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## Effect of different organic wastes on biological properties of maize (*Zea Mays Indendata*) rhizosphere

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### Abstract

This study was carried in order to determine the effects different various organic wastes (tobacco production waste, wheat straw, tea waste and hazelnut husk) under greenhouse conditions on biological properties (microbial biomass C, basal soil respiration, dehydrogenase activity, urease activity and arylsulphatase activity) in clay-loam soil and rhizosphere (*Zea mays indandata*) soil of maize plant. The organic wastes were thoroughly mixed with the soil at a rate equivalent to 50 g kg<sup>-1</sup> on air-dried weight basis. Experimental desing was randomized plot desing with there replications in greenhouse. The moisture content in soil was maintained around 60 % of maximum water holding capacity by weighing the pots everyday. Changes in the biological properties were determined in the soil and rhizosphere (*Zea mays indendata*) samples and root free soil taken in 15, 30, 45, 60, 75 and 90 days after the experiment was conducted. At the end of experiment, all organic waste added soil increased biological properties of soil in comparison with the control (P<0.01) at all experimental periods. Moreover, biological properties in rhizosphere soil were higher than in root free soil at all organic waste application (P<0.01). Increased of organic wastes on soil biological properties had different trend (P<0.01), the most increases in the biological properties in the soil treated with tea wastes and tobacco production waste with supplying of low initial C/N ratio compared to other organic wastes.

**Keywords:** Organic waste, soil, rhizosphere, microbial biomass, basal soil respiration, enzyme activities.

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### Introduction

 The loss of soil organic matter under intensive land use is one of the many factors that degree agricultural soil of Anatolia. Traditional agricultural practices also leads to decrease fertility and, therefore, to declining productivity (Kenenbayev and Kucherov, 1994; Kızılkaya 2004). Soil organic matter is extremely heterogenous ranging from only slightly decomposed plant and microbial residues to highly humified organic substances. The most common practice to preserve and/or restore soil fertility is to add organic matter, which, preferentially, should be sufficiently stabilized to produce beneficial effects (Gallardo-Lara and Nogales, 1987; Mathur et al., 1993). Therefore, different types of organic wastes have increasingly been applied to soils in recent years. Organic wastes applications haven't only increased the soil organic matter, but have also enhanced the soil's C and N contents, and have improved biological activity in soil (Vigil et al., 1991). Plants influence C turnover and organic matter content in soils, both because they provide C inputs for microbiological characteristics in the soil through litter and exudation in the rhizosphere, and because they stimulate the turnover of existing soil C by rhizosphere microorganisms and their activities (Chen et al.,

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2006). The functional capacity of the soil microbial community, as reflected in the activities of enzymes involved in nutrient mineralization processes, varies among soils dominated by plant roots (Waldrop et al., 2000; Kourtev et al., 2003). Nevertheless, there have been relatively few studies that have examined root exudation, microbial rhizosphere community composition and enzyme activities of plants (Kourtev et al., 2003). Recently, Grierson and Adams (2000) indicate that microbial activity and enzyme activities are strongly affected by plant roots.

Microbial activity plays an important role in regulating soil fertility. Indeed, the microbiological processes taking place in soil are at the centre of many ecological functions (Nannipieri et al., 1990), since microbiological activity is related to soil structure, soil fertility, and the transformation of soil organic matter (Ladd et al., 1996). Several microbiological parameters have been used to define the status and sustainable development of soil productivity in agricultural ecosystems (Visser and Parkinson, 1992). There are many methods currently available for studying the microorganisms and their activities at the microhabitat level (Nannipieri et al., 1990). The dependence of the microbiological properties of agricultural soils on site and soil factors has been studied (Vekemans et al., 1989). Some soil biological properties such as enzyme activities, respiratory activity and microbial biomass are used as bio-indicators for soil quality and health in environmental soil monitoring (Rogers and Li, 1985).

The microbial biomass, being containing only 1-3% of total soil carbon and approximately 5% total soil (Smith and Paul, 1990), is an important component of soil organic matter. It is involved in biogeochemical cycles of the main nutritive elements (C-N-P-S) and in related energy flows (Meli et al., 2002; Kızılkaya et al., 2004). Basal soil respiration of soil microflora provides useful information on the physiological condition of the pedoecosystem, even though it is a matter some controversy. This respiratory activity takes into account the use of energy by microflora and expresses the efficiency of organic carbon degradation by soil microorganisms (Wardle and Ghani, 1992). As presence of dehydrogenases, which are intracellular to the microbial biomass, is common throughout microbial species and they are rapidly degraded following the cell death, the measurement of microbial dehydrogenase activity (DHA) in soils and sediments has been used extensively, (Bolton et al., 1985; Rossel and Tarradellas, 1991). Therefore, usage of DHA as an index of microbial activity has been suggested (Benfield et al., 1977; Nannipieri et al., 1990; Tabatabai 1994; Kızılkaya and Hepşen, 2004). Urease (UA) is involved in the hydrolysis of urea to carbondioxide and ammonia, which can be assimilated by microbes and plants. It acts on carbon-nitrogen (C-N) bonds other than the peptide linkage (Bremner and Mulvaney, 1978; Kızılkaya and Bayraklı, 2005). Arylsulphatase (ASA) is the enzyme involved in the hydrolysis of arylsulphate esters by fission of the oxygen-sulphur (O-S) bond. This enzyme is believed to be involved in the mineralisation of ester sulphate in soils (Tabatabai, 1994). Also, it may be an indirect indicator of fungi as only fungi (not bacteria) contain ester sulphate, the substrate of arylsulphatase (Bandick and Dick, 1999; Aşkın and Kızılkaya, 2006; Yertayeva et al., 2019).

The experiment in the present study was conducted in the greenhouse, simulating field conditions of organic matter management with different organic wastes (hazelnut husk, wheat straw, tea waste and tobacco production waste) in soil. The organic wastes used in the research were selected due to their variance in very large interval (C/N; 20 - 171). All organic wastes were sifted from 0.5 mm sieve after grinding in order to eliminate any effect that could be occurred due to magnitude of the particles. Our objectives were to determine the effects of the organic wastes on biological properties such as microbial biomass, basal soil respiration and enzymatic activities (dehydrogenase, urease and arylsulphatase) in rhizosphere and root-free soil.

## Material and Methods

### Material

#### Soil and organic wastes

Surface soil (0-20 cm) used in this experiment is a Typic Udipsamment and contained 20.60 % clay, 18.36 % silt, and 61.04 % sand. Soil texture can accordingly be classified as sandy clay loam (SCL). The pH in water was 8.1, the oxidizable organic matter content was 1.68 %, and the soil C:N ratio was 13.9. The properties of the organic wastes (Wheat straw (WS), Hazelnut husk (HH), Tea (TEW) production waste and Tobacco (TOW) production waste) was expressed on a moist-free basis and analyzed by standard procedures, given in Ryan et al. (2001).

#### Experimental procedure

The soil samples were air-dried in a laboratory and sieved through 0-2 mm screens. The samples (500 g air-dried soil) were placed in 600 ml cylindrical plastic container. The organic wastes (WS, HH, TOW and TEW)

were thoroughly mixed with the soil at a rate equivalent to 5% on an air-dried weight basis. Then, five individuals of maize (*Zea mays indendata*) seeds were placed in the soils. The moisture contents in the soils were adjusted to 60% water holding capacity (WHC) and the containers were incubated in greenhouse for 90 days. The moisture content was maintained throughout the experiment. The maize-planting containers were regarded as rhizosphere and the other containers as root free soil (nonrhizosphere). Changes in the microbiological properties were determined in the root free soil and rhizosphere samples taken in 15, 30, 45, 60, 75 and 90 days after the experiment was conducted. During the sampling of soil the crops were gently pulled out, and the soil remaining on the maize roots was regarded as rhizosphere. At the same time, the root free soil was taken from the nonplanting containers at the same depth. Soil without organic waste addition was used as a control. A randomized complete plot design with three replicates per treatment and soil was used. This greenhouse experiment was total 180 pots. The experiment was performed with the following 10 treatment:

- (1) control for soil (without organic waste addition and plant seed)
- (2) + 50 g kg<sup>-1</sup> hazelnut husk (without plant seed)
- (3) + 50 g kg<sup>-1</sup> wheat straw (without plant seed)
- (4) + 50 g kg<sup>-1</sup> tobacco production waste (without plant seed)
- (5) + 50 g kg<sup>-1</sup> tea production waste (without plant seed)
- (6) control for rhizosphere (without organic waste addition and with plant seed)
- (7) + 50 g kg<sup>-1</sup> hazelnut husk (with plant seed)
- (8) + 50 g kg<sup>-1</sup> wheat straw (with plant seed)
- (9) + 50 g kg<sup>-1</sup> tobacco production waste (with plant seed)
- (10) + 50 g kg<sup>-1</sup> tea production waste (with plant seed)

## Methods

### Total organic C and N contents

Total N in soil was determined by digestion and subsequent measurement by the Kjeldahl method (Bremner, 1965). Whole soil samples were sieved through a 150 µm mesh to determine total organic carbon by the wet oxidation method (Walkley-Black) with K<sub>2</sub>Cr<sub>2</sub>O<sub>7</sub>. C/N ratios in soils were calculated as total organic carbon / total nitrogen (Rowell, 1996).

### Biological properties in soils and rhizosphere

All determinations of microbiological properties were performed for the eachsoil sample in triplicate, and all values reported are averages of the three determinations expressed on an oven-dried sample basis at 105 °C for 48 h.

### Microbial biomass carbon and basal soil respiration

Microbial biomass carbon (C<sub>mic</sub>) was determined by the substrate-induced respiration method of by Anderson and Domsch (1978). A moist sample equivalent to 10 g oven-dry soil or cats was amended with a powder mixture containing 40 mg glucose. The CO<sub>2</sub> production rate was measured hourly using the method described by Anderson (1982). The pattern of respiratory response was recorded for 4 h. C<sub>mic</sub> was calculated from the maximum initial respiratory response in terms of mg C g<sup>-1</sup> soil as 40.04 mg CO<sub>2</sub> g<sup>-1</sup> + 3,75. Data are expressed as mg C g<sup>-1</sup> dry sample.

Basal soil respiration (BSR) at field capacity (CO<sub>2</sub> production at 22 °C without addition of glucose) was measured, as reported by Anderson (1982); by alkali (Ba(OH)<sub>2</sub>·8H<sub>2</sub>O + BaCl<sub>2</sub>) absorption of the CO<sub>2</sub> produced during the 24h incubation period, followed by titration of the residual OH<sup>-</sup> with standardized hydrochloric acid, after adding three drops of phenolphthalein as an indicator. Data are expressed as µg CO<sub>2</sub>-C g<sup>-1</sup> dry sample.

### Enzyme activities

Dehydrogenase activity (DHA) was determined according to Pepper et al (1995). To 6 g of sample 30 mg glucose, 1 ml of 3% TTC (2,3,5-triphenyltetrazoliumchlorid) solution and 2.5 ml pure water were added and the samples were incubated for 24 h at 37°C. The formation of TPF (1,3,5 triphenylformazan) was determined spectrophotometrically at 485 nm and results were expressed as µg TPF g<sup>-1</sup> dry sample.

Urease activity (UA) was measured by the method of Hoffmann and Teicher (1961). 0.25 ml toluene, 0.75 ml citrate buffer (pH, 6.7) and 1 ml of urea substrate solution were added to the 1 g sample and the samples

were incubated. The formation of ammonium was determined spectrophotometrically at 578 nm and results were expressed as  $\mu\text{g N g}^{-1}$  dry sample.

Arylsulphatase activity (ASA) was measured according to [Tabatabai and Bremner \(1970\)](#). 0.25 ml toluene, 4 ml acetate buffer (pH, 5.5) and 1 ml of 0.115 M *p*-nitrophenyl sulphate (potassium salt) solution were added to the 1 g sample and the samples were incubated for 1 h at 37°C. The formation of *p*-nitrophenol (*p*-NP) was determined spectrophotometrically 410 nm and results were expressed as  $\mu\text{g p-NP g}^{-1}$  dry sample.

### Statistical Analysis

All data were analyzed using SPSS 11.0 statistical software (SPSS Inc.). Analysis of variance (ANOVA) was carried out using three factors (plant root, incubation period, organic waste) randomized complete plot design; where significant *F*-values were obtained, differences between individual means were tested using the LSD (Least Significant Difference) test, with a significance level of  $P < 0.01$ . The asterisks, \*, \*\* and \*\*\* indicate significant at  $P < 0.05$ , 0.01 and 0.001 respectively.

## Results

### Composition of organic wastes

Among the OW used in this study, TEW had the highest organic matter (92.72%) while that of TOW was the lowest (66.21%). Regarding N content, TEW again had the highest N content (2.46%) and the lowest N content belong to WS (0.31%). C:N ratio of the OW ranged from 20 to 171 and the highest level C:N ratio observed in WS while that of lowest is TOW. The order of OW associated with C:N ratio was  $\text{WS} > \text{HH} > \text{TEW} > \text{TOW}$ . In addition these OW contained major important nutrients such as  $\text{P}_2\text{O}_5$ ,  $\text{K}_2\text{O}$ , which are agronomically important (Table 1).

Table 1. Composition of organic wastes in measured variables

Organic waste	Organic matter, %	C/N	N (%)	$\text{P}_2\text{O}_5$ (%)	$\text{K}_2\text{O}$ (%)
TEW	92,72	22	2.46	0.48	5.83
TOW	66,21	20	1.97	0.45	4.71
HH	85,34	52	0.96	0.28	5.17
WS	91,17	171	0.31	0.25	4.77

### Nutrients and biological properties in rhizosphere and root free soil

The organic carbon and N contents in rhizosphere and root free soils were significantly greater in all organic waste treatments compared to the control soil ( $P < 0.001$ ). All of the organic waste additions significantly increased total organic C in rhizosphere compared to the root free soil (Figure 1 and 2).

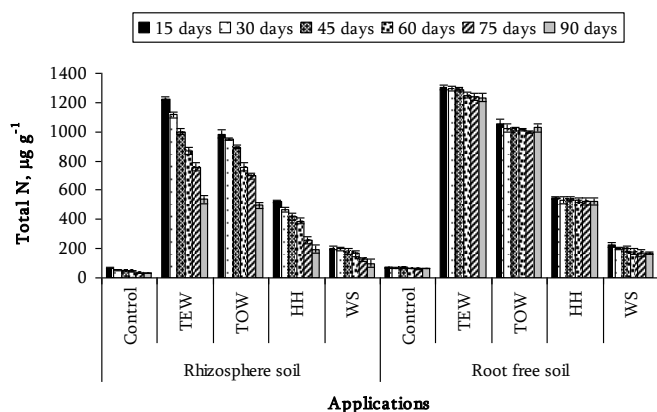


Figure 1. Changes of total nitrogen in rhizosphere and root free soil. Vertical bars are standard errors.

HH = Hazelnut husk, WS = Wheat straw, TOW = Tobacco production waste, TEW = Tea production waste

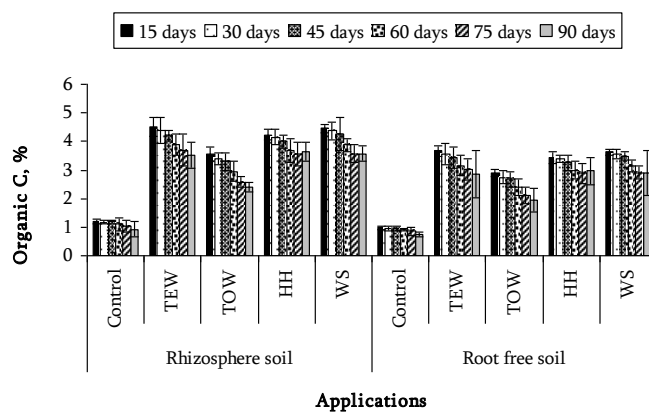


Figure 2. Changes of total organic carbon in rhizosphere and root free soil. Vertical bars are standard errors.

HH = Hazelnut husk, WS = Wheat straw, TOW = Tobacco production waste, TEW = Tea production waste

The effects of different organic waste treatments on biological properties in rhizosphere and root free soils are presented in Table 2-6. Considerable variations in all biological properties, BSR, Cmic, DHA, UA and ASA were found for the different organic wastes and with/without plant roots at different sampling times. Statistically significant variations were found in all biological properties at various organic waste application and sampling times (Table 7 and 8). Biological properties were also affected by incubation period, organic waste and plant root. The analysis of variance of the results obtained in our experiment on the periodic



sampling times with organic waste showed that all factors (organic waste types, plant roots and incubation periods) significantly influenced all biological properties. After organic waste addition a rapid and significant increase in biological properties was observed in waste amended soils followed by a progressive increase in the biological properties in rhizosphere amended with the organic waste. At the end of the experiment, the biological properties measured in waste-treated soils were statistically different from those measured in the control soils.

Table 2. Microbial biomass C (Cmic) in rhizosphere and root free soils ( $\mu\text{g CO}_2\text{-C g}^{-1}$  dry soil). Standard error in parenthesis.

Incubation Days	Organic wastes									
	Control		TEW		TOW		HH		WS	
	Rhizosphere soil									
15 days	2,7	(0,43)	8,4	(0,40)	7,5	(0,98)	6,2	(0,24)	5,6	(0,15)
30 days	3,6	(0,11)	12,2	(0,80)	10,3	(0,16)	8,2	(0,14)	6,2	(0,13)
45 days	4,5	(0,09)	15,5	(0,52)	14,7	(0,36)	8,2	(0,30)	8,9	(0,84)
60 days	5,5	(0,35)	18,3	(0,24)	16,2	(0,82)	13,7	(0,61)	9,9	(0,56)
75 days	7,1	(0,14)	20,8	(0,25)	18,4	(1,27)	15,4	(0,34)	12,8	(0,43)
90 days	9,6	(0,70)	20,9	(0,76)	18,3	(0,53)	17,2	(0,37)	15,2	(0,31)
	Root free soil									
15 days	2,6	(0,37)	8,5	(0,18)	7,8	(0,85)	5,8	(0,22)	4,4	(0,41)
30 days	2,5	(0,16)	11,5	(0,16)	9,6	(0,19)	7,4	(0,39)	5,5	(0,38)
45 days	2,8	(0,16)	12,7	(0,57)	10,5	(0,74)	9,4	(0,74)	7,8	(0,56)
60 days	2,9	(0,61)	13,6	(0,49)	11,4	(0,29)	10,1	(0,08)	8,9	(0,68)
75 days	3,0	(0,48)	13,7	(0,49)	12,7	(0,29)	12,5	(0,37)	9,1	(0,11)
90 days	3,1	(0,22)	15,1	(0,42)	13,2	(0,80)	13,1	(0,27)	11,0	(0,75)

Table 3. Basal soil respiration (BSR) in rhizosphere and root free soils ( $\mu\text{g CO}_2 \text{ g}^{-1}$  dry soil). Standard error in parenthesis.

Incubation Days	Organic wastes									
	Control		TEW		TOW		HH		WS	
	Rhizosphere soil									
15 days	27.5	(4.40)	85.7	(4.12)	29.1	(3.83)	63.9	(2.43)	57.2	(1.53)
30 days	40.1	(1.22)	137.8	(8.97)	116.1	(1.84)	92.2	(1.57)	69.6	(1.49)
45 days	44.4	(0.91)	153.0	(5.10)	144.9	(3.89)	92.2	(2.94)	57.8	(4.31)
60 days	38.3	(2.95)	182.0	(2.95)	161.8	(8.19)	136.5	(6.10)	98.5	(5.56)
75 days	49.9	(5.81)	212.0	(2.50)	134.7	(8.84)	156.5	(3.48)	130.7	(4.39)
90 days	101.7	(7.44)	212.7	(8.10)	194.8	(5.62)	182.5	(3.91)	161.3	(3.25)
	Root free soil									
15 days	17.3	(1.66)	86.8	(1.88)	52.9	(3.89)	42.5	(3.24)	45.6	(4.21)
30 days	28.6	(1.80)	129.8	(1.79)	108.6	(2.14)	83.1	(4.40)	41.6	(3.13)
45 days	18.8	(4.29)	124.9	(5.65)	74.1	(5.74)	92.9	(5.74)	51.7	(3.89)
60 days	28.4	(6.13)	135.4	(4.92)	113.2	(2.90)	100.9	(0.75)	62.4	(4.83)
75 days	30.3	(4.93)	139.6	(4.92)	129.7	(2.90)	127.6	(3.75)	93.1	(1.10)
90 days	33.4	(2.33)	160.6	(4.50)	140.0	(8.51)	139.6	(2.91)	116.5	(8.01)

## Discussion

### Total organic carbon and nitrogen

Total organic C contents in rhizosphere were higher than in root free soil at all organic waste applications (Figure 1). Treatments of TEW and WS gave the highest organic C content in rhizosphere and root free soil compared to the control treatment. In addition, N contents in TOW and TEW treated soils in rhizosphere were significantly greater in all organic waste treatments compared to the control treatment and root free soil. Total N in root free soil were higher than in rhizosphere at all treatments. These situations might be related organic matter and N contents of organic wastes which contain different amounts of organic matter and N (Table 1) and N uptake by plant roots. The differences of C/N ratios of rhizosphere and root free soil were statistically significant for all OW treatments. The TOW and TEW treatments had lower C/N ratio in rhizosphere and root free soil than those in other treatments (HH and WS) (Figure 1). All these changes mostly depended on the characteristics and initial level of organic C and N contents of organic wastes. In general, C/N ratios in rhizosphere and root free soil were lower in soil treated with organic wastes of initial low C/N ratios (TOW and TEW), while treatments with high initial C/N ratios (WS and HH) caused high C/N

ratios in rhizosphere and root free soil. Figure 1 shows that the organic C in rhizosphere were higher than in control treatment and in root free soils at all sampling times and organic waste treatments. This situation might be related supply of organic C material from plant exudates such as polysaccharides, mucigel, carbohydrates and amino acids, and dead cells of root hairs (McGill et al., 1986; Huang and Schoenau, 1998).

Table 4. Dehydrogenase activity (DHA) in rhizosphere and root free soils ( $\mu\text{g TPF g}^{-1}$  dry soil). Standard error in parenthesis.

Incubation Days	Organic wastes									
	Control		TEW		TOW		HH		WS	
	Rhizosphere soil									
15 days	16,9	(0,65)	87,2	(3,40)	76,9	(2,31)	46,2	(1,83)	22,5	(1,43)
30 days	20,8	(0,69)	103,7	(5,96)	80,2	(2,12)	55,5	(2,67)	27,4	(2,94)
45 days	24,5	(0,59)	110,2	(1,90)	96,4	(3,07)	55,5	(2,25)	49,6	(12,52)
60 days	27,9	(2,34)	137,0	(3,56)	116,0	(5,46)	75,7	(4,03)	50,3	(2,39)
75 days	32,4	(2,14)	156,5	(5,68)	138,6	(1,85)	96,1	(4,08)	69,2	(3,24)
90 days	39,9	(2,79)	193,0	(8,73)	171,3	(1,74)	118,0	(10,04)	78,7	(2,66)
	Root free soil									
15 days	14,9	(0,60)	80,4	(1,84)	69,0	(1,18)	39,9	(2,06)	20,1	(0,93)
30 days	14,3	(0,80)	84,5	(3,56)	73,2	(2,18)	45,3	(1,29)	21,9	(1,63)
45 days	15,3	(0,91)	99,3	(3,70)	91,0	(2,75)	58,6	(2,75)	25,5	(1,46)
60 days	15,9	(0,32)	111,2	(3,66)	98,2	(2,81)	60,4	(3,02)	31,3	(2,14)
75 days	15,9	(1,36)	132,5	(3,66)	112,6	(2,81)	79,2	(3,51)	50,0	(2,29)
90 days	16,3	(0,94)	138,6	(8,40)	127,9	(4,00)	87,9	(4,89)	66,1	(7,78)

Table 5. Urease activity (UA) in rhizosphere and root free soils ( $\mu\text{g N g}^{-1}$  dry soil). Standard error in parenthesis.

Incubation Days	Organic wastes									
	Control		TEW		TOW		HH		WS	
	Rhizosphere soil									
15 days	7,0	(0,54)	8,7	(1,06)	16,7	(2,54)	8,0	(0,75)	7,6	(1,17)
30 days	17,2	(2,29)	34,0	(1,95)	32,4	(0,74)	26,1	(1,79)	21,5	(0,92)
45 days	21,8	(1,93)	38,7	(1,69)	39,4	(1,18)	26,1	(2,33)	26,4	(2,09)
60 days	25,6	(1,24)	42,8	(2,24)	46,2	(2,74)	32,3	(1,93)	30,5	(2,14)
75 days	32,4	(2,14)	58,2	(3,02)	58,6	(1,85)	42,8	(2,27)	40,2	(1,53)
90 days	32,9	(2,70)	74,7	(3,09)	71,3	(1,74)	58,6	(3,01)	44,3	(2,73)
	Root free soil									
15 days	5,4	(1,18)	9,1	(1,57)	13,5	(2,85)	8,9	(0,84)	7,1	(1,59)
30 days	14,2	(2,30)	27,2	(1,31)	27,8	(2,91)	23,7	(2,15)	19,2	(1,00)
45 days	13,5	(1,90)	34,3	(1,31)	33,0	(1,27)	25,3	(1,27)	21,7	(1,86)
60 days	15,9	(0,32)	38,4	(2,19)	39,6	(0,96)	29,8	(1,69)	26,5	(1,98)
75 days	15,9	(1,36)	38,9	(2,19)	39,6	(0,96)	32,8	(2,69)	30,0	(2,29)
90 days	19,1	(1,24)	58,6	(2,70)	52,7	(2,84)	40,9	(2,33)	35,6	(0,97)

Table 6. Arylsulphatase activity (UA) in rhizosphere and root free soils ( $\mu\text{g p-NF g}^{-1}$  dry soil). Standard error in parenthesis.

Incubation Days	Organic wastes									
	Control		TEW		TOW		HH		WS	
	Rhizosphere soil									
15 days	22,7	(2,12)	84,4	(4,34)	67,4	(1,90)	47,1	(1,42)	31,1	(1,03)
30 days	28,8	(2,81)	87,3	(3,94)	81,2	(2,79)	59,3	(4,35)	35,0	(2,44)
45 days	31,9	(1,81)	103,1	(4,39)	96,0	(5,13)	59,3	(1,45)	41,4	(3,33)
60 days	39,5	(1,26)	122,3	(2,82)	105,1	(4,96)	84,6	(2,78)	62,4	(3,61)
75 days	45,6	(1,98)	165,0	(9,58)	134,3	(7,86)	97,8	(3,06)	74,5	(2,66)
90 days	58,1	(1,98)	180,4	(10,83)	151,5	(5,50)	129,6	(8,26)	92,5	(2,85)
	Root free soil									
15 days	10,7	(1,34)	38,9	(1,80)	30,8	(1,20)	26,9	(2,97)	17,3	(2,04)
30 days	13,4	(1,01)	61,3	(4,35)	59,5	(2,18)	36,5	(2,99)	19,2	(1,00)
45 days	15,7	(1,18)	74,9	(4,29)	63,0	(1,27)	43,6	(1,27)	33,4	(3,70)
60 days	20,5	(1,30)	94,7	(3,61)	82,1	(2,95)	59,0	(1,91)	44,4	(2,73)
75 days	20,6	(0,97)	124,6	(3,61)	105,7	(2,95)	74,9	(4,16)	73,6	(2,48)
90 days	25,52	(1,01)	141,6	(8,04)	122,0	(4,47)	96,9	(2,37)	66,6	(2,78)

Table 7. Results of ANOVA for BSR and Cmic

Variables	BSR		Cmic	
	F-value	LSD $_{\alpha=0.01}$	F-value	LSD $_{\alpha=0.01}$
Plant root (Pr)	73.210***	8.516	1272.261***	0.200
Incubation days (Id)	67.507***	14.749	981.591***	0.347
Pr x Id	5.060***	20.859	117.093***	0.490
Organic wastes (Ow)	128.453***	13.464	2099.943***	0.316
Pr x Ow	0.435***	19.041	17.559***	0.447
Id x Ow	2.913***	32.981	25.607***	0.775
Pr x Id x Ow	1.217***	46.642	6.615***	1.096

Table 8. Results of ANOVA for enzyme activities

Variables	DHA		UA		ASA	
	F-value	LSD $_{\alpha=0.01}$	F-value	LSD $_{\alpha=0.01}$	F-value	LSD $_{\alpha=0.01}$
Plant root (Pr)	699.030***	1.558	684.391***	0.774	1843.665***	1.492
Incubation days (Id)	851.059***	2.751	1393.715***	1.341	1436.351***	2.584
Pr x Id	46.014***	3.890	67.705***	1.897	8.664***	2.359
Organic wastes (Ow)	3688.078***	2.511	713.288***	1.224	2484.417***	3.665
Pr x Ow	12.647***	3.551	8.349***	1.731	39.071***	3.336
Id x Ow	42.717***	6.150	35.649***	2.999	57.859***	5.778
Pr x Id x Ow	5.413***	8.698	2.475***	4.241	4.246***	8.172

### Biological properties

Different organic waste application significantly affected the levels of biological properties in the rhizosphere, when compared with the control treatment and root free soils. Table 2-6 shows that the BSR, Cmic and enzyme activities (DHA, UA and ASA) in rhizosphere were higher than in control treatment and in root free soils at all sampling times and organic waste treatments. This situation might be related the supply of organic material from plant roots and plant exudates. The supply of organic material from plant roots is crucial to soil microbial communities whose growth is carbon limited. The type and amount of nutrients released will affect both the microbial biomass and their activity. This primary carbon supply to the soil system arrives through plant litter and more directly from roots. These include the release of plant exudates, many of which appear to be simply lost by leakage from the root. Plant exudates contain carbohydrates, amino acids, organic acids, lipids, hormones, vitamins and enzymes. These organic substances are stimulated for soil microbiological activity. It is well known that root-derived organic C from root exudates stimulates the growth of microorganisms and increases microbial activity in the rhizosphere (Toal et al., 2000; Kourtev et al., 2003; Bais et al., 2004). Results from this study also showed the greater biological properties (Cmic, BSR, and enzyme activities such as DHA, UA and ASA) in all organic waste added soils under plant roots compared with root-free soil. Greater biological properties in all organic waste added soils under rhizosphere after 90 days contributed to greater under root free soil. It is likely that increased levels of organic C and N due to root exudation could have led to greater microbial activity. The amount of root-derived C flow through the rhizosphere has a significant impact on transformations of soil organic C, N, P and S (Helal and Sauerbeck, 1989). It has been established that soluble organic C and N in mineral soils is mainly derived from root derived from root exudates and root residues (McGill et al., 1986; Huang and Schoenau, 1998). In the present study, organic C accumulated in the rhizosphere soil, and concentrations of organic C were significantly greater in the rhizosphere compared with root free soil. There were significant relationships observed between biological properties and organic substrates in rhizosphere and root-free soil, indicating that greater value of biological properties such as Cmic, BSR and enzyme activities in the rhizosphere may be partly attributed to increased levels of organic C. It has been suggested that both plant roots and microorganisms produce UA and ASA (Speir and Ross, 1978). It was found that UA and ASA were higher in the rhizosphere compared with control and root-free soil (Table 5 and 6), which is consistent with findings from several other studies (Tarafdar and Jungk, 1987). Moreover, UA and ASA were directly related to value of microbial biomass and their activities in soils.

These tables shows that both rhizosphere and root free soils BSR, Cmic and enzyme activities (DHA, UA and ASA) in all organic waste treatments were higher than in control treatment at all sampling times. This situation may be related carbon source of organic wastes and increased the organic matter level, which consequently elevated the biological properties of soil. For this reason, increased soil organic matter content is correlated positively with microbiological activity in soil, generally. The organic waste treatments had consistent or significant effect on the soil biological properties. This indicated accumulation of organic matter and improvement in nutrient status of soil, as microbial biomass and their activity is a labile

reservoir of plant nutrients (Jenkinson and Ladd, 1981). The BSR, Cmic and enzyme activities (DHA, UA and ASA) in rhizosphere and root free soil for all organic waste treatments was similar in all sampling times (Table 2-6). Addition of organic material increases the microbial activity in soil (Pascual et al., 1997). García-Gil et al. (2000) reported increases microbial biomass and their activity in soil organic waste application to soil. Such increases in rhizosphere and root free soil BSR and Cmic were probably caused by the higher level of soluble organic-C in organic wastes. Availability of biogenic material for biomass stimulation (Jenkinson and Ladd, 1981) induced the increase in soil microbial activity of enriched soils. The increase may also correspond to the growth of the zymogenous population associated with organic matter enrichment (Jenkinson et al., 1987) and incorporation of exogenous microorganisms (Perucci, 1992). The source of enzymes in soil is definitely known, additionally presumed to originate from microorganisms, plant roots and soil animals (Tabatabai, 1994). However, evidence could be obtained from the present study that DHA, UA and ASA of root free soil and rhizosphere were positively related to Cmic and their activity. Perucci (1992) also found positive correlation between enzyme activities and microbial activity. Addition of all organic wastes increased the enzyme activities of rhizosphere and root free soil. This could have originated from the higher amounts of enzymes in the viable microbial populations and the increased levels of accumulated extracellular enzymes (UA and ASA) in the soil matrix. Presence of enzymes in organic matter (Dick and Tabatabai, 1984) may also contribute enzymes directly to soil on addition.

The highest BSR, Cmic and enzyme activities (DHA, UA and ASA) were generally found in rhizosphere and root free soil at TOW and TEW treatments. There have been numerous studies (Pascual et al. 1997; Madejón et al., 2001) on the effects of organic wastes on microbial activity and their enzymatic activities in soil. These studies generally indicated larger effects in organic matter or organic waste treated soils than in control or non-treated soils. However, in most studies it was possible to establish relationships between and biological properties and the magnitude of the effects of organic waste type's especially chemical composition, nutrient content and C/N ratio. Similarly, Martens et al. (1992) suggested that variation in the nature of organic materials variably stimulated the microbial activity and production of enzyme activity in soil. In this research, higher enzyme activities (DHA, UA and ASA) of soil treated with TOW and TEW was associated with their quality in respect to their capacity of microbial biomass production. This situation might be related initial C/N ratios of organic wastes. Organic wastes their C/N ratios are the most important factors that the effects on soil biological properties. Moreover, low C:N ratio and nutrient (N,P,K) sources are essential for the build-up of Cmic and the production or synthesis of enzymes (Alexander, 1977). This can obviously be explained by the input of nutrients in organic wastes and lower C/N ratios prevalent in TEW and TOW. This rose with organic waste, particularly when TOW and TEW were added since this contains a high proportion of easily biodegradable compounds compared with HH and WS. Nitrogen content is also important in determining microbial decomposition rates of organic waste. Higher decay rates are found with increased nitrogen supply (Marinucci et al., 1983). This is similar to the role played by nitrogen in decomposition of other types of organic matter (Mann, 1976; Berg et al., 1982). In addition, nitrogen content of organic wastes has only a positive effect on decomposition rates (Valiela et al., 1985). Because nitrogen has a positive effect on decomposition rates, the trend of increasing nitrogen content within decomposing wastes during the microbially-dominated stage of decomposition is important.

## Conclusion

According to data, this showed a clear relationship between organic wastes and biological properties. We assume that the replacement of organic waste has stimulating effects on biological properties such as Cmic, BSR and enzyme activities (DHA, UA and ASA) in rhizosphere and root free soil, due to the quantity and quality of the organic waste incorporated into soil, and the microbial growth caused by the addition of organic compounds to the soil. Organic materials are possibly the most important C source for microorganisms. It consists mainly of root exudates and organic waste degradation products. Differing organic waste inputs in the system were reflected by the C and N contents which, however, varied much more between the systems than did biological properties. In general, initial low C/N ratios of organic wastes application (TEW and TOW) caused the most beneficial effects on biological properties in rhizosphere and root free soil among the investigated types of organic waste on clay loam soils. The use of these organic wastes can contribute to an enhancement of the level of organic matter and the fertility of the agricultural soils. Furthermore, organic waste had a stronger impact on biological properties in rhizosphere compared to root free soil. Hence, it can be concluded that the biological properties was clearly governed by the organic waste incorporated into soil under the conditions of the investigated greenhouse experiment. At the same time this practice seems to be a potentially effective way of recycling wastes and solving the problem of their disposal.



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