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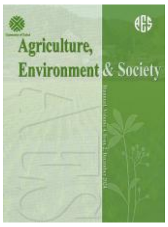

Enhancing soil biological activity and net primary productivity through no-tillage, wheat residues, and corn-bean intercropping

Farideh Akbari [a], Mehdi Dahmardeh [*b], Ali Morshedi [c], Ahmad Ghanbari [b], Soroor Khorramdel [d]

[a] Ph.D Student, Department of Agronomy, Faculty of Agriculture, University of Zabol, Zabol, Iran
[b] Department of Agronomy, Faculty of Agriculture, University of Zabol, Zabol, Iran
[c] Department of Soil and Water Research, Chaharmahal and Bakhtiari Agriculture and Natural Resources Research Center, AREEO, Shahrekord, Iran
[d] Department of Agrotechnology, Faculty of Agriculture, Ferdowsi University of Mashhad, Mashhad, Iran

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ABSTRACT

Conservation management practices play a crucial role in enhancing soil microbial activity, which, in turn, drives improvements in net primary productivity (NPPc) and facilitates efficient carbon allocation. This study presents an innovative approach to examining the synergistic effects of tillage systems, varying wheat residue levels, and corn-bean intercropping patterns on soil biological activity and NPPc. The study was conducted as a carefully designed field experiment, utilizing a split-split plot arrangement within a randomized complete block design (RCBD) with three replications, at the Agricultural Research, Education and Extension Organization in Shahrekord during 2017-2018 growing season. The experiment included three tillage systems (conventional, minimum, and no-tillage) as the main plots, four levels of crop residue (0%, 30%, 60%, and 90% of the straw yield of wheat) as the subplots, and five intercropping patterns (corn monoculture, bean monoculture, and corn-bean ratios of 2:2, 3:1, and 1:3) as the sub-subplots. The results indicated that the highest soil microbial respiration was observed in the no-tillage system combined with 60% wheat residues and a corn-bean intercropping ratio of 2:2. Soil microbial biomass carbon increased significantly under the no-tillage system with 90% wheat residues and the same intercropping ratio of 2:2. The carbon allocation coefficient in seeds, straw, roots, and extra-root structures increased under no-tillage compared to conventional tillage by 10.09%, 11.84%, 52.18%, and 62.67% for corn, and by 12.68%, 7.85%, 35.71%, and 34.92% for beans, respectively. Corn exhibited higher NPPc and allocated more carbon to its aerial organs than beans. This study demonstrates that the combination of no-tillage systems and wheat residue management in diverse intercropping patterns not only synergistically enhances soil microbial activity and NPPc but also promotes soil health and supports sustainable agricultural productivity.

Highlights

- No-tillage combined with 60% wheat residues and a 2:2 corn-bean intercropping ratio achieved the highest soil microbial respiration.
- Soil microbial biomass carbon was maximized under no-tillage with 90% wheat residues and the same 2:2 intercropping pattern.
- Compared to conventional tillage, no-tillage increased carbon allocation to seeds, straw, roots, and extra-root structures by 10-63% in corn and 8-36% in beans.

1. Introduction

Agriculture contributes approximately 13.5% to climate change, primarily through activities such as deforestation, livestock farming, the use of nitrogen fertilizers, and intensive plowing, all of which release and increase greenhouse gases in the atmosphere (Mohammed et al., 2020). Intensive agriculture has led to a significant reduction in soil organic matter, carbon sequestration,

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[*] Corresponding author.
E-mail address: dr.dahmardeh@uoz.ac.ir

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enzyme activities, and ultimately soil quality by performing activities such as intensive tillage to achieve maximum crop production (Cárceles Rodríguez et al., 2022; Hussain et al., 2021). Soil quality is the permanent ability of soil to perform its functions as a vital living system within the ecosystem and under different exploitations, in addition to maintaining biological fertility, enabling the soil to improve the quality of water and air and support human habitation and the health of humans, animals, and plants (Tahat et al., 2020). There is a strong relationship between sustainable agriculture and soil quality. Part of the instability of agricultural systems is due to the reduction of soil quality over time, so maintaining its quality is an important strategy for economic progress and improving environmental quality (Cárceles Rodríguez et al., 2022; Tahat et al., 2020). Based on this, the selection of the type of management operations and land exploitation should be made taking into account the maintenance of soil quality and its improvement as well as carbon sequestration (Cárceles Rodríguez et al., 2022; Lal et al., 2018).

Carbon sequestration is a method of decreasing carbon dioxide (CO_2) emissions into the atmosphere by involving long-term storage of CO_2 in soil, which helps to mitigate the effects of global warming (Hussain et al., 2021). Increasing carbon sequestration improves soil fertility, increases soil water holding capacity, prevents water and wind erosion, and ultimately increases biomass production and plant productivity (Ghimire et al., 2022). By increasing carbon sequestration in the soil, reducing atmospheric carbon concentration, and decreasing greenhouse gas emissions, conservation management systems (such as the addition of plant residues, conservation tillage, cultivation of perennial plants, maintaining crop straw on the soil surface, rotation, and multi-cropping systems), as the most important management strategy, can play a role in mitigating the effects of climate change, improving soil quality, and achieving agricultural sustainability (Cárceles Rodríguez et al., 2022).

In production systems, conversion of conventional tillage to conservation tillage (minimum tillage and no-tillage) can transform the soil system from a source of carbon dioxide production to its reservoir. This plays a crucial role in enhancing soil quality, decreasing the entry of carbon dioxide into the atmosphere, and combating climate change (Hussain et al., 2021). In conservation tillage systems, the reduced use of agricultural machinery leads to lower consumption of fossil fuels and, consequently, less carbon dioxide released into the atmosphere. Additionally, more plant residues are left on the soil surface (Wang et al., 2020a). Conservation tillage reduces the mineralization of organic matter by reducing soil disturbance. The result is that a larger soil organic carbon reserve is created compared to conventional tillage (Cárceles Rodríguez et al., 2022). The positive effects of conservation tillage on soil quality, environment, and soil water conservation as compared to conventional tillage were highlighted by many researchers (Mondal et al., 2020).

Plant residues, as a suitable conservation agricultural management, are the main source of carbon input to agricultural ecosystems, so returning them to the soil can enhance the soil's organic carbon content and mitigate climate change. This is linked to plant yield promotion and ecologically sustainable agriculture (Wang et al., 2020b). Plant residues can maintain soil fertility, increase the concentration of soil organic matter, conserve water in the soil, reduce evaporation, reduce temperature fluctuations, improve soil quality (physical, chemical, and biological properties) and stimulate microbial activities (Fu et al., 2021). Improving the activity of microorganisms and higher soil microbial biomass carbon (MBC) and nitrogen due to the return of plant residues to the soil were reported by many researchers (Choudhary et al., 2018; Li et al., 2018). Intercropping is an efficient planting strategy that maximizes the benefits of time and space, reduces the loss of water, soil, light, fertilizer, and other resources, increases diversity in crop production, suppresses weeds and insect pests, enhances crop yields, decreases soil water evaporation and leakage, improves crop water-use efficiency, promotes the rapid growth of soil microorganisms, and maximizes soil advantages and quality (Xiao et al., 2023). Compared to mono-cropping, intercropped soils have higher microbial biomass and respiration (Chen et al., 2019), which may be because intercropping can enhance biomass and litter, and promote nitrogen production (Cong et al., 2015).

In addition to the physicochemical properties of the soil, its biological properties (e.g., microbial biomass and respiration) are also important for soil quality evaluation. Microbial biomass is responsible for the decomposition of plant residues and the release of nutrients in the soil and acts as an accessible source of nutrients. Microbial biomass carbon is an estimate of the activity and vita of the soil microbial mass and is one of the indicators sensitive to environmental variables. This parameter is used to analyze the effect of oil management practices, environmental factors and stresses on soil microbial population (Gayan et al., 2023; Ramesh et al., 2019). Soil microbial respiration is one of the oldest characteristics of determining microbial activity in the soil, which indicates the availability of soil organic matter and the intensity of its decomposition (Feketeová et al., 2021). Humidity, temperature, nutrient availability, soil structure, soil carbon storage, substrate quality and quantity, microbial activity and biomass, land management and use, and fertilizer consumption are among the factors that affect microbial respiration (Anjum and Khan, 2021; Bian et al., 2022; Fang and Moncrieff, 2005). By increasing the amount of available oxygen for soil microorganisms, conventional tillage increases microbial activities, which leads to further decomposition of soil organic matter, and subsequently, soil quality decreases. This has been reported by many researchers (Akbolat et al., 2009; Cárceles Rodríguez et al., 2022). The researchers stated that the no-tillage system with residue retention resulted in higher soil MBC, nitrogen, and microbial quotients (Li et al., 2018). In a continuous rice and wheat rotation, no-tillage, and residue cycling compared to conventional tillage and residue removal raised soil MBC by 29% and 56%, respectively (Choudhary et al., 2018). In a pigeon pea-soybean intercropping system, conservation tillage systems recorded significantly higher soil MBC and nitrogen levels than conventional tillage without crop residues (Kumar and Babalad, 2018). Increasing the organic manure levels from 0 to 20 Mg ha^{-1} has increased the soil microbial respiration by 88% (Anjum and Khan, 2021).

Plants can store much organic carbon in their above-ground and below-ground organs by absorbing carbon dioxide during photosynthesis. Since the type of agricultural system management has a significant effect on the amount of plant production, it is necessary to pay attention to the amount of net primary production (NPP) in plants as an estimate of the amount of carbon dioxide absorbed by plants (Lambers et al., 2008). In other words, determining the NPP of plants in different agricultural systems can be considered as a sustainable strategy to reduce carbon dioxide concentration in the atmosphere (Mohammadi et al., 2017). Net primary production is the amount of carbon fixed in different above-ground and below-ground organs of the plant, which returning it to the soil can decrease the concentration of carbon dioxide in the atmosphere and increase carbon sequestration in the soil (Gan et al., 2009; Lambers et al., 2008). The researchers stated that any management factor (tillage type and crop residues) that is effective in the growth and development of the plant significantly affects the net production of the plant, which represents the absorbed carbon (Mohammadi et al., 2017; Ryals and Silver, 2013). In this order, determining the amount of NPP in plants on the one hand shows the production of plant biomass and, on the other hand, it is an estimation of the amount of carbon dioxide absorbed from the atmosphere. Therefore, it seems that the selection of plant species such as corn that have higher biomass production and also using conservation management can be considered as a sustainable strategy to increase carbon sequestration in the soil and reduce carbon dioxide concentration in the atmosphere in the future. In addition, by calculating the amount of primary net production and estimating the relative coefficients of carbon allocation to different organs of important plant species, the contribution of each plant organ can be estimated from the amount of carbon dioxide absorbed. Considering that by choosing the most appropriate tillage method, cultivation pattern, and nutritional management, it is possible to increase the amount of carbon allocated in different organs of plants and prevent the release of carbon into the atmosphere; Therefore, the present study was conducted to investigate the effect of different tillage systems, intercropping, and plant residue management on soil biological activity, NPP, and carbon allocation to different organs of corn (*Zea mays* L.) and bean (*Phaseolus vulgaris* L.).

2. Materials and methods

2.1. Experimental design and treatments

This experiment was done as a split-split plot arrangement in a randomized complete block design with three replications. The main factor was the tillage system, which had three levels: 1- Conventional tillage (Moldboard plow and disc), 2- Minimum tillage (disc and furrower), and 3- No-tillage. Crop residues as a sub-factor included four levels (0, 30, 60, and 90% of the weight of wheat residues). The intercropping pattern of corn (K.S.C. 704 cultivar) and bean (Sadri cultivar) was a sub-sub factor. Intercropping by alternative method had five levels, including corn sole cropping, bean sole cropping, and corn and bean ratios of 2 corn rows:2 bean rows, 3 corn rows:1 bean row, and 1 corn row:3 bean rows.

2.2. Preparation of seedbed and cultivation

This experiment was carried out during the growing season of 2017 in the Research Farm of the Agricultural Research, Education and Extension Organization of Shahrekord (Longitude: 50˚56′ E, Latitude: 32˚21′ N, Altitude above sea level: 2066 m). The average rainfall and the average temperature during the growing season are presented in Table 1. Temperature plays an important role in determining the population of soil fungi. Most fungi grow optimally at a temperature of 15 to 22 degrees Celsius and excessive temperature increase has an adverse effect on them. In the present study, the increase in microbial biomass carbon in the second sampling stage (100 days after planting) indicates that the temperature is favorable for fungal activity (Table 4). Before sowing the seeds, the physicochemical properties of soil (depth 0 to 30 cm) were measured according to the standard methods (Westerman, 1990) and are presented in Table 2.

Table 1. Data on average monthly temperature and total monthly precipitation for the year 2017

Month	Average monthly temperature (˚C)	Total monthly precipitation (mm)
20 March – 19 April	10.3	74.2
20 April – 20 May	17.0	4.9
21 May – 20 June	21.2	0.0
21 June – 21 July	25.2	0.0
22 July – 21 August	22.3	0.0
22 August – 21 September	18.6	0.0
22 September – 21 October	12.3	1.7

Table 2. Physico-chemical properties of soil in the Research Farm

Soil texture	pH	Electrical conductivity (ds m^{-1})	Organic carbon (%)	Total N (%)	P (ppm)	K (ppm)
Clay Loam	7.86	0.735	0.624	0.087	8.6	217

In the early spring of 2017, a piece of field with an area of, 2400 m^2 was selected. To apply the treatment of crop residues, the residues of wheat field cultivated in the fall of the previous year, which amounted to 8200 kg ha^{-1}, were weighed, and the required amount was added to the field according to the relevant treatments. Then, the tillage operation was done. The chemical characteristics of the wheat residues used are presented in Table 3. It should be noted that in this experiment, the C:N ratio was considered to be equal to 20 to prevent the immobilization of soil nitrogen by microorganisms (Barbarick, 2006). For this purpose, according to the C:N ratio of wheat residues (Table 3), nitrogen mineral fertilizer (urea) was added to the residues before planting to bring the C:N ratio of the residues to the desired amount. The amount of urea fertilizer required to prevent immobilization was estimated at 43 kg ha^{-1}.

Table 3. Chemical characteristics of wheat residues

C (g kg^{-1})	**K (g kg^{-1})**	**P (g kg^{-1})**	**N (g kg^{-1})**	**C: N**	**C: P**
520	11.1	2	8.6	60.46	**260**

The field used was divided into three blocks, and each block was divided into three main plots with a distance of 2.5 m from each other. Each main plot was divided into four sub plots with a distance of 2 m from each other to apply wheat residue treatment. Then, each of the sub plots was divided into five parts with a distance of 1 m to apply the treatment of intercropping patterns. Each sub-sub-plot had six planting rows with a length of 2 m and a row distance of 60 cm. The distance between the two plants on the row for bean and corn was considered to be 5 cm and 20 cm, respectively. The plant density for beans and corn was considered to be 33 and 8 plants m^{-2}, respectively. Sowing depth was 3-5 cm. Seeds were prepared from the Agricultural Research, Education, and Extension Organization of Shahrekord and disinfected by Mancozeb fungicide at a rate of one per thousand. Sprinkler irrigation was done at the beginning of the growing season with an interval of four days and after that with an interval of one week. Sowing and weed control were done manually by workers. Microbiological indicators were evaluated as follows.

2.3. Microbial biomass carbon

For this purpose, soil samples (depth 0-30 cm) were prepared from all the plots during two stages of the growing season (60 days after planting, equal to the flowering of beans, and the end of vegetative growth of corn, and 100 days after planting, equal to the filling of the bean pods, and the seed dough stage of corn). Soil samples were fumigated with chloroform and extracted with potassium sulfate solution (Sparling and West, 1988). Organic carbon in the resulting extract was determined and converted to microbial biomass carbon (MBC). First, wet field soil (150 g) was fumigated with chloroform for 24 h (150 g of soil for the control sample without fumigating with chloroform). Fumigated soil (5 g) was mixed with 25 mL of potassium sulfate solution (0.5 M). The resulting mixture was shaken for 30 minutes and then filtered. The same steps were performed for the control soil (without fumigation). The organic matter in the extract was decomposed by potassium dichromate solution. The remaining potassium dichromate was measured through titration with ferrous ammonium sulfate (Schwedt and Schnepel, 1981). For this, in a round-bottom flask, soil extract (10 mL) was mixed with potassium dichromate solution (10 mL) and 10 mL of concentrated sulfuric acid containing silver sulfate. Then, distilled water (100 mL) and detector solution (0.3 mL), ferrous ammonium sulfate, were added. In the case of the control, it was done in the same way. The amount of soil organic carbon was calculated from the following formula:

$$\frac{(\mathrm{B}-\mathrm{S}\ \&\ \mathrm{C})\times 0.3\times \mathrm{V}\times 1000\times 100}{\mathrm{ml}\times \mathrm{sw}\times \%\ \mathrm{dm}}=\mathrm{mgc}\ (\mathrm{Organic})\mathrm{g}^{-1}\mathrm{dm} \qquad (1)$$

B: The average volume of ferrous ammonium sulfate consumed for the standard control (mL),

S: The average volume of ferrous ammonium sulfate consumed for the samples (mL)

C: The average volume of ferrous ammonium sulfate consumed for the control of samples (mL)

0.3: Conversion factor (1 mL of 0.1 M ferrous ammonium sulfate is equivalent to 0.3 mg of carbon)

1000: Conversion factor

V: Extract volume (mL), Ml: Filtered extract volume (mL), SW: Initial soil weight (mL), dm: Dry mass calculation factor (mL).

Finally, the organic carbon obtained from the above calculations was placed in the following formula and the MBC in mg of carbon per 100 g of dry soil was obtained.

$$\mathrm{MBC}=\frac{\mathrm{S}-\mathrm{C}}{0.35} \qquad (2)$$

S: The average number of mg of organic carbon per 100 g of fumigated dry soil; C: The average number of mg of organic carbon per 100 g of dry soil of control (without fumigation); 0.35: The conversion factor of organic carbon to microbial carbon

2.4. Soil microbial respiration

This trait was measured using closed containers and titration of the remaining soda with acid (Anderson et al., 1993; Stotzky, 1965). First, soil (50 g) was poured into the bottom of the container. Then 0.1 M soda (25 mL) was poured into a small beaker and placed inside the container containing the soil sample, and the lid of the container was closed. Also, three to five containers without soil but containing soda (as a control) were prepared and placed in the incubator at 25 °C for three days and nights. After the end of the incubation period, the amount of remaining soda was measured through titration. In this way, the available soda was transferred to the Erlenmeyer flask, and 0.1 M barium chloride (5 mL) was added to it. Then, 3 to 4 drops of phenolphthalein were added, and the remaining soda was titrated with hydrochloric acid (0.1 N) while shaking (until the color changed from red to colorless). Finally, the respiration rate was calculated with the following formula.

$$\mathrm{CO_2}=\frac{(\mathrm{V_0}-\mathrm{V})\times 1.1}{\mathrm{dwt}}=\frac{\mathrm{mg}}{\mathrm{sw}/\mathrm{t}} \qquad (3)$$

t: Incubation time (h), V0: Average acid consumption for control sample titration (mL), V: Amount of acid used for soil sample titration (mL), dwt: Dry weight of 1 g of wet soil, Sw: Dry soil weight (g), 1.1: Conversion factor (1 mL of 0.05 M NaOH is equal to 1.1 mg of CO_2)

2.5. Metabolic quotient

The metabolic quotient was calculated as the ratio between the CO_2 (mg of carbon) released/h from each g of soil and the soil MBC (g) and reported in mg C-CO_2 mg^{-1} MBC day^{-1} (Anderson et al., 1993).

2.6. Evaluation of primary net productivity and carbon allocation to plant organs

The primary net productivity of organs was calculated from the total carbon allocated to seed (CP), other aerial organs or straw (CS), root (CR), and extra-root (CE) (Bolinder et al., 2007). At the end of the growing season, five plants from each plant were randomly selected and harvested completely from the soil along with the roots. The plant samples were divided into two parts, shoot and root, and weighed. The ratio of shoot to root was calculated by weight for each plant. The amount of net primary productivity based on carbon (NPPc) for each plant was calculated through the following formula (Houghton et al., 2001).

$$NPPc = CP + CS + CR + CE \tag{4}$$

The amount of C in each of these four fractions (and thus also NPP) can be estimated from agricultural yields, using published or assumed values for harvest index (HI), S:R ratios, plant C in root exudates, and C concentrations in the plant parts. In a grain crop, for example, assuming the C concentration of all plant parts is 0.45 g g^{-1} (Bolinder et al., 2007). Therefore, the following formulas were used to calculate allocated carbon content to different organs:

$$CP = \text{Economic organ yield} \times 0.45 \tag{5}$$
$$CS = (\text{Economic organ yield} \ (1\text{-HI}) / \text{HI}) \times 0.45 \tag{6}$$
$$CR = (\text{Economic organ yield} / S{:}R \times HI) \times 0.45 \tag{7}$$
$$CE = CR \times 0.65 \tag{8}$$

NPPc, Net primary production based on carbon in the whole plant; CP, Carbon in the economic organ; CS, Carbon in the straw; HI, harvest index; CR, Carbon in the root; S:R, the ratio of shoot to root; CE, Carbon in extra-root.

Carbon in root secretions based on different references (Bolinder et al., 2007; Gill et al., 2002) was considered to be about 65% of the carbon in the root. The relative coefficients of carbon allocation to different organs were calculated with the following formulas (Bolinder et al., 2007).

$$RP = CP / NPPc \tag{9}$$
$$RS = CS / NPPc \tag{10}$$
$$RR = CR / NPPc \tag{11}$$
$$RE = CE / NPPc \tag{12}$$

In the above formulas, RP, RS, RR, and RE represent the relative coefficient of carbon allocation to seeds, other aerial organs, roots, and extra-root, respectively.

2.7. Statistical Analysis

A split-split plot arrangement in a randomized complete block design with three replications was used for analysis. The first factor was three tillage systems. The second factor was four levels of wheat residues. The third factor was five intercropping patterns. Data were tested for normality before statistical analysis and were normal in all traits. Statistical analysis of variance was done in a split-split plot arrangement using the general linear model procedure of SAS statistical software version 9.2 (SAS, Institute Inc. 2009). Differences between means were compared by the Duncan multiple range test ($p \leq 0.05$).

3. Results

3.1. Soil microbial biomass and respiration

These parameters were investigated in this study as criteria for evaluating soil biological activity under the influence of tillage system, crop residues, and intercropping patterns. The tillage systems, crop residues, intercropping pattern, and their triple interaction had a significant effect ($p \leq 0.05$) on soil microbial biomass and respiration. Based on the results, the highest soil microbial respiration in both sampling stages (60 and 100 days after planting) was related to the no-tillage system along with 60% wheat residues and the intercropping ratio of 2 corn rows: 2 bean rows (Table 4). The lowest microbial respiration was observed in pure corn cultivation along with conventional tillage and 30% wheat residues, which was not statistically different from 0% wheat residues (Table 4). In the no-tillage system, the increase in the percentage of wheat residues in all three ratios of intercropping led to an increment in soil microbial respiration compared to sole cropping systems (Table 4).

In general, the simultaneous presence of species with different functions in intercropping systems led to an increase in soil microbial respiration compared to sole cropping systems. Superior cultivation patterns compared to the corn and bean sole cropping systems led to an increase of 49.50%, 24.79%, 44.10%, and 34.14% in soil microbial respiration during 60 and 100 days after planting, respectively (Table 4).

The triple interaction of tillage systems, wheat residues, and intercropping on soil MBC showed that the highest amount in both sampling stages (60 and 100 days after planting) was related to the no-tillage system along with 90% wheat residues and the intercropping ratio of 2 corn rows: 2 bean rows. The lowest amount of soil MBC was obtained in the conventional tillage systems along with 0% wheat residues and pure corn cultivation (Table 5; Figure 1).

Conservation tillage systems lead to reduced soil disturbance and greater retention of crop residues on the soil surface. These factors contribute to soil fertility, aggregate stability, improved water infiltration, better temperature and pH regulation, increased nutrient availability, and organic carbon accumulation. As a result, conservation tillage significantly influences soil microbial activity and subsequently affects plant growth (Lal, 2007). On the other hand, increasing tillage intensity accelerates the decomposition of organic matter, reducing the essential substrate for the growth and activity of soil microorganisms.

In this regard, Abbott and Murphy (2007) reported that tillage operations have a significant impact on soil microbial communities, and intensive tillage practices lead to a long-term decline in soil microbial growth and activity. Similarly, Chirinda et al. (2010) stated that the lowest microbial respiration rates were observed under intensive tillage systems.Studies on no-till systems have shown that microbial populations and, consequently, CO_2 production from microbial activity are significantly higher in no-till systems compared to intensive tillage systems (Levanon et al., 1993).

Other studies have shown that the addition of plant residues to soil significantly increases CO_2 emissions, which reflect the respiratory activity of soil microorganisms and the decomposition of added organic matter (Hadas et al., 2004).

Table 4. Mean comparison of the soil microbial respiration (mg Co_2 g^{-1} soil day) under the influence of the triple interaction of tillage systems, wheat residues, and corn-bean intercropping patterns during two stages of sampling

		60 days after planting				100 days after planting			
		Wheat residues (%)				Wheat residues (%)			
Tillage systems	Intercropping pattern	0	30	60	90	0	30	60	90
Conventional tillage									
	Corn sole cropping	0.0082^{x}	0.0124^{v-x}	0.0201^{r-x}	0.022^{r-w}	0.051^{xz}	0.034^{z}	0.095^{v}	0.098^{v}
	Bean sole cropping	0.0085^{x}	0.016^{u-x}	0.023^{r-v}	0.03^{q-t}	0.052^{x-z}	0.034^{z}	0.097^{v}	0.141^{qt}
	2 corn rows:2 bean rows	0.0208^{r-x}	0.024^{r-v}	0.033^{qp}	0.052^{no}	0.075^{w}	0.061^{w-y}	0.101^{v}	0.171^{n-p}
	3 corn rows:1 bean row	0.0103^{wx}	0.017^{t-x}	0.026^{q-u}	0.046^{op}	0.057^{w-y}	0.043^{yz}	0.099^{v}	0.153^{p-r}
	1 corn row:3 bean rows	0.0177^{s-x}	0.0203^{r-x}	0.031^{q-s}	$0.049^{o}p$	0.061^{w-y}	0.046^{yz}	0.099^{v}	0.160^{o-q}
Minimum tillage									
	Corn sole cropping	0.0203^{r-x}	0.051^{op}	0.065^{nm}	0.107^{ij}	0.071^{wx}	0.188^{l-n}	0.131^{s-u}	0.116^{uv}
	Bean sole cropping	0.024^{r-v}	0.057^{no}	$0.^{087k}$	0.119^{e-i}	0.097^{v}	0.21^{3h-k}	0.142^{q-t}	0.126^{ut}
	2 corn rows:2 bean rows	0.052^{no}	0.0806^{kl}	0.116^{f-i}	0.132^{b-e}	0.149^{q-s}	0.235^{fg}	0.221^{g-j}	0.208^{h-l}
	3 corn rows:1 bean row	0.039^{qp}	0.076^{k-m}	0.1^{j}	0.129^{e-f}	0.099^{v}	0.214^{g-j}	0.179^{m-o}	0.136^{r-t}
	1 corn row:3 bean rows	0.047^{op}	0.079^{kl}	0.106^{ij}	0.131^{c-e}	0.137^{r-t}	0.224^{g-j}	0.203^{j-l}	0.178^{m-o}
No-tillage									
	Corn sole cropping	0.0459^{op}	0.072^{lm}	0.101^{j}	0.111^{h-j}	0.192^{k-m}	0.209^{h-l}	0.229^{f-h}	0.221^{g-j}
	Bean sole cropping	0.053^{no}	0.0807^{lk}	0.121^{e-h}	0.113^{g-j}	0.205^{i-l}	0.227^{f-i}	0.246^{ef}	0.226^{f-i}
	2 corn rows:2 bean rows	0.072^{lm}	0.124^{d-h}	0.151^{a}	0.136^{b-d}	0.262^{ed}	0.263^{e-d}	0.33^{a}	0.309^{b}
	3 corn rows:1 bean row	0.072^{lm}	0.117^{f-i}	0.143^{a-c}	0.116^{f-i}	0.257^{e}	0.234^{gf}	0.302^{b}	0.28^{cd}
	1 corn row:3 bean rows	0.072^{lm}	0.121^{e-h}	0.144^{ab}	0.126^{d-g}	0.257^{e}	0.256^{e}	0.311^{b}	0.294^{bc}

In each stage of sampling, data with the same letter are not significantly different (Duncan multiple range test $p \leq 0.05$)

Table 5. Mean comparison of the soil microbial biomass carbon (mg C g^{-1} soil) under the influence of the triple interaction of tillage systems, wheat residues, and corn-bean intercropping patterns during two stages of sampling

		60 days after planting				100 days after planting			
		Wheat residues (%)				Wheat residues (%)			
Tillage systems	Intercropping pattern	0	30	60	90	0	30	60	90
Conventional tillage									
	Corn sole cropping	1.61^{x}	2.99^{v-x}	4.85^{s-u}	8.71^{op}	3.01^{d}	3.84^{cd}	10.53^{za}	14.75^{x}
	Bean sole cropping	1.64^{x}	3.12^{v-x}	5.85^{r-t}	8.78^{op}	3.96^{cd}	3.97^{cd}	10.96^{z}	16.33^{uw}
	2 corn rows:2 bean rows	2.83^{v-x}	4.34^{t-v}	7.1^{qr}	10.75^{n}	5.05^{bc}	6.41^{b}	12.95^{y}	18.84^{t}
	3 corn rows:1 bean row	2.38^{wx}	3.53^{u-w}	5.63^{r-t}	10.18^{no}	4.08^{cd}	4.26^{cd}	11.52^{yz}	17.02^{uv}
	1 corn row:3 bean rows	2.6^{v-x}	3.68^{u-w}	6.8^{qr}	10.57^{n}	4.16^{cd}	4.97^{bc}	11.6^{yz}	17.13^{u}
Minimum tillage									
	Corn sole cropping	6.66^{qr}	18.49^{kl}	20.64^{j}	25.44^{h}	9.27^{z}	24.19s	25.82^{p-r}	27.06^{n-q}
	Bean sole cropping	6.17^{q-s}	18.52^{kl}	22.4^{i}	29.08^{g}	11.57^{yz}	25.13^{rs}	25.89^{p-r}	28.28^{mn}
	2 corn rows:2 bean rows	9.6^{no}	20.21^{jk}	23.7^{gi}	30.53^{g}	15.54^{v-x}	26.56^{o-r}	27.89^{m-o}	37.71^{l}
	3 corn rows:1 bean row	7.83^{qp}	19.4^{kj}	22.43^{i}	29.37^{g}	12.02^{yz}	25.2^{rs}	26.86^{n-q}	29.41^{m}
	1 corn row:3 bean rows	8.88^{op}	20.18^{jk}	23.64^{i}	30.42^{g}	15.13^{wx}	25.52^{q-s}	27.2^{n-p}	32.34^{l}
No-tillage									
	Corn sole cropping	16.8^{m}	37.97^{f}	39.37^{e-f}	67.92^{c}	36.84^{k}	45.41^{i}	59.91^{f}	60.34^{f}
	Bean sole cropping	17.5^{lm}	38.4^{ef}	39.4^{e-f}	68.05^{c}	37.64^{jk}	45.45^{i}	63.04^{e}	60.92^{f}
	2 corn rows:2 bean rows	19.85^{kj}	39.85^{ed}	40.74^{d}	75.11^{a}	39.13^{j}	50.34^{g}	86.76^{ab}	87.5^{a}
	3 corn rows:1 bean row	18.83^{kl}	38.97^{e-f}	39.41^{e-f}	68.63^{c}	37.84^{jk}	45.57^{i}	67.73^{d}	84.06^{c}
	1 corn row:3 bean rows	19.68^{kj}	38.98^{e-f}	40.3^{d}	70.75^{b}	37.92^{jk}	47.63^{h}	85.39^{bc}	85.81^{b}

In each stage of sampling, data with the same letter are not significantly different (Duncan multiple range test $p \leq 0.05$)

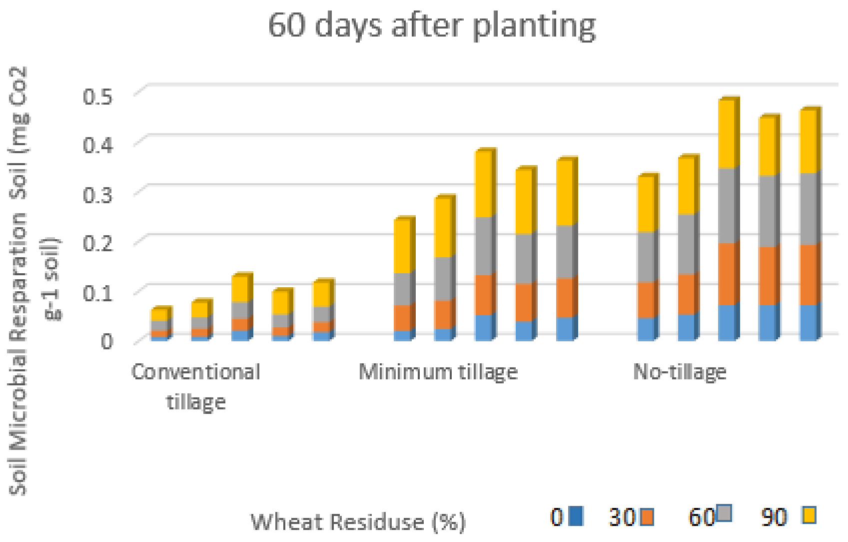

Figure 1. The interaction of wheat residues and tillage systems on soil microbial respiration.

3.2. Metabolic quotient

The triple interaction of tillage, wheat residues, and intercropping on the metabolic quotient was significant ($p \leq 0.05$) only in the second sampling (100 days after planting) and had no effect on this trait in the first sampling. According to the results obtained from sampling 100 days after planting, the highest amount of metabolic quotient was obtained in the common tillage system along with 0% wheat residues and pure corn cultivation. The lowest amount was related to the no-tillage system along with 90% wheat residues and pure corn cultivation, which was not significantly different from pure bean cultivation (Table 6).

Table 6. Mean comparison of the soil metabolic quotient (mg C-CO$_2$ mg^{-1} MBC day^{-1}) under the influence of the triple interaction of tillage systems, wheat residues, and corn-bean intercropping patterns at the sampling of 100 days after planting

Tillage systems	Intercropping pattern	Wheat residues (%)			
		0	30	60	90
Conventional tillage					
	Corn sole cropping	1.7^{a}	$0.885^{\text{e-i}}$	$0.902^{\text{e-h}}$	0.664^{no}
	Bean sole cropping	1.31^{c}	$0.856^{\text{ e-k}}$	$0.885^{\text{e-i}}$	$0.863^{\text{e-k}}$
	2 corn rows:2 bean rows	1.48^{b}	$0.951^{\text{d-f}}$	$0.779^{\text{i-m}}$	$0.907^{\text{e-g}}$
	3 corn rows:1 bean row	1.39^{c}	1.00^{d}	$0.859^{\text{e-k}}$	$0.898^{\text{e-g}}$
	1 corn row:3 bean rows	1.46^{b}	$0.925^{\text{d-g}}$	$0.836^{\text{f-k}}$	$0.934^{\text{e-g}}$
Minimum tillage					
	Corn sole cropping	$0.765^{\text{j-n}}$	$0.777^{\text{k-n}}$	0.507^{qr}	$0.428^{\text{r-u}}$
	Bean sole cropping	$0.838^{\text{f-k}}$	$0.847^{\text{e-k}}$	$0.548^{\text{p-r}}$	$0.445^{\text{q-u}}$
	2 corn rows:2 bean rows	$0.958^{\text{d-f}}$	$0.884^{\text{e-i}}$	$0.789^{\text{ h-l}}$	0.551^{pq}
	3 corn rows:1 bean row	$0.823^{\text{g-k}}$	$0.849^{\text{e-k}}$	$0.666^{\text{ l-o}}$	$0.462^{\text{q-t}}$
	1 corn row:3 bean rows	$0.905^{\text{e-g}}$	$0.877^{\text{e-i}}$	$0.746^{\text{ k-n}}$	0.55^{pq}
No-tillage					
	Corn sole cropping	0.521^{pq}	$0.46^{\text{q-t}}$	$0.382^{\text{s-v}}$	$0.366^{\text{t-w}}$
	Bean sole cropping	0.544^{qr}	$0.499^{\text{q-s}}$	$0.411^{\text{r-u}}$	$0.37^{\text{t-w}}$
	2 corn rows:2 bean rows	$0.669^{\text{ l-o}}$	0.522^{pq}	$0.38^{\text{ s-v}}$	$0.353^{\text{u-w}}$
	3 corn rows:1 bean row	$0.679^{\text{ l-o}}$	0.513^{qr}	$0.445^{\text{q-u}}$	0.333^{vw}
	1 corn row:3 bean rows	$0.677^{\text{ l-o}}$	0.532^{pq}	$0.364^{\text{t-w}}$	$0.342^{\text{u-w}}$

Data with the same letter are not significantly different (Duncan multiple range test $p \leq 0.05$)

3.3. Net primary productivity based on carbon

Based on the results, the triple interaction of tillage, wheat residues, and intercropping had a significant effect ($p \leq 0.05$) on the NPPc in both corn and bean plants. Results indicated that the highest amount of NPPc for corn (1173.72 g m^{-2} year) was obtained in the no-tillage system along with 90% wheat residues and pure corn cultivation, and there was no significant difference between no-tillage and minimum tillage.

Greater biomass production in maize resulted in a higher net primary production of carbon (NPPc) in pure maize cultivation compared to other cropping patterns. Consequently, in mixed cropping systems where maize had a higher proportion, NPPc was also greater. Wang et al. (2010) suggested that to enhance carbon sequestration efficiency in agriculture, cropping systems should be designed to include crop rotation and intercropping, which increase biomass production and consequently enhance carbon sequestration in soil. Based on experimental results, it can be concluded that cropping systems should be designed to include multiple plant species, ensuring high plant diversity and incorporating components with high NPPc. This approach not only increases the quantitative potential for carbon sequestration but also improves the qualitative aspect by diversifying organic matter inputs to the soil. Management practices aimed at increasing plant production can significantly enhance soil carbon storage potential (Derner and Schuman, 2007).

The lowest amount (828.47 g m^{-2} year) was related to the conventional system along with 0% wheat residues and the intercropping ratio of 1 corn row: 3 bean rows (Table 7). The highest amount of NPPc for bean (58.34 g m^{-2} year) was achieved in the no-tillage system, along with 90% wheat residues and the intercropping ratio of 3 corn rows: 1 bean row. Its lowest amount for bean was related to the conventional system, along with 0% wheat residues and pure bean cultivation (Table 7).

Among the intercropping patterns, the highest and lowest NPPc for beans were obtained in the ratio of 3 corn rows: 1 bean row and pure bean cultivation, respectively. Also, the highest and lowest NPPc for corn among intercropping patterns were observed in pure corn cultivation and 1 corn row: 3 bean rows, respectively (Table 7). By increasing the percentage of wheat residues, the amount of NPPc in both corn and bean plants increased (Table 7). In each tillage system, the amount of bean NPPc was higher in different patterns of intercropping compared to pure bean cultivation. For example, in the no-tillage system, the ratio of 2 corn rows:2 bean rows along with 30%, 60%, and 90% wheat residues, respectively, caused an increase of 8.77%, 6.22%, and 15.38% in the amount of bean NPPc compared to pure bean cultivation (Table 7).

Table 7. Mean comparison of the net primary productivity based on carbon (NPPc) in corn and bean plants (g m^{-2} year) under the influence of the triple interaction of tillage systems, wheat residues, and corn-bean intercropping patterns

		Corn				Bean			
		Wheat residues (%)				Wheat residues (%)			
Tillage systems	**Intercropping pattern**	**0**	**30**	**60**	**90**	**0**	**30**	**60**	**90**
Conventional tillage									
	Corn sole cropping	902.61^{pq}	943.28^{m-o}	995.78^{h-k}	984.81^{i-l}	-	-	-	-
	Bean sole cropping	-	-	-	-	18.7 ^{t}	29.24 $^{l-n}$	30.67 ^{lm}	30.32^{l-n}
	2 corn rows:2 bean rows	891.87^{q}	940.94^{m-p}	994.83^{h-k}	968.13^{j-m}	22.09^{rs}	34.97 ^{k}	37.08 ^{k}	36.63 ^{k}
	3 corn rows:1 bean row	835.27^{r}	935.86^{m-p}	965.04 $^{k-m}$	962.66 $^{k-m}$	23.16^{q-s}	35.11 ^{k}	37.68 ^{k}	38.07 ^{k}
	1 corn row:3 bean rows	828.47^{r}	929.04^{m-q}	948.27 $^{l-o}$	954.02^{l-n}	20.71^{ts}	29.59 $^{l-n}$	36.67 ^{k}	30.91 ^{l}
Minimum tillage									
	Corn sole cropping	922.06^{n-q}	1031.5^{d-h}	1071.79^{c}	1167.9^{a}	-	-	-	-
	Bean sole cropping	-	-	-	-	23.17 $^{q-s}$	37.75 ^{k}	45.11^{h-j}	44.7 $^{h-j}$
	2 corn rows:2 bean rows	921.77^{n-q}	1025.51^{e-h}	1069.06^{cd}	1062.41^{c-e}	27.93^{m-p}	41.97 ^{j}	54.32^{bc}	48.82^{d-f}
	3 corn rows:1 bean row	916.54^{n-q}	1008.92^{g-i}	1068.69^{cd}	1058.7^{c-f}	28.63 $^{l-o}$	46.79^{e-i}	56.32^{ab}	51.49^{cd}
	1 corn row:3 bean rows	845.82 ^{r}	1005.56^{g-i}	1033.8^{c-h}	1034.35^{c-h}	24.69^{p-r}	38.19 ^{k}	47.28^{e-i}	47.78 $^{e-h}$
No-tillage									
	Corn sole cropping	934.04^{m-p}	1021.47^{f-i}	1148.47^{a}	1173.72 ^{a}	-	-	-	-
	Bean sole cropping	-	-	-	-	23.16^{q-s}	37.71^{k}	38.2 ^{k}	45.43^{f-i}
	2 corn rows:2 bean rows	919.41^{n-q}	1018.22^{g-i}	1107.77^{b}	1067.7^{cd}	27.05^{n-p}	44.23^{ij}	48.63 $^{d-g}$	49.8 ^{de}
	3 corn rows:1 bean row	910.58^{o-q}	1008.23^{g-i}	1113.97^{b}	1156.1 ^{a}	27.53^{m-p}	44.25^{ij}	48.82 $^{d-f}$	58.43 ^{a}
	1 corn row:3 bean rows	851.71^{r}	996.76^{g-k}	1058.18^{c-f}	1036.0^{c-g}	25.66^{o-q}	42.00 ^{j}	45.25^{g-j}	47.58^{e-i}

In each plant, data with the same letter are not significantly different (Duncan multiple range test p≤0.05)

Table 8. Mean comparison of the relative coefficient of carbon allocation to different organs of corn and bean plants under the influence of the double interaction of tillage systems and wheat residues

		Corn				Bean			
Tillage systems	**Wheat residues (%)**	**RP**	**RS**	**RR**	**RE**	**RP**	**RS**	**RR**	**RE**
Conventional tillage									
	0	0.384^{h}	0.343^{i}	0.0614^{i}	0.04^{i}	0.377^{h}	0.361^{e}	0.091^{h}	0.059^{f}
	30	0.416^{ef}	0.392^{fg}	0.0885^{f}	0.057^{g}	0.413^{fg}	0.372^{ed}	0.101^{ef}	0.066^{ed}
	60	0.421^{d-f}	0.395^{f}	0.088 ^{f}	0.06^{fg}	0.425^{ef}	0.376^{c-d}	0.103^{e}	0.067^{ed}
	90	0.438^{c-e}	0.407^{e}	0.092^{e}	0.06^{fg}	0.426^{ef}	0.377^{c-e}	0.105^{e}	0.067^{ed}
Minimum tillage									
	0	0.409^{gf}	0.386^{g}	0.072^{h}	0.0473^{h}	0.402^{g}	0.372^{ed}	0.095^{hg}	0.061^{f}
	30	0.443^{bc}	0.414^{de}	0.111^{c}	0.072^{e}	0.444^{cd}	0.377^{c-e}	0.118^{d}	0.077^{c}
	60	0.455^{ab}	0.432^{b}	0.112^{c}	0.0755^{d}	0.466^{ab}	0.38^{b-d}	0.132^{bc}	0.086^{ab}
	90	0.46^{a}	0.444^{a}	0.117^{b}	0.0828^{b}	0.475^{a}	0.389^{a-c}	0.128^{c}	0.083^{b}
No-tillage									
	0	0.398^{gh}	0.377^{h}	0.076^{g}	0.049^{h}	0.4^{g}	0.361^{e}	0.098^{fg}	0.063^{ef}
	30	0.435^{cd}	0.41^{e}	0.097^{d}	0.0631^{f}	0.434^{de}	0.394^{ab}	0.106^{e}	0.069^{d}
	60	0.444^{a-c}	0.424^{c}	0.116^{b}	0.0788^{c}	0.456^{bc}	0.403^{a}	0.145^{a}	0.09^{a}
	90	0.444^{a-c}	0.419^{cd}	0.122^{a}	0.086^{a}	0.451^{c}	0.405^{a}	0.136^{b}	0.087^{ab}

In each column, data with the same letter are not significantly different (Duncan multiple range test p≤0.05)

3.4. Relative coefficient of carbon allocation and allocated carbon content to different organs

The results showed that only the double interaction of the tillage system and wheat residues had a significant effect ($p \leq 0.05$) on the relative coefficient of carbon allocation and allocated carbon content to different organs of corn and bean, including seeds, straw, root, and extra-root. In each tillage system, increasing the percentage of wheat residues led to an increase in the relative coefficients of carbon allocation in both corn and bean plants (Table 8). The highest amount of relative coefficients of carbon allocation in different organs of both corn and bean plants was related to conservation tillage systems. In corn, the highest amount of RP (0.460) and RS (0.444) was related to the 90% wheat residues along with the minimum tillage system, and the maximum value of RR and RE was observed in the 90% wheat residues along with the no-tillage system (Table 8).

In bean, the highest amounts of RP (0.475) and RR (0.145) were related to the 90% wheat residues along with the minimum tillage system, and the 60% wheat residues along with the no-tillage system, respectively. The maximum value of RS and RE for bean was obtained in the 60% wheat residues along with the no-tillage system, which had no significant difference with the 90% wheat residues along with the no-tillage system (Table 8). The lowest number of relative coefficients of carbon allocation in different organs of both corn and bean plants was related to the conventional tillage system, along with 0% wheat residues. Also, the results showed that in both corn and bean plants, the contribution of root secretions from the total carbon in the plant was lower compared to other organs. Corn with a carbon allocation coefficient equal to 0.04-0.08 had the lowest amount of carbon allocated to root secretions (Table 8).

The interaction of tillage and plant residues on the allocated carbon content to different organs (CP, CS, CR, and CE) of corn and beans is given in Table 9. The results indicated that the highest amount of CP for corn (445.26 g Carbon m^{-2} year) was obtained in the minimum tillage system, along with 90% wheat residues. The highest value for beans (21.83 g Carbon m^{-2} year) was observed in the minimum tillage system along with 90% wheat residues, which was not significantly different from the no-tillage system along with 60% wheat residues (Table 9). The highest amount of CS for corn (452.78 g Carbon m^{-2} year) and bean (16.97 g Carbon m^{-2} year) was observed in the minimum tillage system along with 90% wheat residues, which was not significantly different from the minimum tillage system along with 60% wheat residues (Table 9). The use of plant residues and conservation tillage (minimum tillage and no-tillage) due to the improvement of soil physical and biological characteristics for root growth led to an increase in the CR. So that the highest amount of CR for corn (135.95 g Carbon m^{-2} year) and bean (4.57 g Carbon m^{-2} year) was obtained in the no-tillage system along with 90% wheat residues and the no-tillage system along with 60% wheat residues, respectively (Table 9). The highest amount of CE for corn (88.33 g Carbon m^{-2} year) and bean (2.97 g Carbon m^{-2} year) was observed in the no-tillage system along with 90% wheat residues and the no-tillage system along with 60% wheat residues, respectively (Table 9).

Table 9. Mean comparison of allocated carbon content (g Carbon m^{-2} year) to different organs of corn and bean plants under the influence of the double interaction of tillage systems and wheat residues

Tillage systems	Wheat residues (%)	Corn				Bean			
		CP	CS	CR	CE	CP	CS	CR	CE
Conventional tillage									
	0	382.89^{c}	304.95^{f}	59.^{25f}	39.45^{h}	11.49^{h}	11.42^{e}	3.81^{e}	2.47^{d}
	30	426.1^{ab}	387.72^{ed}	85.81^{d}	55.77^{f}	14 ^{f}	12.47^{d}	3.88^{e}	2.52^{d}
	60	427.56^{ab}	393.48$^{c\text{-}e}$	92.35^{cd}	60.03^{e}	15.89^{e}	14.03^{c}	3.98^{ed}	2.69^{c}
	90	429.07^{ab}	398.04^{cd}	95.03^{c}	62.58^{e}	16.46^{ed}	14.92^{bc}	3.89^{ed}	2.53^{d}
Minimum tillage									
	0	423.47^{ab}	379.53^{e}	68.72^{e}	46.67^{g}	12.68^{g}	11.83^{ed}	3.86^{e}	2.51^{d}
	30	431.18^{ab}	422.77^{b}	96.88^{c}	62.97^{e}	18.6^{c}	15.41^{b}	4.14^{cd}	2.69^{c}
	60	444.33^{a}	445.86^{a}	117.01^{b}	76.05^{cd}	19.94^{b}	16.91^{a}	4.4^{ab}	2.86^{ab}
	90	445.26^{a}	452.78^{a}	135.89^{a}	83.4^{b}	21.83^{a}	16.97^{a}	4.25^{bc}	2.76^{bc}
No-tillage									
	0	411.04^{b}	378.51^{e}	70.98^{e}	46.14^{g}	12.78^{g}	12.16^{ed}	3.83^{e}	2.49^{d}
	30	430.81^{ab}	406.33^{c}	113.41^{b}	73.72^{d}	17.11^{d}	14.92^{bc}	4.14^{cd}	2.69^{c}
	60	433.76^{a}	423.83^{b}	121.02^{b}	78.66^{c}	20.95^{a}	16.92^{a}	4.57^{a}	2.97^{a}
	90	444.14^{a}	437.21^{ab}	135.95^{a}	88.33^{a}	19.41^{bc}	15.41^{b}	4.4^{ab}	2.86^{ab}

In each column, data with the same letter are not significantly different (Duncan multiple range test $p \leq 0.05$)

4. Discussion

Conservation tillage leads to the reduction of soil disturbance and the preservation of more plant residues on the soil surface. This contributes to soil fertility, stability of aggregates, better water penetration, excellent regulation of temperature and pH, availability of nutrients, and accumulation of organic carbon. Thus, it is effective on the activity of soil microorganisms and affects plant growth (Cárceles Rodríguez et al., 2022; Williams et al., 2020). On the other hand, increasing the intensity of tillage increases the decomposition rate of organic matter and reduces the primary material required for the growth and activity of soil microorganisms. In this regard, the researchers reported that tillage operations have a significant impact on the activity of soil microbial communities, and the implementation of intensive tillage leads to a decrease in the growth and activity of the soil microbial population and soil microbial respiration in the long term (Baghel et al., 2018). Also, the researchers stated that the lowest rate of soil microbial respiration was observed in the conventional tillage and the highest rate was related to the conservation tillage (Nunes et al., 2018). The results of our study were in line with the results of the above studies.

It has been reported that in the no-tillage system, the microbial population and consequently the production of CO$_2$ due to the activity of the soil microbial population were much higher than the intensive tillage system (Wang et al., 2016). The main factor in increasing soil respiration is related to the availability of organic carbon in the soil and observed a significant and high positive correlation between organic carbon and soil respiration (Bera et al., 2018). Increasing soil organic carbon content as the primary material required for the growth of soil microbial communities improves the activities of soil microbial communities in the long term (Kumar, 2011). Preservation of plant residues by increasing organic matter leads to increased respiration in the soil. The high rate of soil respiration indicates the high quality of the soil and more microbial activity in the soil (Anjum and Khan, 2021; Cárceles Rodríguez et al., 2022). Plant residues can be used as a source of carbon and energy by the population of heterotrophic soil microorganisms and increase soil microbial respiration as an indicator of microbial activity (Anjum and Khan, 2021; Fu et al., 2021; Li et al., 2018). Other studies showed that the amount of carbon dioxide emission, which represents the respiratory activity of soil microorganisms and, as a result, the decomposition of organic matter added to the soil, increases significantly with the addition of plant residues to the soil (Lv et al., 2020; Siedt et al., 2023).

The increase in microbial respiration in intercropping can be due to the increase in the diversity of plants in multi-cropping systems and consequently the improvement of the conditions for the activity and growth of soil microbial communities through the root secretions of different species in intercropping and also the increment of some elements, especially nitrogen, through the selection of leguminous plants in intercropping (el Zahar Haichar et al., 2014). The researchers reported an increase in soil microbial respiration with an increment in the diversity of crops in soybean-millet intercropping (Aziz et al., 2013). Also, obtained similar results. They showed an increase in soil microbial respiration in wheat-groundnut intercropping compared to the sole cropping of both species (Lin et al., 2010). The increase of soil microbial activities in multi-cropping systems compared to sole cropping systems leads to the improvement of services such as recycling of nutrients and soil fertility, which was consistent with the findings obtained in this study. In this context, we showed that nitrogen and phosphorus contents were higher in bean-corn intercropping patterns, which had more microbial respiration, compared to their monocultures (Akbari et al., 2019).

According to the results, MBC content was higher in the no-tillage system compared to the minimum tillage system and also in the minimum tillage system compared to the conventional tillage system. These results can be due to the presence of plant residues on the soil surface and more carbon accumulation in the conservation tillage system. Following the increase in soil carbon, the microbial population has also increased, which leads to an increase in MBC (Bera et al., 2018; Cárceles Rodríguez et al., 2022). Similarly, the reported higher MBC under conservation agriculture practices compared to conventional tillage (Baghel et al., 2018).

The researchers declared that multi-cropping systems have a higher microbial population and biomass compared to pure species cultivation (Rivest et al., 2010). The main reason for the higher microbial biomass and population in intercropping systems is the greater availability of nutrients, especially phosphorus, for microorganisms (Scott et al., 2012). The study of soybean-sugarcane intercropping concluded that the soil microbial population in the intercropping increased significantly compared to the pure cultivation of both species (Li et al., 2013). These researchers found that the population of bacteria, fungi, and actinomycetes increased by 42.62%, 14.5%, and 78.5%, respectively, in soybean-sugarcane intercropping compared to pure sugarcane cultivation and by 188%, 183%, and 73% compared to pure soybean cultivation. Crop diversification can raise soil microbial diversity and activities because the roots of crops release exudates in intercropping systems, contributing to greater microbial biomass (el Zahar Haichar et al., 2014). Similarly to our results, an increment in MBC with intercropping compared with monoculture (Lopes and Fernandes, 2020). Decreased soil bulk density due to the use of organic inputs has been reported as one of the reasons for increasing microbial activity and microbial carbon in the soil because the decrease in bulk density increases root growth and its secretions and leads to the stimulation of soil microbial activity (Chowdhury et al., 2015; Fu et al., 2021). The herb residue vermicompost improved soil MBC, respiration, and some enzyme activities (Lv et al., 2020). Higher MBC values were found in residue treatments compared to the no-residue treatments (Chowdhury et al., 2015). It has been reported that the lower value of MBC in non-residue treatments resulted in a higher rate of microbial respiration per unit of MBC (i.e., a higher metabolic quotient) than in the treatments with residue, which could be related to the stressful conditions created by lower nutrient levels and carbon availability for microorganisms in the non-residue treatments (Chowdhury et al., 2015). Therefore, adding organic matter to the soil in conservation tillage systems increases MBC. The presence of organic matter and higher water-holding capacity in conservation tillage caused an increase in the carbon content of the microbial living mass compared to conventional tillage (Bescansa et al., 2006), which was consistent with the results of this study. The increasing trend of MBC in the second stage of sampling (100 days after planting) showed that wheat residues with a C:N ratio of 60.47 and a lignin content of 14% (Han and Rowell, 1997) were slow to decompose, and for this reason, microbial activities for decomposing polysaccharide compounds continue until the end of the growing season. The decomposition of soybean residues occurs very quickly in the early stages compared to wheat residues. They also stated that there is a slight difference in the decomposition process of corn and wheat residues, which is related to the difference in the level of soluble N substrate and the C:N ratio in these residues (Kelley et al., 2003). Also, temperature plays an important role in determining soil fungi population. Most fungi grow optimally at a temperature of 15-22 °C, and increasing the temperature too much has the opposite effect on them and may weaken the capacity of soil fungi to decompose organic matter and to facilitate plant nutrient absorption (Che et al., 2019). In the present study, the increase of MBC in the second stage of sampling (100 days after planting) showed that the temperature was favorable for the activity of fungi. The researchers reported that the fungi population decreases significantly in the summer months and increases with the onset of the first rain (Toberman et al., 2008). The result of these studies can be an emphasis on the possible effect of temperature on the change process of MBC in the present study.

Metabolic quotient is an important biological indicator that is used to determine the state of stress in soil ecosystems. Its value increases under stress conditions because soil microorganisms need more energy to maintain their living biomass, and their respiration rate increases. Also, under stress conditions, the MBC decreases faster than the soil organic carbon. High metabolic quotient values reflect CO_2 emissions higher than the maintenance requirement of microbial populations, which decreases the ability of soil to sequester carbon (Ashraf et al., 2022). The metabolic quotient value in wheat residues was significantly lower than the control, as reported by other researchers (Nava-Arsola et al., 2024; Paz-Ferreiro et al., 2012). Also, metabolic quotient values were higher in non-residue treatments than in residue treatments (Chowdhury et al., 2015). This indicates that adding organic matter reduced the microorganisms' stress by using the added carbon source to increase their microbial biomass and maintain their metabolic activity, promoting the microbial carbon accumulation (Nava-Arsola et al., 2024). There are different results about the effect of intercropping on metabolic quotient. For example, soybean–Aruana Guinea grass intercropping decreased metabolic quotient, while soybean–Congo grass intercropping increased metabolic quotient (Batista and Vilela, 2023). This difference in metabolic quotient can be due to environmental conditions, soil characteristics and the type of plants used in intercropping.

In line with our results, they reported that the metabolic quotient was always lower under the no-tillage system (Hu et al., 2013). Two hypotheses are proposed to justify the difference in the amount of metabolic quotient among the studied treatments. In the first hypothesis, the lower metabolic quotient indicates the lower level of stress in the microbial community in the no-tillage system and severe stress in the conventional tillage system. An increase in metabolic quotient, as a stress-sensitive index, has been reported in the soil ecosystem by salt accumulation, acidification, or changing environmental conditions (Ashraf et al., 2022; Hou et al., 2019). Different environmental conditions, such as the difference in the entry of fresh organic matter into the soil and the quality of organic carbon in the studied soil, can be the cause of the difference in the metabolic quotient amount. In the second hypothesis, a lower metabolic quotient can indicate a change in the microbial community. The management that causes the increase of plant residues on the soil surface stimulates the activity of fungi (Anderson, 2003; Fu et al., 2021). Fungi stabilize more decomposable carbon in their living mass than bacteria. Therefore, fungi have a better efficiency in converting carbon into their living mass and have a lower metabolic quotient (Chen et al., 2019; Six et al., 2006). It seems that more increase of organic matter on the soil surface in the no-tillage system caused more activity of fungi population and decreased metabolic quotient. Among soil microorganisms, the lowest metabolic quotient is related to fungi. On the other hand, bacteria, especially opportunistic bacteria, whose relative abundance in the soil increases dramatically due to tillage and soil disturbance and the use of accessible carbon from broken aggregate, have a high metabolic quotient and spend a significant part of the absorbed carbon in respiration. An increase in microbial activity per unit of living mass indicates higher levels of available organic carbon, but more use of it to obtain the necessary energy to maintain the existing state (catabolism) and less use of energy for growth (anabolism) (Islam and Weil, 2000). In many studies, the metabolic quotient is used as a suitable index to evaluate the response of soil microorganisms to ecosystem disturbances, cell physiological conditions, and environmental stresses, such as salinity, pollution, and drought (Ashraf et al., 2022). However, it is not possible to check this hypothesis with the present data, and it is necessary to measure the population of fungi and bacteria separately.

More biomass production of corn plants led to higher corn NPPc in pure corn cultivation compared to other cultivation patterns. Therefore, in intercropping systems where the proportion of corn plants was higher, the amount of corn NPPc was higher in them. By examining the carbon cycle in Iran's agricultural system, the results indicated that the amount of NPPc in corn is higher (Nasiri Mahalati et al., 2014). Increasing preservation of plant residues in soil has been reported as one of the important factors in increasing carbon sequestration potential and NPPc (Wang et al., 2010)

Decreased NPPc in the conventional tillage system was consistent with the results obtained by other researchers (Lu et al., 2018; Yeboah et al., 2016). The increment in soil nutrients related to conservation tillage resulted in a higher carbon and nitrogen accumulation in plant tissues, which could contribute to organic carbon when the straw is recycled. This was also reported by others (Yeboah et al., 2016).

In investigating the role of species diversity on the carbon cycle, it was reported that the increase in species diversity leads to an increase in carbon storage in the soil (Lange et al., 2015). Also reached similar results that the increase in biological diversity in farms leads to an increase in soil carbon storage (Smukler et al., 2010). The amount of carbon sequestration increased with the increase and preservation of biodiversity and estimated the value of carbon sequestration to be around 1.6 million dollars by 2050, while this amount in the common scenario without preserving biodiversity was estimated to be equal to 0.9 million dollars (Nelson et al., 2009). As previously mentioned, due to the high NPPc in corn, it seems that in systems where the corn plant is one of its components, the potential of entering carbon and thus carbon sequestration increases. The researchers declared that to increase the efficiency of carbon sequestration in agriculture, different cultivation systems in the form of crop rotation and intercropping should be designed in such a way that the potential of entering and sequestering carbon in the soil increases with the increase in biomass production (Wang et al., 2010).

According to the test results, it can be said that the design of cultivation systems should be done in such a way that, in addition to the presence of several plant species and, in other words, high plant diversity, its components have high NPPc. In this way, in addition to increasing the potential of carbon sequestration quantitatively, due to the variety of residues returned to the soil, it can also be improved qualitatively. Management actions aimed at increasing plant production can have a significant potential to improve or increase carbon storage in the soil (Cárceles Rodríguez et al., 2022; Mohammadi et al., 2017). Management practices can affect the storage of carbon or its loss in the soil through changes in the physical and chemical characteristics of the soil, root morphology, soil moisture, and the speed of microbial activities (Cárceles Rodríguez et al., 2022; Fu et al., 2021; Mondal et al., 2020).

Soil improvement by adding organic matter leads to an increase in the availability of nutrients and is a common method in cropping systems to increase NPPc (Blair et al., 2006; Mohammadi et al., 2017). Organic manure, plant residues, municipal waste, and sewage sludge are common organic matter forms. Adding organic matter to the soil directly increases soil carbon reservoirs and indirectly leads to an increase in carbon storage potential by stimulating plant growth. It has been reported that high soil microbial activities could accelerate soil nutrient cycling and contribute to the growth and production of plants (Lv et al., 2020). The soil respiration increased (18-19%) over three years following the addition of organic matter to the soil (Ryals and Silver, 2013). Therefore, management techniques, such as adding organic material compost, can lead to a potential increase in soil carbon resources in the long term. They also showed that the NPPc was higher in plots that received organic fertilizer compared to the control. Adding organic matter and other management operations can lead to an increase in soil carbon resources compared to chemical fertilizers, which only provide nutrients for a short time. This can probably have positive effects on increasing NPPc via increasing water holding capacity and nitrogen availability (Mohammadi et al., 2017). Adding organic matter to the soil causes the slow release of elements during decomposition, resulting in a long-term sustainable increase in NPPc (Blair et al., 2006). By adding organic and inorganic fertilizers to a pasture, they observed a similar pattern of increased organic carbon and microbial activity (Kowaljow et al., 2010). Also, it has been reported that the addition of compost on the soil surface led to an increase in the NPPc of aerial and underground organs during three growing seasons, which increased the annual carbon input from the vegetation (Ryals and Silver, 2013). Also reported is that the RE in corn plants is low and around 0.089. They attributed the reason for this to the physiological structure of this plant (Bolinder et al., 2007).

It seems that the reason for this is related to the lower contribution of the underground organs of corn plants, which has caused a decrease in the contribution of root exudates. The higher root exudation for beans can relate to its structure and characteristics, such as the ability to fix nitrogen. The increase in plant production and carbon allocation with the use of crop residues in the conservation tillage system can be due to the improvement of the organic carbon content and biological activity of the soil and the reduction of soil bulk density. The researchers stated that by organic fertilizer management, the RR and RE in the corn can be increased and preserved in the soil (Mohammadi et al., 2017).

It seems that the gradual release of nutrients from organic inputs has established a proper balance between the growth of vegetative and reproductive organs (Lambers et al., 2008; Mohammadi et al., 2017).

The use of intensive tillage operations has hindered the growth of roots due to the destruction of the soil structure and its compression, and as a result, the reduction in the allocation of photosynthetic assimilates for its growth. On the other hand, the use of organic inputs has improved the root growth and allocation of photosynthetic assimilates to the roots due to the improvement of soil characteristics (Abbott and Murphy, 2007) and the gradual availability of nutrients during the plant growing season (Fu et al., 2021; Mondal et al., 2020). The amount of CE is affected by the amount of CR (Bolinder et al., 2007). The researchers reported that using organic manure and reducing the use of tillage operations increased the amount of carbon allocated to root due to the improvement of the soil's physical and biological characteristics for root growth (Mohammadi et al., 2017).

5. Conclusion

Generally, wheat residue had the potential in combined with the no-tillage system and maize-bean intercropping to improve soil microbial activities and increase bean NPPc and carbon allocation. In no-tillage systems, the presence of wheat residues in the soil increased soil organic carbon, and the lack of destruction and manipulation in the soil improved the conditions for the development of the soil microbial community. The highest amount of microbial biomass carbon and respiration was obtained in the no-tillage system and 2 maize rows:2 bean rows along with the highest percentage of wheat residues (90% and 60%, respectively). Increasing organic matter on the soil surface in the no-tillage system caused more activity of the fungal population and decreased metabolic quotient. Such that the lowest amount of metabolic quotient was obtained in the no-tillage system along with 90% wheat residues and pure maize cultivation. The results showed that the application of wheat residues coupled with conservation tillage could improve soil microbial activity, and stimulate NPPc and carbon allocation, thus creating a synergic relationship. The highest relative coefficients of carbon allocation in different organs of maize and bean plants were related to conservation tillage systems with the highest amount of wheat residues (90% and 60%). The greatest value of NPPc for maize was obtained in the no-tillage system with 90% wheat residues and pure maize cultivation. Therefore, through the use of conservation tillage and wheat residues, it is feasible to enhance the NPPc amount and carbon allocation to various organs (particularly roots) in maize and bean plants. This also results in carbon retention in the soil and prevents its release into the atmosphere by adding the residues of these plants (especially maize). Hence, it is suggested to consider the improvement of soil organic matter through the use of plant residues along with the implementation of conservation tillage and intercropping as a sustainable and ecological strategy to improve soil biological activities and plant productivity. Also, these results would be the main step in sustainable agriculture wherein plant residues along with conservation tillage and intercropping could be applied for sustainable production of plants, improvement of soil characteristics and prevention of carbon emissions into the atmosphere.

Conflict of interest

There is no conflict of interest.

Consent for publications

All authors read and approved the final manuscript for publication.

Availability of data and material

All the data are embedded in the manuscript.

Authors' contributions

All authors contributed to the study conception and design. Material preparation, data collection and analysis were performed by F. Akbari, M. Dahmardeh, and A. Morshedi. The first draft of the manuscript was written by F. Akbari and M. Dahmardeh and all authors commented on previous versions of the manuscript. All authors read and approved the final manuscript.

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Ethics approval and consent to participate

No humans or animals were used in the present research. The authors have adhered to ethical standards, including avoiding plagiarism, data fabrication, and double publication.

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