} - S_{i0})] \quad [6]$$

where, P_p is the primary sand particle content with the same size as the aggregates of the size class after the complete disruption of the whole soil. The whole soil DL (max) is calculated using Eq. [5] with DLSi replaced by DLSi (max). The NSI is then computed as follows:

$$NSI = [DL / DL max] \quad [7]$$

The DL was divided by the DL (max) to normalize the DL for the maximum possible disruption based on the primary sand particle-size distribution.

The percentage of aggregate destruction (PAD) is another suitable index used to appraise soil physical structure, which is based on dry-sieved aggregates' mean weight diameter (dry MWD) and wet-sieved aggregates' mean weight diameter (wet MWD), which was calculated (Cates *et al.*, 2016) as follows:

$$PAD = [(M_d - M_w) / M_d] \times 100 \quad [8]$$

where, M_d and M_w represent the weight of dry MWD >0.25 mm and the weight of wet MWD >0.25 mm, respectively.

To determine the soil microbiological activity, 500 g of the rhizosphere soil sample was collected from the plot

in each replication in every sampling. After removing the roots, the samples were passed through a 5-mm sieve. In addition, after removing the residues, the samples were passed through another 2-mm sieve. The number of microbial populations was determined using a fluorescence microscope (Li *et al.*, 2002). For this purpose, 10 μL of the final solution was placed on a slide and spread uniformly with a needle. The slide had a concentric circle with the specific surface area of 1 cm^2 . For the purpose of bacterial staining, 5 mL of an orange acridine solution was added to the slide. After drying the slides, the number of bacteria, lightened by staining, was counted using a 500x magnification fluorescence microscope. In the end, the number of bacteria in each gram of the dry soil was calculated according to the following equation:

$$\text{Number of bacteria} = [(BF \times 10^4 \times DF/AF)] \quad [9]$$

where BF represents the number of bacteria counted on the slide, DF is the dilution factor (150), and AF shows the area of a microscopic field on the slide.

To measure microbial biomass carbon, the fumigation-extraction method (Sparling & West, 1988) was used. In this method, the soil samples were sterilized by chloroform and extracted using a potassium sulfate solution. In the end, the organic carbon content of the extract was measured, with the microbial biomass carbon obtained as follows:

$$\text{Microbial biomass carbon} = [(S - C)/0.35] \quad [10]$$

where S represents organic carbon (mg) per 100 grams of the dry soil, and C is organic carbon (mg) per 100 grams of the non-fumigated dry soil (control).

Statistical analysis

The analysis of variance for different tillage methods and plant residues on soil stability indices, microbial biomass carbon, and the number of bacteria was performed following the ANOVA technique with the SPSS V.19.0 (SPSS Inc., Chicago, IL, USA, 2010). Comparisons between statistical averages of the various treatments were made with the Tukey test ($\alpha \leq 0.05$). Computation and preparation of figures were done using Excel 2016.

Results

The proportion of dry-sieved aggregates for each size class

In this study, the proportion of aggregate weight values was significantly higher for 9.5–2, 2–1, and 1–0.25 mm

size classes, *i.e.* macroaggregates, in the treatments of MT and NT systems (Fig. 1). As a result of the less frequent tillage practices, the highest Pai, for 9.5–2 mm aggregates, was assigned to NT and MT (Fig. 1).

Soil structure stability indices

There was a significant difference between the effects of different tillage methods on the WSSI, with NT and CT having had the highest and the lowest WSSI values (Table 2). The lowest and highest WSSI values were found for 0% of plant residues (0.25) and 70% of plant residues (0.29), respectively (Table 2). The interaction effects of tillage and plant residues are shown in Fig. 2a. Among the studied treatments, the highest and lowest WSSI values were found for NT with 70% of residues (0.35) and CT with 0% of (0.17) residues, respectively. The average WSSI value in NT with 70% of plant residues was 98% higher than that in CT with no plant residues.

The results showed that NT led to a significant increase in the NSI (Table 2). Accordingly, the amount of NSI in different tillage systems was as such NT>MT>CT (0.81>0.73>0.42). In addition, there was a significant difference in the effects of the amount of plant residues on the NSI. In view of this, the highest and lowest NSI values were obtained for 70% of plant residues and no-plant residues, respectively. The interaction effects of the tillage treatments and plant residues showed that tillage systems (NT, MT, and CT) significantly increased the NSI value by adding plant residues, in contrast to no plant residues (Fig. 2b).

In this study, the PAD index increased significantly with an increase in tillage intensity (Table 2). According to the results, the lowest and highest PAD values were

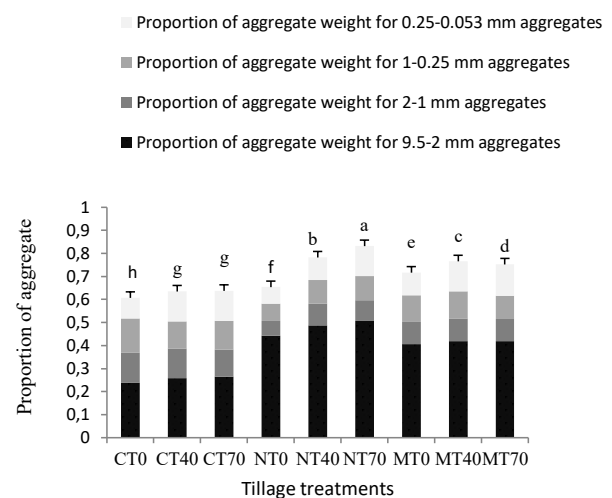


Figure 1. The proportion of aggregate weight for each size class in different treatments. CT, conventional tillage. MT, minimum tillage. NT, no tillage.

Table 2. Effects of tillage treatments and plant residues on soil stability indices ($p < 0.05$)

Treatments	WSSI	NSI	PAD
NT	0.3272 ^a	0.8116 ^a	59.07 ^c
MT	0.2841 ^b	0.7335 ^b	69.42 ^b
CT	0.1986 ^c	0.4257 ^c	86.06 ^a
0% plant residue	0.2526 ^b	0.6329 ^c	74.06 ^a
40% plant residue	0.2657 ^b	0.6583 ^b	71.30 ^b
70% plant residue	0.2915 ^a	0.6796 ^a	69.18 ^c

NT: no tillage. MT: minimum tillage. CT: conventional tillage. WSSI: whole soil stability index. NSI: normalized stability index. PAD: percentage of aggregate destruction. In each column, figures followed by different lower case letters are significantly different.

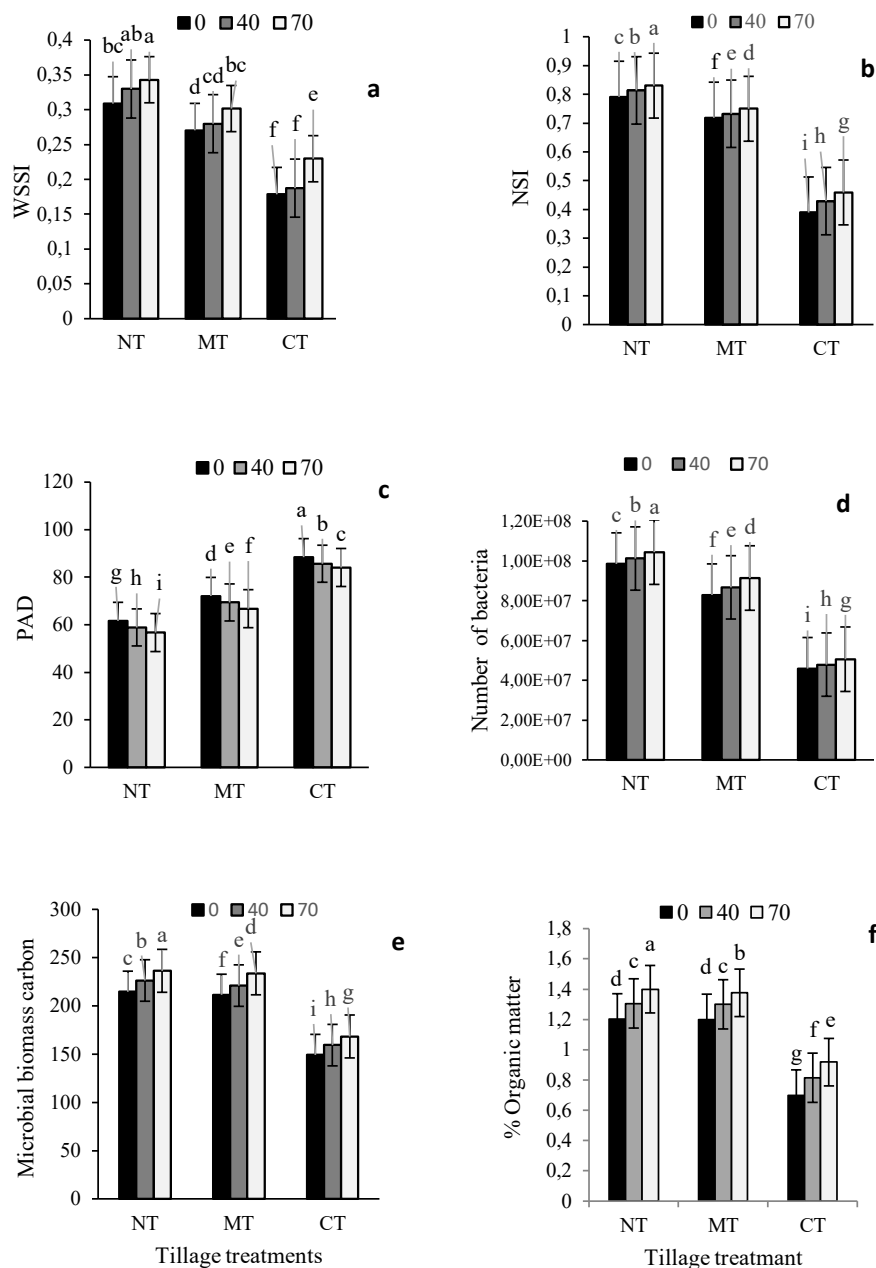


Figure 2. The interaction effects of tillage treatments and plant residues on (a) WSSI (whole soil stability index), (b) NSI (normalized stability index), (c) PAD (percentage of aggregate destruction), (d) number of bacteria, (e) microbial biomass carbon and (f) organic matter. CT, conventional tillage. MT, minimum tillage. NT, no tillage.

obtained with 70% and 0% of crop residues, respectively (Table 2). The highest and lowest PAD values were obtained for CT with 0% of residues (88.43) and NT with 70% of plant residues (56.71), respectively (Fig. 2c).

Number of bacteria and microbial biomass carbon

The analysis results of the variance of the effects of tillage treatments and plant residues on the number of bacteria and microbial biomass carbon are shown in Table 3.

In tillage treatments, the number of bacteria increased significantly with a decrease in tillage intensity (Table 4). In general, an increase in the amount of plant residues in soil as well as a decrease in tillage intensity were the major factors stimulating the activity of useful microorganisms in soil.

The highest and lowest number of bacteria per gram of dry soil was found for NT with 70% of residues (104.4×10^6) and CT with 0% of residues (45.8×10^6), respectively, which highlights the effects of tillage management practices, *i.e.* tillage and crop residues, on soil number of bacteria. The comparison of the means showed that CT system significantly reduced the bacterial population, so that in treatment CT with no plant residues a reduction of 1.27 times that of the NT treatment with 70% of plant residues was obtained (Fig. 2d).

In this study, NT had the highest and CT had the lowest amount of microbial biomass carbon (Table 4). In addition, the highest amount of microbial biomass carbon was found for 70% plant residues, and the lowest amount of microbial biomass carbon was obtained for no plant residues (Table 4).

In the tillage treatments, the microbial biomass carbon increased significantly after using plant residues (Fig. 2e). Among the studied treatments, the highest and lowest values of microbial biomass carbon were obtained for NT with 70% of residues (236) and CT with 0% of residues

(149), respectively. Regarding the effects of tillage management practices, *i.e.* tillage and crop residues, on soil microbial biomass carbon, it was observed that the microbial biomass carbon in NT with 70% of plant residues was 58% higher than that of CT with no plant residues.

The organic matter increased by reducing tillage intensity. In addition, among the studied treatments, the highest and lowest values for the organic matter were obtained for NT with 70% of residues (1.4%) and CT with 0% of residues (0.7%), respectively (Fig. 2f).

Discussion

Effects of tillage methods and plant residues on Pai

The Pai values of the aggregates larger than 2 mm were lower in the CT treatments than in NT and MT. It seems that the breakdown of the macroaggregates due to compression by tractor wheels increased the microaggregates. Intensive traffic by the machinery in inadequate tillage practices can lead to degradation of soil structure, due to the gradual loss of stable aggregates and organic matter in soil. The fewer number of tractor wheels in the NT systems has been effective in the formation of the macroaggregates (Fig. 1).

According to Tisdall & Oades (1982), the formation and stability of micro-aggregates and macro-aggregates are interrelated processes. They believed that compared to micro-aggregates, macro-aggregates are less stable and are more susceptible to disruption by soil management practices, cultivation, and environmental disturbance (*e.g.* wet-dry cycles).

Kalhor *et al.* (2017) found that macro-aggregates are composed of organic binding agents. Accordingly, the protective mechanism effects of macro-aggregates on SOC are stronger than those of micro-aggregates (Hurisso *et al.*, 2013). Naidu *et al.* (1996) reported that tillage

Table 3. The analysis of variance for different tillage methods and plant residues on microbial biomass carbon, and the number of bacteria

Source	Mean square	
	No. of bacteria	Microbial biomass C
REP	0.89 ^{ns}	1.1825 ^{ns}
Tillage practices	6833.78**	12701.28**
Plant residues	91.61**	994.36**
Tillage×plant residues	2.74 ^{ns}	3.7632*
Error	0.98	0.82

** , * : Significant differences at 1% and 5% levels, respectively. ns: no significant difference

Table 4. Effects of tillage treatments and plant residues on number of bacteria and microbial biomass carbon ($p < 0.05$).

Treatments	No. of bacteria	Microbial biomass C
NT	101.38 ^a	225.74 ^a
MT	87.00 ^b	222.05 ^b
CT	48.12 ^c	158.91 ^c
0% plant residue	75.77 ^c	191.74 ^c
40% plant residue	78.58 ^b	202.20 ^b
70% plant residue	82.14 ^a	212.76 ^a

NT: no tillage. MT: minimum tillage. CT: conventional tillage. In each column, figures followed by different lower case letters are significantly different.

operations increased bulk density of soil and decreased aggregate stability.

Retention of crop residues on the soil surface under NT, due to increased microbial activity and production of binding agents, significantly improved macroaggregate formation compared with CT. These findings are consistent with those reported by Lichter *et al.*, (2008) and Golchin *et al.* (1994). Nichols & Toro (2011) reported the presence of coarse aggregates in zero and NT systems, native rangelands, as well as medium grazing rangelands. SOM participates in various formation and stability processes of soil aggregates (Six *et al.*, 2004). Soil aggregate stability is a definitive variable in understanding soil erosion processes in agricultural and forest soils (Garcia-Orenes *et al.*, 2012; Haregeweyn *et al.*, 2013). CT practices may expose the soil to erosion and reducing tillage intensity can help mitigate soil erosion and generally improves biological and physical soil health. Research on soil aggregate stability is an essential requirement for the importance of on-site and off-site water erosion damage (Montanarella *et al.*, 2016).

Effect of tillages systems and plant residues on soil stability indices

According to Zhang *et al.* (2015), a decrease in tillage disturbances increases the stability of soil aggregates. The reason is that the frequent tillage of farmlands destroys the soil particle structure, increases soil aeration, and deteriorates protection of soil particles, thereby resulting in loose soil structure and an increase in the damage to the soil structure (Su *et al.*, 2017). Zhou *et al.* (2020) found out that a decrease in soil aeration with NT in the farmland rapidly increased SOC storage with the rotation of legumes and the gramineae, and improved stabilization of soil aggregates.

CT reduced aggregate stability. However, the aim of tillage operations in crop production is to prepare soil for sowing and plant growth, but an intensive soil tillage is

the most destructive force of soil resources (Madari *et al.*, 2005). Not using tillage on a long-term basis without the movement of the soil could lead to more serious compaction problems than the passage of heavy machinery used in traditional tillage. The continuous non-use of tillage could increase bulk density (*i.e.* $\geq 1.30 \text{ g cm}^{-3}$), followed by crop yield loss, which occurs due to insufficient soil aeration (Reynolds *et al.*, 2009). According to Blanco-Canqui & Ruis (2018), shortly after conversion from CT to NT up to 4–5 years, deterioration in soil properties, *i.e.* a decrease in hydraulic conductivity, a decrease in soil porosity, and a corresponding increase in dry bulk density occurred regardless of the soil type.

Low aggregate stability in CT system could be due to the high soil displacement rate and pressures on aggregates during tillage operations. Six *et al.* (2000) investigated the effects of soil structure as well as SOM and minerals on the NSI, and they reported that the NSI decreased with an increase in tillage intensity. Accordingly, the NT was arranged in the form of native plants > NT > CT. In addition, there was a significant difference in the effects of the amount of plant residues on the NSI. In view of this, the highest and lowest NSI values were obtained for 70% of plant residues and no-plant residues, respectively. Thus, adding plant residues leads to the increased microbial activity, which ultimately leads to an increase in soil structure stability. One of the reasons for an increase in aggregate stability is the increase in the microbial population and accordingly in microbial activities, with metabolites produced by them. These metabolites include polysaccharides, and fungal mycelia followed by crop residue decomposition that bind soil particles to each other, thereby forming larger aggregates. Another basic component in various NT and MT systems is crop straw return. The level of SOM has been reported to be low in long-term tillage systems without residue return (Lenka & Lal, 2013).

Reduced tillage resulted in higher soil aggregate stability than CT. These observations are consistent with findings from Eastern and Western Kenya (Gicheru *et al.*, 2004; Kihara *et al.*, 2011), according to which NT and

MT resulted in a higher NSI. Sparrow *et al.* (2006) investigated the possible impacts of tillage systems and plant residue management on soil properties in the long term. They found out that moderate tillage practices increased organic and inorganic materials and helped maintain soil moisture. Zhao *et al.* (2017) reported that management operations significantly affected the stability and size distribution of soil aggregates by changing the SOM in soil and size fractions by highly intense cultivation or management-related hydro regime. Lehrs *et al.* (2012) reported that land use and associated management, such as plant rotation, fertilization, drainage, and irrigation affect soil structure and properties by influencing destruction forces and aggregate forming processes. Moreover, a number of studies have examined the relationship between soil physicochemical properties and aggregate stability (Chaplot & Cooper, 2015). The organic carbon produced by decomposition of plant residuals acts as a cementing agent, which leads to accumulation of soil primary particles and formation of stable aggregates. Conservational tillage as a sustainable agricultural measure, could lead to reversed soil degradation, an increase in crop production, and an improvement in socio-economic conditions of small land-holder farmers (Pradhan *et al.*, 2017).

According to the results, the lowest and highest PAD values were obtained with 70% and 0% of crop residues, respectively. It could be argued that the organic carbon derived from the decomposition of plant residues acted as a cementation agent, which led to the accumulation of primary soil particles. In addition, it reduced the PAD and aggregate stability due to the same force between inorganic particles and organic polymers. Besides, the increased soil aggregate stability had a positive association with the amount of the soil material (Blanco-Canqui & Lal, 2009). This study showed that soils of higher stability are more resistant to soil degradation. Upon reducing tillage and adding plant residues, the PAD index decreased significantly. Soil aggregate stability seems to better reflect the actual vulnerability of topsoils to physical degradation (Stanchi *et al.*, 2015). The maximum amount of NT compared to CT, in terms of soil aggregation, has been reported by Lichter *et al.* (2008) and Kumar *et al.* (2012).

Effect of tillage methods and plant residues on some soil microbiological properties

The application of the NT and MT increased soil microbial population. Hence, modified soil physical and chemical qualities under NT and MT cause significantly different habitats for microorganisms and result in shifts of soil microbial community structure (Kandeler *et al.*, 1999; Helgason *et al.*, 2009).

According to Zhou *et al.* (2020), crop rotation could increase the energy required for microbial life activities

in the soil, produce cementing substances that form soil aggregates, weaken destruction of soil aggregates and decrease the damage to the soil structure.

CT can lead to soil microbial communities prevailed by aerobic microorganisms, while NT and MT practices increase microbial population and activity (Staley, 1999) as well as microbial biomass (Kandeler *et al.*, 1999; Balota *et al.*, 2003). Lupwayi *et al.* (2001) stated that soil disruption by plowing is the major factor in exerting tillage effects. This type of disruption could lead to a population decrease, diversity of soil microorganisms, mechanical degradation, and soil compaction. Doran & Zeiss (2000), in contrast, showed that the number of optional and total anaerobic organisms in no-till soils decreases much less with an increase in depth compared to the number of aerobic bacteria and fungi. In addition, they stated that plowed soils contain a significant number of fungi as well as aerobic and autotrophic nitrifiers compared to NT soils. Some researchers state that soil conservation management could lead to an increase in microbial biomass and microbial activity (Hoitink & Boehm, 1999).

Consuming plant residues could be substantially dependent on the availability of the organic matter. In other words, any factors leading to an increase in the amount of organic carbon in soil could eventually affect soil biological activities (Bastian *et al.*, 2009). Changes in tillage, residues, and rotation practices induce major shifts in the number and composition of soil fauna and flora, including pests and beneficial organisms (Andersen, 1999). However, healthy soil with high disease suppressive potentials will result in sustainable agricultural production only if underlying agronomic management practices support the development of a high soil physical and chemical quality, and if they are economically feasible. Although both NT and CT with plant residues resulted in acceptable amounts of microbial biomass, NT with plant residues increased bacterial and microbial biomass carbon to a greater extent. It may stem from the fact that in NT and MT systems, microbial activity is reduced and more resistance is expected due to plant residues on soil surface. Due to the surface placement of crop residues, those under NT are most likely to be less active so as to balance the carbon mineralization rate to microbial needs, thereby making the system more efficient and perhaps more resilient (Kaschuk *et al.*, 2010). Li *et al.* (2008) stated that the application of mineral and organic fertilizers combined with the return of plant residues increased microbial biomass carbon and microbial biomass nitrogen. In addition, soil microorganisms under CT have low C-utilization efficiency, so they are more metabolically active (Martínez *et al.*, 2013). In most cases, the increased microbial biomass and activity have been attributed to management practices, *i.e.* the maintenance of crop residues and MT, or adding the organic matter directly to soil (Van Bruggen & Semenov, 2000). Lenka & Lal (2013) reported long-term tillage

without crop residue returning leads to a low SOC level. It was shown for various soil types and climatic regions that tillage systems that minimize soil disturbance MT and NT generally increase the storage of SOM compared to conventionally tilled soils (Kushwaha *et al.*, 2001; Jacobs *et al.*, 2009). Wright *et al.* (2007) stated that the application of a MT system could improve the SOC percentage and increase soil microbial population.

Meng *et al.* (2014) reported that the main reason for the decrease in aggregate stability and water-stable aggregates was the reduction in the SOM. Blanco-Canqui & Lal (2008) reported that the amount of the organic matter increased in the surface layers of the soil in NT systems. According to in the results of this research, the organic matter in MT and NT systems with plant residues was higher than in CT; thus, NT and MT states are necessary for protecting the SOM. The strong linearity correlation between MWD and SOC suggested that the SOC content in the WSA of 0.106-0.25 mm played a major role in soil aggregate stability (Zhou *et al.*, 2020).

NT, and also controlled reduced tillage, can be of vital importance to maintain the physical, chemical and biological quality of the soil, in addition to reducing the need for phytosanitary products. According to the findings from this study, it can be concluded that desirable management practices, such as the application of NT systems and plant residues could increase the formation and sustainability of aggregates. In addition, the number of bacteria and the microbial biomass carbon increased significantly in the tillage treatments upon the addition of plant residues. On this basis, we could recommend that a stepwise approach be adopted to the practice of conservation agriculture, especially for resource-constrained farmers. This approach mainly aims to improve biomass productivity and the resultant amount of crop residues retained as soil cover.

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