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Research Article

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The effects of long-term application of organic and inorganic fertilizers on soil and plant Nitrogen-Carbon contents.

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ABSTRACT
Soil and crop management such as chemical fertilizer applications hurt the soil's biological quality and productivity. The most well-known organic fertilizer such as animal manure, compost and mycorrhizal fungi are significantly contributing to soil organic carbon sink and consequently, organic fertilizers have a positive effect on the soil biological diversity and productivity. To increase soil carbon content, long-term organic fertilizer applications increase soil carbon budget hypothesis was tested under field conditions. The aim is to investigate the effects of different organic and inorganic fertilizer applications on the soil and wheat plant carbon and nitrogen content. A long-term field experiment was established in 1996 until the present time. In 2018, before cultivation control, Mineral fertilizer (NPK), Animal manure (25 ton ha^{-1}), Compost (25 ton ha^{-1}) and Compost+Mycorrhiza (10 ton ha^{-1}) were applied. Adana-99 varieties of wheat seeds were sown. After harvest, soil and plant samples were taken and soil-plant carbon-nitrogen analyses were made. Results show that C % and N % contents of the plant seed, shoot, and root have higher concentrations than that of the control treatment. When the TC %, OC %, and N % contents of the soil at different depths (0-15 cm and 15-30 cm) and in the rhizosphere (R) and no rhizosphere (NR) area were examined, compost and animal manure treatments have higher content. At the rhizosphere area 0-15 cm depth the soil % OC, and % IC contents were statistically significant. The highest values of soil OC %, C: N and IC % contents at Rhizosphere 0-15 cm depth were obtained in animal manure with 2.94 %, 10.06, and 3.90 % respectively. Organic fertilizers application increased soil TC %, OC % and N% contents as well as contribute to the carbon and nitrogen budget. The results found support our hypothesis.
Keywords: Wheat, Carbon, Organic Carbon Nitrogen, organic mineral and fertilizer.

INTRODUCTION
The world population was 250 million in the 1000's, it increased to 6.1 billion in 2000 and it is expected to be 9.8 billion by 2050 (Kopittke et al., 2019). Depending on the population growth, an increase in food demand occurs, which causes more agricultural production per unit area. In this situation, human life mainly depends on sustainability (Aznar-Sanchez et al., 2019) and manageability of agricultural production. Due to the desire to obtain more product, it causes to use of excess chemical fertilizers and other inputs in to agricultural production systems. Mineral fertilizer applications to the soil reduce soil health and subsequently, there were decreases in organic carbon soil microbial carbon (TMC) due to reducing the microbial activities. It has been reported that excessive mineral nitrogen application reduces soil microbial diversity which may be due to the formation of an acidic environment in the soil as a result of high concentration of ammonium in soil (Zhang et al., 2015). Decreased microbial activity may be caused a serious risk for the sustainability of soils health. For a sustainable agriculture, it is necessary to manage the land use agro-ecologically. Soil carbon must be reintroduced to the soil for a healthier management of agriculture. In addition to organic matter, soil-plant management practices, including the use of organic and inorganic fertilizers, significantly affect the soil organic carbon pool and agronomic yield (Ortas and Lal, 2014). For the proper management of soils, the organic material taken from the soil must be returned to the soil.

Long-term organic fertilizers application to the soil instead of mineral fertilization will increase the microbial activity of the soils and also will allow the addition of organic carbon in the soils. Addition of organic matter to soil significantly affects soil carbon dynamics such as soil carbon pool (Ortas et al., 2013). Organic material sources can be listed as animal manure, composts, stubbles, urban wastes, biochar, leonardite, green manure, humic acids (Gezgin, 2018). In addition to these resources, mycorrhiza applications are also an extremely important applications for the atmospheric carbon fixation and microbial activity of the soil. Increasing the amount of organic carbon in soils is extremely important to increase plant growth and consequently mitigate atmospheric CO_2 concentration. Increased CO_2 emission to atmosphere caused climate changes. Batjes (1996) indicated that land use changes and predicted global warming, through climate effect on net primary productivity, the plant community and soil conditions, may have significant effects on the size of the soil organic carbon pool and directly affect the concentration of atmospheric greenhouse gases.

In this context, the aim of the study is that organic (including mycorrhizal inoculation) and inorganic fertilizers applied to the soil will increase the total carbon and organic carbon content of the soil and have a long-term effect on the soil carbon budget. The tested hypothesis was that organic fertilizer sources (including mycorrhizal inoculation) application to the soil increase the soil organic carbon and nitrogen concentration.

MATERIALS AND METHODS

The trial was established at Menzilat soil series (Typic Xerofluvents) in 1996 and continues to the present day. Trial located on the Research Farm of the Çukurova University in the eastern part of the Mediterranean region of Adana-Turkey. An image of the trial area is given in figure 1 and soil properties are given in Table 1.

The experiment was established in 1996 with composed of 5 treatments. The treatments were (i) control; (ii) traditional N-P-K fertilizers (160 kg N ha-1 83 kg K ha-1 as K_2SO_4, and 26 kg P ha-1 as $(NH_4)_2SO_4$, as 3Ca $(H_2PO_4)_2.H_2O$); (iii) compost at 25 Ton ha-1; (iv) animal manure at 25 Ton ha-1; and (v) mycorrhiza-inoculated compost at 10 Ton ha-1. Annually, the organic fertilizers (animal manure, compost, and mycorrhizae) were uniformly spread on the soil surface just before sowing and were incorporated into the surface 0-15 cm layer with a disc harrow. Similar tillage practices were followed for the control and fertilizer-treated plots.

Soil samples were taken from 2 different depths (0-15 cm and 15-30 cm) and 2 different areas ((Rhizosphere (R) and Non-rhizosphere (NR)) for each plot in May 2019. Plant samples were also taken on the same date for seed, shoot and root samples as well.

Soil samples were air dried, ground, and passed through a 2 mm sieve for soil analysis. Also, for determining total C and N concentrations soil was anymore ground and sieved through a 0.25-mm. Soil samples analyzed by the dry combustion method at 900-C using a C and N elemental analyzer (Fisher-2000). Inorganic C was determined by measuring the total $CaCO_3$ content using a calcimeter (schibler type) device.

The SOC concentration was analyzed by subtracting soil inorganic C from total C (Ortas et al., 2013; Ortas and Bykova, 2020). Total N concentration was measured by the Carbon nitrogen analyzer device.

Plant tissue total C and N concentrations were analyzed by the dry combustion method at 900-C using a C and N elemental analyzer (Fisher-2000) as well.

Obtained data Analyses were analyzed with JMP8 statistical software to assess the effects of different treatments and soil depths on soil properties. Treatment means were compared using the least significant difference test (P <0.05). Furthermore, the Principal component analyzes PCA analyzes were made by XLSTAT Statistical Software.

Table 1. Soil Characteristics of Menzilat Soil in 1996 (Ortas and Lal, 2014).

Parameters	Unit	0-15 cm Depth	15-30 cm Depth
Clay		318.8 ±30.6	333.4±21.8
Silt		360.9 ±87	379.5±13.4
Sand		320.3 ±23.0	287.2±16.4
Organic C Soil		0.96 ±0.08	0.78 ±0.08
İnorganic Carbon	g kg^{-1} soil	3.77 ±0.35	3.97 ±0.42
Total N		0.08 ±0.01	0.07 ±0.01
CEC	$Cmol^{+}$ kg^{-1}	20.50 ±2.00	17.90±1.64
pH	H_2O	7.58 ±0.66	7.60 ±0.71
Salt	%	0.05 ±0.00	0.04 ±0.00
P		22.60 ±2.16	20.20±2.00
Fe		5.43 ±0.82	5.66 ±0.58
Mn	mg kg^{-1}	5.74 ±0.32	5.31 ±0.59
Zn		0.52 ±0.05	0.23 ±0.02
Cu		1.86 ±0.19	1.56 ±0.16
AMF spores	10 g^{-1} soil	64.00 ±11.70	44.00±2.62

Mean of three replicates ±SD

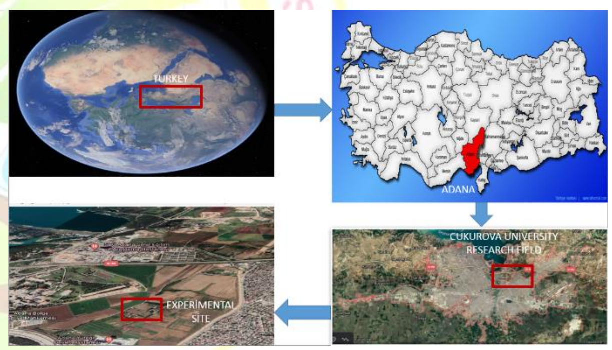


Figure 1. A View of the Trial Area

RESULTS AND DISCUSSION

Soil Organic C, Inorganic C, N, and C: N Ratio

Results of organic and inorganic C, N and C: N concentrations for different organic and inorganic treatments are presented in Table 2. İn rhizosphere soil depth 0-15 cm depth, different Organic and inorganic treatments had significantly impacted (P<0.05) on soil organic and inorganic carbon concentrations and C: N ratio.

In the 0, 0-15 cm rhizosphere soil depth, higher values for the soil organic carbon concentration and C: N were obtained in animal manure compared to the control treatment. With different fertilization applications, the values obtained for the inorganic carbon were found to be higher in the control application compared to the animal manure-treated plots.

While the highest value for organic carbon concentration at R0-15 cm depth was 2.94 % in animal manure applied plot soils, the lowest value was 1.18 % in control plot soils. Similarly, in terms of C: N, the highest value at R0-15 cm depth was found to be 10.06 in animal manure, while the lowest value was 6.05 in control application. The highest value for inorganic carbon at a depth of R0-15 cm was 3.90% in the control application, while the lowest value was measured in animal manure applied to soil as a 3.57 %.

When the effects of different organic and inorganic fertilizer applications on the nitrogen concentration of the soil were examined, the highest values were obtained in animal manure at 2 different soil depths of rhizosphere and non-rhizosphere zone, but the lowest values were obtained in the control plot soils. The highest N concentration was found as 0.29 % in animal manure at R0-15 cm depth, the lowest value was found as 0.13 % at 15-30 cm depth in control plot soils.

Table 2. Effect of Inorganic and Organic Fertilizers on IC %, OC %, N %, and C:N of the Soil 2019

APPLİCATİONS	AREA	DEEP	IC %		OC %		N %		C: N	
Control	R	0-15	3,90	±0,06A	1,18	±0,41B	0,19	±0,02	6,03	±1,57B
		15-30	3,74	±0,08	1,14	±0,74	0,18	±0,01	6,28	±4,02
	NR	0-15	3,57	±0,08	0,61	±0,91	0,16	±0,04	3,17	±4,51
		15-30	3,69	±0,07	1,34	±0,35	0,13	±0,08	12,45	±6,84
Mineral Fertilizer	R	0-15	3,73	±0,10BC	2,04	±0,48AB	0,22	±0,03	9,10	±1,03A
		15-30	3,76	±0,26	1,04	±0,68	0,19	±0,01	5,42	±3,37
	NR	0-15	3,60	±0,06	1,25	±1,25	0,21	±0,10	5,08	±2,71
		15-30	3,52	±0,10	1,33	±0,36	0,18	±0,03	7,77	±3,66
Animal manure	R	0-15	3,57	±0,06C	2,94	±0,52A	0,29	±0,02	10,06	±1,44A
		15-30	3,54	±0,05	1,63	±0,40	0,24	±0,04	6,90	±1,93
	NR	0-15	3,55	±0,14	1,39	±0,48	0,22	±0,05	6,35	±1,75
		15-30	3,64	±0,05	1,45	±0,43	0,27	±0,05	5,82	±2,97
Compost	R	0-15	3,77	±0,16AB	1,84	±1,06AB	0,28	±0,08	6,10	±2,46B
		15-30	3,78	±0,13	1,28	±0,74	0,21	±0,03	5,84	±2,66
	NR	0-15	3,69	±0,12	1,13	±0,12	0,21	±0,03	5,42	±0,74
		15-30	3,63	±0,15	1,82	±0,48	0,19	±0,04	9,89	±3,52
Compost+ Mycorrhiza	R	0-15	3,69	±0,01BC	1,60	±0,39B	0,22	±0,03	7,31	±0,86AB
		15-30	3,68	±0,08	1,51	±0,50	0,19	±0,04	7,86	±1,24
	NR	0-15	3,80	±0,14	1,23	±0,63	0,20	±0,09	6,01	±1,98
		15-30	3,66	±0,13	1,12	±0,11	0,17	±0,06	7,27	±3,60

A, B, C: The difference between the averages indicate by different letters is significant in its column. Mean of three replicates ±SD. R: Rhizosphere NR: non-rhizosphere

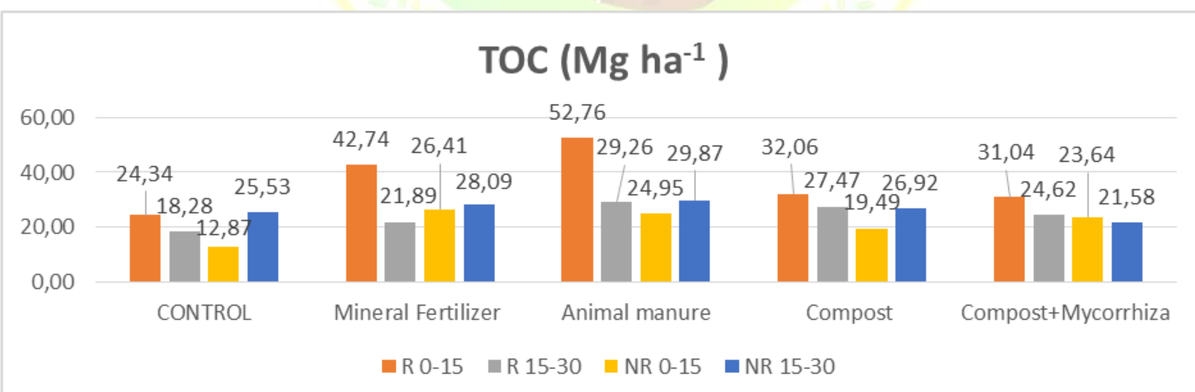


Figure. 2. Effect of inorganic and organic fertilizers in soil total organic carbon 2019.

Soil Total Organic Carbon and Total Nitrogen

Total soil organic carbon and nitrogen content increased in all organic fertilizer applied plot soils compared to the control. The highest values were generally measured in animal manure. when the total organic carbon content is examined. The highest value was found in the animal manure as 52.76 Ton ha^{-1} at R0-15 cm soil depth, while the lowest value was found as 12.87 Ton ha^{-1} at NR0-15 cm depth. For total nitrogen amount, the highest value was found in animal manure as 5.23 Ton ha^{-1} at R0-15 cm depth, while the lowest value was 6.64 Ton ha^{-1} at NR15-30 cm depth (Figure 2 and Figure 3).

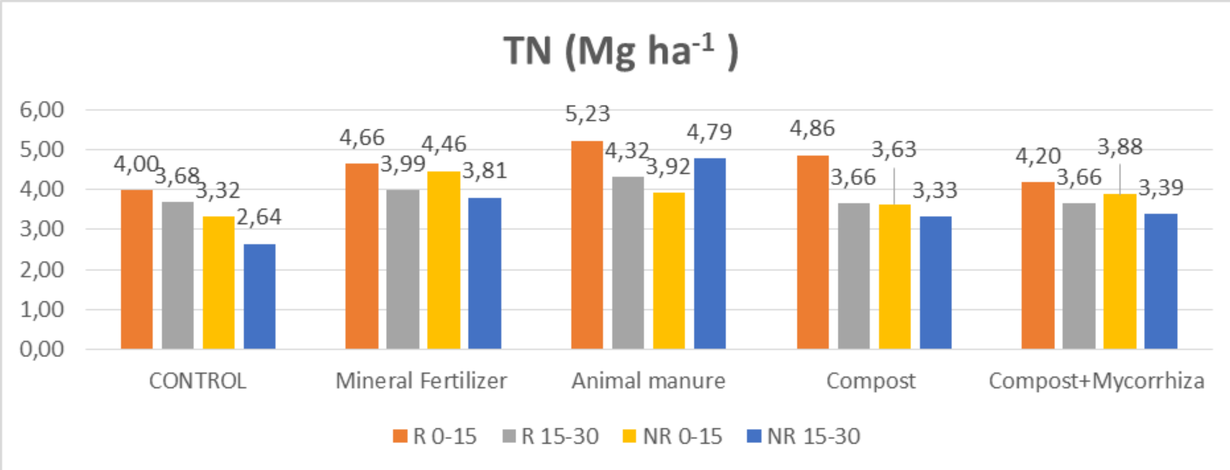


Figure. 3. Effect of inorganic and organic fertilizers on soil total nitrogen 2019

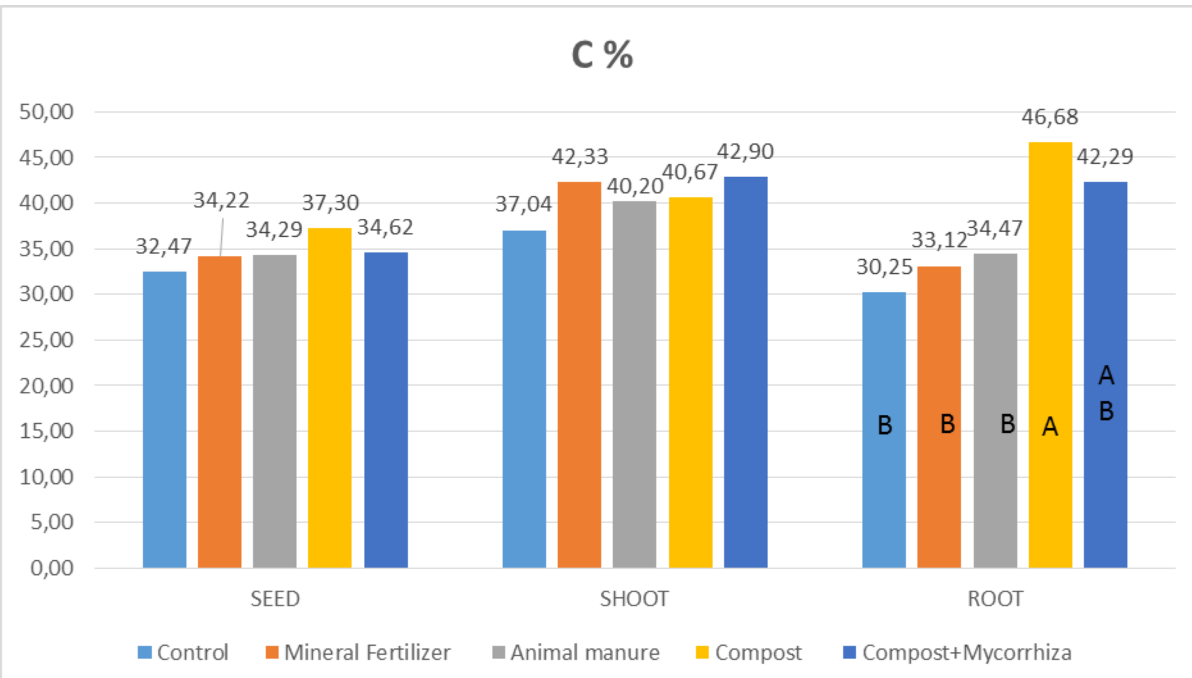


Figure 4. Effect of inorganic and organic fertilizers on plant carbon concentration 2019

Plant (Seed, Shoot and Root) C % and N % Concentration

After harvest plant tissue (seed, shoot, and root) % C and % N concentration were examined. It is seen that all applications, especially organic fertilization applications were higher than the control treatments. The highest C % values were generally obtained in compost and compost + mycorrhiza applied soils. In terms of plant C % the highest values for seed, shoot and root were found to be 37.30% in compost, 42.9% in animal manure and 46.68% in compost respectively. Similarly, in terms of nitrogen, the highest values for seed, shoot and root part were found as 2.25% in mineral, 1.5% in compost+mycorrhiza, and 2.39% in compost treatments respectively.

All measured results were statistically analyzed to find a relationship in between measured parameter related with fertilizer. The principal component analysis (PCA) analysis technique was applied. The results of different organic fertilizer applications are shown that 47.86 percentages of variation were fine on the X axis and 22.63 % of the variation was find on the y-axis. It is seen that plant-related parameters mainly are related in compost and Compost+Mycorrhiza applied plots. Animal manure and mineral fertilization were found to be close to each other on many measured parameters (Figure 6). The control application is far from all applications and is located on the negative part of the XY axis. While there was no effect of control application on the results obtained in the experiment with these results, it was seen that organic fertilizer applications were related to many measured parameters (Figure 6).

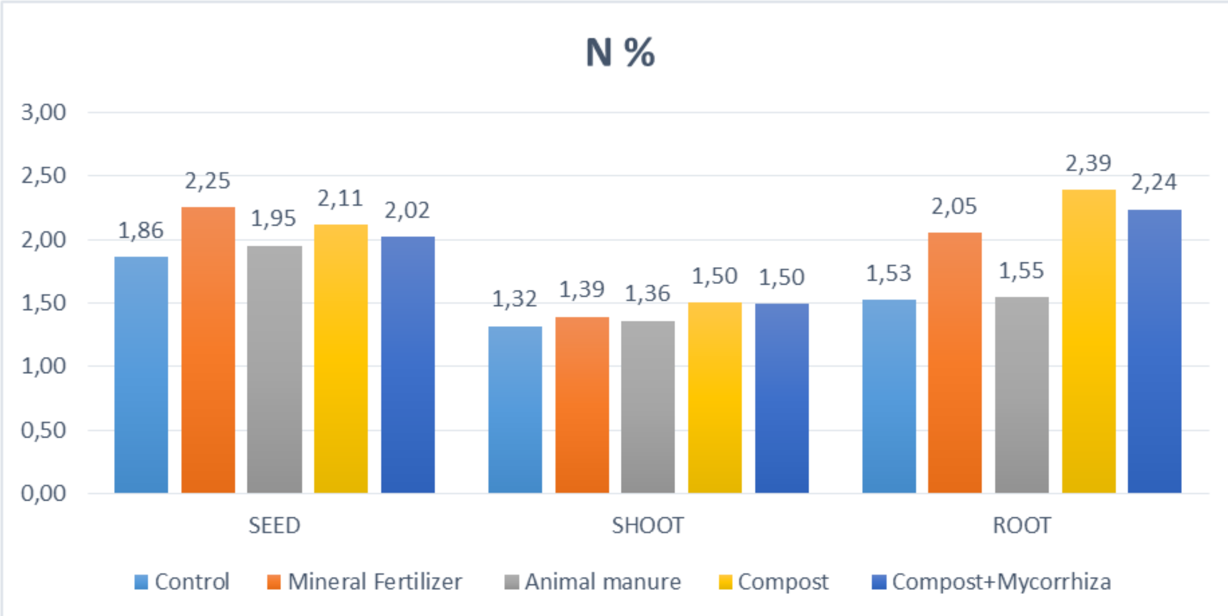
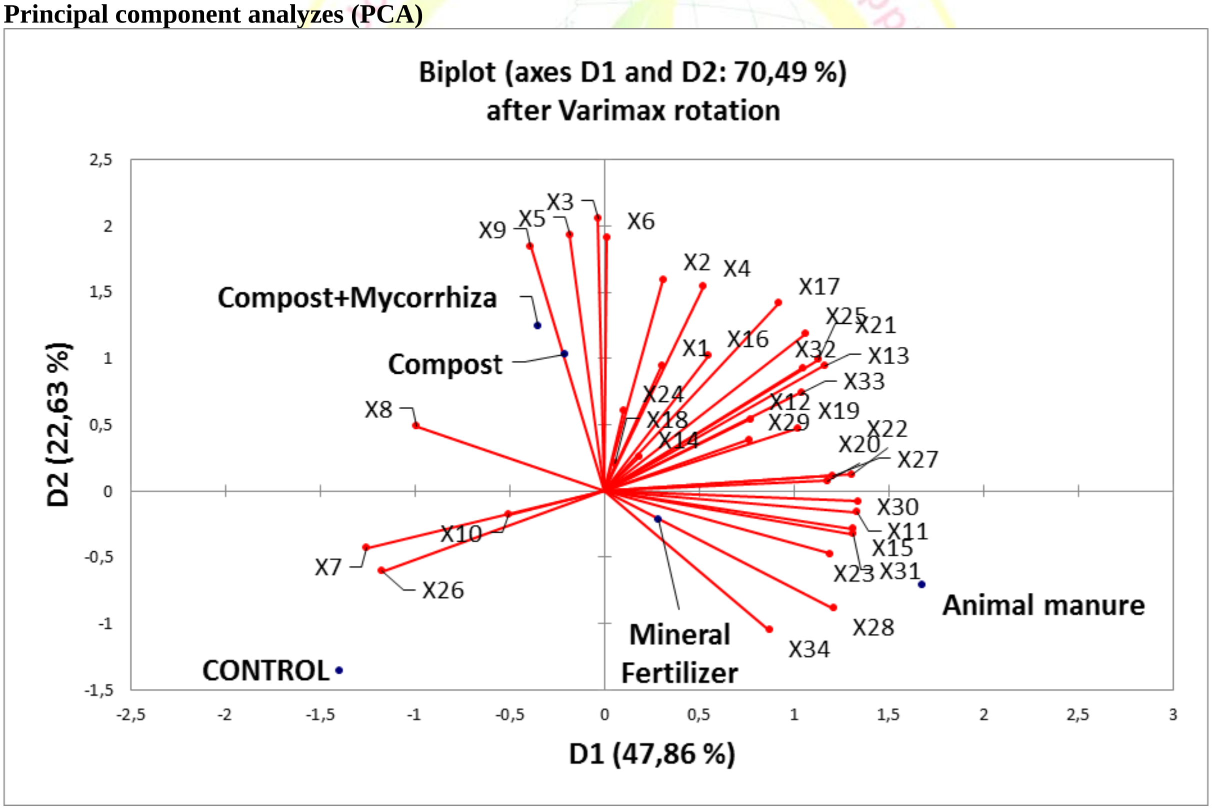


Figure 5. Effect of inorganic and organic fertilizers on plant nitrogen concentration 2019

Principal component analyzes (PCA)



X1 Seed N %,X2 Seed C % X3 Shoot N %, X4 Shoot C %, X5 Root N %, X6 Root C %, X7 R 0-15 IC %, X8 R 15-30 IC %, X9 NR 0-15 IC %, X10 NR 15-30 IC %, X11 R 0-15 OC %, X12 R 15-30 OC %, X13 NR 0-15 OC %, X14 NR 15-30 OC %, X15 R 0-15 TC %, X16 R 15-30 TC %, X17 NR 0-15 TC %, X18 NR 15-30 TC %, X19 R 0-15 N %, X20 R 15-30 N %, X21 NR 0-15 N %, X22 NR 15-30 N %,X23 R 0-15 C:N, X24 R 15-30 C:N, X25 NR 0-15 C:N, X26 NR 15-30 C:N, X27 R 0-15 TN (Ton ha-1), X28 R 15-30 TN (Ton ha-1), X29 NR 0-15 TN (Ton ha-1), X30 NR 15-30 TN (Ton ha-1), X31 R 0-15 TOC (Ton ha-1), X32 R 15-30 TOC (Ton ha-1), X33 NR 0-15 TOC (Ton ha-1), X34 NR 15-30 TOC (Ton ha-1).

Figure 6. Effect of several inorganic and organic fertilizers on Principal component analyzes (PCA) 2019

Organic and inorganic fertilizer treatments compared to control treatments had significantly impacted on soil organic carbon and carbon-nitrogen ratio (C: N) concentrations, which in R0-15 cm soil depth. While higher values for the organic carbon and C: N were obtained in animal manure compared to the control in the 0-15 cm rhizosphere soil depth, When the effects of different organic and inorganic fertilizer applications on the nitrogen concentration of the soil were examined, the highest values were obtained in animal manure at 2 different depths and in 2 different areas, but the lowest values were obtained in the control application. Ortas et al. (2013) reported that animal manure, compost, and compost+mycorrhiza applications applied to the soil increased the OC, N %, and C: N parameters compared to the control. In general mean of soil C: N ratios of soil organic matter range from 9.9 for arid soil to 25.8 for Histosols soil (Batjes, 1996). Results of animal and mineral fertilizer 0-15 cm rhizosphere soil are in parallel with the suggested C:N ratio by (Batjes, 1996; Van Groenigen et al., 2017). Abak and Sakin (2018) report that to balance or increase the C: N ratios of soils in the upper horizon (0-25) ~ 6:1-10:1 C: N ratios, it is important to mix post-harvest wastes with the soil and use organic fertilizers such as animal manure.

As can be seen in table 2, C: N ratio is in between 6 to 10. Since we determined total C and N concentration by CN analyzer compared to soil organic carbon analyses by Walkley-Black methodology our C: N ratio results are seeming to be lower than the suggested ratio.

Total soil organic carbon and nitrogen content increased in all organic applications compared to the control treatments. It has been reported by Malhi et al. (2006) that the amount of TOC and nitrogen is lower in the areas where stubble is not applied compared to the areas where stubble is applied. In addition, it has been reported that green manure supplementation improves the level of organic matter in the soil and increases the amount of N content (McVay et al., 2006). The application of organic fertilizers to the soil increases the organic carbon and total nitrogen content of the soil (Bokhtiar and Sakurai, 2005).

When the C % and N % contents of the plant (seed, shoot, and root) were examined, it was seen that all applications, especially organic fertilization applications had higher concentrations than the control application.

Previously Özkan et al. (2013) reported that different organic and inorganic fertilizers applied in greenhouse pepper cultivation have been reported to increase the N % of pepper compared to control treatment. Also for spring barley in 2008 and spring wheat in 2010, root C was higher in organic than in inorganic fertilizer-based systems reported (Chirinda et al., 2012). In general, compost and compost + mycorrhiza added plot soil have higher C concentrations. Also, Animal manure, compost, and com+myorriza applied plant C and N concentrations are higher than control treatments.

CONCLUSION

It was determined by the data that the application of long-term organic fertilizers increased the TC, TOC, TN concentration of the soil and the total carbon-nitrogen pool more than the control treatment.

The results obtained by processing the data are compatible with our hypothesis and support the following results.

1. The application of organic fertilizers had a positive effect on 0-15 cm rhizosphere soil TC, OC, N concentrations, and C: N ratio than the control treatment.
2. As a result of the application of organic fertilizers, the amount of TOC and TN increased more than the control.
3. Organic fertilizers had a greater effect on the plant (seed, shoot, and root) tissue carbon and nitrogen concentrations.

Applications for food safety and sustainable agriculture management should be implemented by taking into account climate change. Organic fertilizers should be used in agriculture to increase the carbon and nitrogen budget of the soil and to provide sufficient food security.

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