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Mycorrhizal fungi and poultry wastes enhanced soil microbial biomass, metabolic quotient, infection rate, growth and yield of the eggplant

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Abstract---One benefit of incorporating poultry wastes and arbuscular mycorrhizal fungus (AMF) into soils is an increase in crop yield. However, experimental results vary and depend on the kind of poultry wastes, the experimental setup, and the characteristics of the soil. Thus, the underlying causes have not yet been fully understood. In other circumstances, mycorrhizal inoculation and amendment with poultry wastes may have clear impact or negative impacts on soil microbes and plant growth. The experiment was conducted to evaluate the interaction between the fungal mycorrhiza, Italian, local poultry wastes and their effects on the soil biological properties, growth and yield of eggplant *Solanum melongena* L. The mycorrhiza inoculation included three levels (0, 20, and 40 g. spore. The Italian poultry wastes – Atalpolina was added based on three levels (0, 150, 300) kg / 1000 m², while the local poultry was added at three levels (0, 8, 12) tons/ha. The results showed the mycorrhizal fungi (40 g) significantly improved microbial activity, mycorrhizal infection rate, the microbial equation, the percentage of organic carbon and the total yield whereas the local poultry wastes at a level of (12 tons/ha) enhanced microbial activity, microbial biomass carbon (MBC), mycorrhizal infection rate, metabolic quotient, microbial equation, organic carbon %, total yield. We concluded that the large amounts of inoculation of mycorrhizal fungi with the local poultry wastes were more effective in improving MBC and soil quality as well as the yield of the plant.

Keywords---poultry wastes, mycorrhiza, microbial activity, MBC, metabolic quotient, plant yield.

Introduction

Eggplant is one of the most important vegetables in the world. It is rich in antioxidants and vitamins, especially vitamin C. It has substances that promote the health of the heart and arteries, regulating blood pressure, lowering cholesterol in the blood, and reducing toxic metals in the body such as mercury and lead. It also helps in treating diabetes and other diseases (Daunay, 2000). Organic wastes are important agricultural factors in increasing the productivity of vegetable crops. Previous studies have focused on the use of chemical fertilizers, which have proven negative impacts on human, animal health and the environment. Therefore, attention was directed to the use of organic wastes that contain nutrients in a balanced and sufficient way for plant growth. The results of a study by Hao et al. (2008) showed that the addition of organic wastes increased the amounts of nutrients in the plant that encourages the growth of the plant, so the use of these wastes at the right time and in ideal quantities contributes greatly to increasing production. Several studies have been conducted on the possibility of using different types of organic wastes to improve growth and increase the yield in eggplant, Al-Maliki et al. (2022) showed an increase in eggplant productivity after inoculation of the plant with mycorrhizal fungi compared to the control treatment. Mycorrhizal fungus is an important element that may regulate microbial community and organic carbon in the soil as well as nutrients absorption. Mycorrhiza increases phosphorous uptake using the enzyme phosphatase (Qiu et al., 2019) which leads to the promotion of plant growth. In addition, the mycorrhizal fungi secrete glomalin, which contributes to soil fertility (López-Merino. et al., 2015). Similarly, mycorrhizal fungi increase nutrient uptake through symbiotic relationships between roots and plants (Shi et al., 2020).

Mycorrhizal fungi affect soil microbial biomass (SMB) by modulating root exudates after root colonization (Xiao et al., 2019), leading to improved soil properties and plant productivity. Soil microbial biomass (SMB) is an important aspect of soil biological activity and biochemical processes because it is responsible for plant development. Metabolic quotient (qCO_2) is another beneficial process that reflects improvements in soil quality and it is a respiration rate per unit of microbial biomass and is used as an indicator of the ecophysiological circumstances of soil microbes (Anderson and Domsch 1993). It is a sensitive measure of soil carbon quality and can effectively reflect the influence of fungi on the behaviour of soil microbes. As far as the Solanaceae family is concerned, however, interactions of mycorrhizal fungi and poultry wastes are still not studied in the development of soil-microbe relationships. The study aimed to

1. Find the effect of poultry wastes and mycorrhizal fungi on the microbial biomass carbon (MBC) and growth and yield of eggplant
2. Knowing the best level of poultry wastes and mycorrhizal fungi inoculation and their effects on soil biological properties and plant growth.

Materials and Methods

The experiment was conducted in one of the farms of Al-Fayhaa Company for the production of the eggs in Babylon province. The eggplant crop (*Solanum*

melongena L) was cultivated for the agricultural season 2020/2021 near the village of Sinjar, which is about 3 km north of the city of Hilla. Soil texture was silty clay loam with a clay content of 34.13%, silt 51.01%, sand 14.84%. A bulk density was 1.33 g / cm³, and the electrical conductivity of the soil was 2.87 ds / m, while pH was 7.8. The mycorrhizal inoculation was brought from the Agricultural Research Department of the Ministry of Science and Technology and it had (32 spores) in one gram of soil. In addition, two types of poultry wastes were used, namely Iraqi poultry wastes and Italian poultry wastes, which are described in Table (1).

Table (1) some measurements for the Iraqi and Italian poultry wastes

fertilizer type	C/N	C%	N%
local poultry	12.7	52	4.07
Italian poultry	10.3	41	4

Microbial activity in the soil

The activity of microorganisms was measured using a method of alkali trap (Anderson, 1983), where (20) g of soil were placed in a tight glass bottle, then a small beaker was placed containing 20 ml of 1 molar NaOH solution. Then the cover of the bottle was placed tightly, then the bottles were placed under a temperature of 25 °C for periods (24 hours, 7 days, 14 days, and 30 days). We stirred the solution of NaOH with HCl of 0.5 molar until it changes the pink colour to colourless. The amount of CO₂ released was calculated using the following equations:

$$\text{mEq CO}_2 = \text{mEq NaOH} - \text{mEq HCL} \dots\dots\dots (1)$$

$$\text{mEq} = \text{Volume} \times \text{molarity} \dots\dots\dots (2)$$

$$\text{mg CO}_2 = \text{milliequivalent CO}_2 \times \text{equivalent weight CO}_2 \dots\dots\dots (3)$$

Measuring microbial biomass C

Biomass was measured according to the method described by Horwath et al. (1996) by finding the amount of mineralized carbon during ten days of fumigation incubation (CFI) chloroform and then subtracting the amount of mineralized carbon dioxide in the killed samples from the non-acid-killed samples. 20 grams of soil was placed in a tight glass bottle, then a small beaker was placed containing 20 ml of chloroform acid, CH₂Cl₃, which was placed in a water bath for (30 seconds) and the process was repeated four times for four minutes and the samples were left for (24) hours. Then the samples were left open for a period of (3) days to volatilize the smell resulting from the acid, and then the samples were incubated according to the method of measuring the activity of microorganisms above, where the biomass mg/g was calculated on the basis of Dalaal (1998) and Al-Maliki et al. (2020) and according to the following equations

$$\text{Microbial biomass C (mg C/g soil)} = (0.71 \times \text{CO}_2\text{F} - 0.23 \times \text{CO}_2\text{C}) / \text{kC} \dots\dots\dots (4)$$

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Where:-

Microbial biomass C: biomass carbon

Kc: - a value of 0.45 at a temperature of 25 °C (Jenkinson and Powlson 1976; Dalal, 1998).

Metabolic quotient ($\mu\text{g C g}^{-1} \text{ soil}$)

The metabolic quotient **was calculated by dividing the microbial activity at the** incubation period of ten days to the biomass which was resulted from the ten days incubation based on Dalaal, (1998), Al-Maliki et al., (2020) and Insam, (1988) and according to the following equations: -

Metabolism Equation = microbial activity/Biomass MBC..... (5)

Soil microbial quotient (%)

We obtained it by dividing biomass to organic carbon (Insam, 1988)

Soil Microbial Equation = (MBC Biomass) / (Total Organic Carbon)..... (6)

Mycorrhizal infection rate

The method described by Phillips and Hayman, (1970) was adopted.

The AM fungi colonization rate = infected root segments/total root segments * 100% (7)

The percentage of organic carbon (%)

It was estimated by following the method of Walkly and Black, (1947) described in Page et al., (1982) through the wet oxidation of organic matter by potassium dichromate and in the presence of sulfuric acid.

Total yield (tons/ha)

The mature plant was planted on 4/15/2021 manually after the appearance of the final maturity signs of the crop total yield was estimated according to the following equation:

Total yield = (Total Product of the Experimental Unit / Area of the Experimental Unit) * 10000 m²..... (8)

8- Statistical analysis:

The data were analyzed using the Genstate statistics program with the least significant difference (0.05).

Results and discussion

Microbial activity

The results in Table (2) showed that the addition of mycorrhizal fungi to the soil led to a significant increase in the activity of soil microbes. The highest increase in microbial activity was (108.74) mg CO₂. g. soil⁻¹ in M2 treatment The addition of Iraqi and Italian organic wastes to the soil led to a significant increase in the

activity of microbes. It was noted that the treatment of n (i2) recorded the highest average of microbial activity (117.707 mg CO₂.g soil⁻¹). Regarding incubation time, There was a significant increase in the microbial activity at (10 days) incubation (132.606 mg CO₂. gm soil⁻¹) compared to the rest of the other days. There was a significant interaction between time, organic wastes and biofertilizer levels where the combination of (m2I1) at the incubation period (10 days) gave the highest average of microbial activity (153.23 mg CO₂.gm soil⁻¹). The reason for such interaction may be due to the addition of mycorrhiza at the level of (40 g.soil⁻¹) which caused an increase in the activity of microorganisms resulting from the increase in the number of spores that enhanced the fungal numbers and thus increased the fungal hyphae that increase the absorption of nutrients from the soil to the host plant as a part of the symbiotic relationships. Furthermore, the effect was clear when the addition of Iraqi poultry wastes was at the level of (12 tons/ha). The reason for an increase in the activity of soils microorganisms is due to the increase in nutrients, organic carbon and nitrogen, which provides a suitable platform for the growth of mycorrhiza and soil microbes because they obtain an energy source, thus increasing the activity of microorganisms and supplying the plant with the necessary nutrients as shown in Table (2) (Turk et al., 2006). This is also consistent with the study prepared by Hsieh and Hsieh (1990).

Table (2) the effect of adding mycorrhiza, Italian, Iraqi poultry wastes and incubation on the activity of microorganisms (mg CO₂.gm soil⁻¹)

Bio fertilization levels (A)	Organic wastes Levels (B)	Incubation periods (D)				average organic wastes	Biofertilizer average
		24 hours	10 days	20 days	30 days		
0	0	87.3	99	45.2	87.57	79.76	91.214
	P1	102.5	103.5	45.73	57.5	77.307	
	P2	130.6	101.27	45.5	87.03	91.1	
	I1	122.5	119.53	46.37	96.6	96.25	
	I2	131.4	125.47	47.37	96.57	100.202	
M1	0	88.2	122.47	46.87	97.7	88.81	105.9
	P1	102.3	140.83	49.97	111.77	101.21	
	P2	143.5	142.67	50.77	111.8	112.18	
	I1	134.5	141.8	50.43	111.77	109.62	
	I2	152.6	144	54	120.23	117.707	
M2	0	87.7	146.63	54.7	119.67	102.17	108.74
	P1	102.4	145.33	54.6	118.6	105.23	
	P2	137.1	151.47	56.93	102.5	112	
	I1	128.5	153.23	57.7	103.27	110.67	
	I2	142.3	151.9	57.4	102.87	113.61	
L.S.D		For incubation period with interaction ab=7.399				B=4.273	A=3.31
Organic wastes Incubation *		24 hours	10 days	20 days	30 days	average organic wastes	
0		87.7	101.26	45.48	87.37	80.45	

P1	102.4	122.49	46.87	96.96	92.18	
P2	137	141.77	50.39	111.78	110.23	
I1	128.5	145.32	54.43	119.5	111.97	
I2	142.1	152.2	57.34	102.88	113.63	
L.S.D	Incubation *organic = 3.397					
Bio Fertilization * Incubation	24 hours	10 days	20 days	30 days	Bio fertilizer average	
0	114.8	130.97	50.49	103.73	99.99	
M1	124.2	134.3	51.25	103.76	103.73	
M2	119.6	132.55	50.96	103.59	101.67	
L.S.D	2,631 Incubation * Biofertilization					
l.s.d for incubation period = 13.45		132.606	50.9	101.69	average of incubation periods	

(0) control without addition) (M1 Mycorrhizal fungi 20 g/soil) (M2 Mycorrhizal fungi 40 g/soil) (P1 Italian poultry wastes 150 kg/ha) (P2 Italian poultry wastes 300 kg/ha) (I1 Iraqi poultry wastes 8 tons/ha) (I2 Iraqi poultry wastes 12 tons/ha)

Soil microbial biomass (MBC) (mg C/g soil)

The results showed in Table (3) that the addition of Mycorrhizal fungi to soil did not significantly affect the MBC. Welc et al. (2010) mentioned that Mycorrhizal fungi reduced the amounts of microorganisms in the presence of a high amount of cellulose in soil. It is clear that the type of organic matter is the decisive factor in determining the effect of Mycorrhizal fungi on biomass. In a study by Hodge (2000), it was mentioned that the Mycorrhizal fungi alone may increase the amounts of microbes in the soil, reduce or not be affected, so the type of organic matter may be the main reason for this conflict at these studies. The results in Table (3) also indicated the superiority of treatment (I2) in which MBC was (67.6 mg C/g soil) compared to the control treatment, (45.4 mg C/g soil). As for the Italian poultry wastes (Atalpolina), they did not differ from the control treatment. The results also indicated in Table (3) that the combination of i2 with m2 caused a higher MBC (68.2% mg C/g soil). The MBC is the main indicator in the decomposition of organic matter and the release of nutrients and this is consistent with this mentioned by Al-Maliki et al. (2020). The increase in soil microbial biomass in the Iraqi poultry wastes and Mycorrhizal fungi treatment may be due to the high percentage of organic carbon C mentioned in Table (10) and the contents of the available nutrients in addition to the mycorrhizal-microbes secretions which assisted MBC to be raised and this is consistent with Akmal et al., (2004). An amendment with organic matter not only improves soil fertility but also stimulates the development of AM fungi in the soil, which increases soil biomass and biota activity and this is in accordance with what was mentioned in the study prepared by Calvet et al. (1992).

Table (3) Effect of adding Mycorrhizal fungi and the Italian, Iraqi poultry wastes and their effects on MBC (mg C/g soil)

Bio fertilization (A)	Organic wastes Levels (B)					Bio fertilizer average
	0	P1	P2	I1	I2	
0	45.4	52.9	50.2	50.1	66.9	53.1
M1	46.1	53	50.5	50.6	67.6	53.6
M2	46.7	53.1	50.8	51.2	68.2	54
Fertilization average	46.1	53	50.5	50.6	67.6	
L.S.D	AB=20.69		B=11.94		A=9.25	

Mycorrhizal infection rate (%)

The results in Table (4) indicated that the addition of Mycorrhizal fungi significantly affected the Mycorrhizal infection rate, where the treatment M2 was higher than the rest of the treatments, which gave the highest increase (45.8%) compared to the control treatment that did not record any root infection (0.0%). The reason for the increase in the infection rate at the recommended level of (40 g.soil⁻¹) may be due to the increase in the number of spores in the soil, thus increasing the fungal numbers which lead to a definite rise in the mycorrhizal infection. Also, the mycorrhizal fungi have the ability to have a symbiotic relationship with the root hairs and then grow quickly, Furthermore, the extra mycelium might extend from the infected root and it occupies large areas of the surrounding root and soil to provide the plant with nutrients, especially phosphorous and nitrogen (Powell, 1984). The results in Table (4) also indicated that the treatment of the Iraqi poultry wastes (12 tons/hectare) had the highest rate of mycorrhizal fungi (54.8%). The reason why the Mycorrhizal infection rate was higher when adding Iraqi wastes may be due to the increase in the percentage of carbon and nitrogen in the residues of Iraqi wastes, which encouraged the fungi to grow correctly so that the infection rate became higher in Iraqi poultry wastes. As for the combinations of A2 with I2 treatment which achieved the highest rate (54.8%). The reason for such increase may be due to the fact that the recommended dose for the spores of the mycorrhiza (40 g.soil⁻¹) caused an increase in the number of spores in the soil, thus boosting the fungal community.

Table (4) Effect of adding Mycorrhiza and Italian, Iraqi poultry wastes and their effects on the Mycorrhizal infection of the roots(%)

Bio fertilization (A)	Organic wastes Levels (B)					Bio fertilizer average
	0	P1	P2	I1	I2	
0	0	0	0	0	0	0
M1	24	33	33	43.2	54.8	37.6
M2	43.2	54.8	33	43.2	54.8	45.8
Fertilization average	33.6	43.9	33	43.2	54.8	

L.S.D	AB=11.53	B=6.66	A=5.16
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Metabolic quotient ($\mu\text{g C g}^{-1} \text{ soil}$)

The metabolic quotient is one of the most important environmental physiological indicators for the growth of soil microbes and is used to determine the extent of developments in soil quality and plant growth (Al-Maliki et al., 2020; Insam, 1988). The results of table (5) indicate that the addition of Mycorrhiza did not have a significant effect on the metabolic quotient. The treatment of (I2) for Iraqi poultry wastes with organic wastes (12 tons/ha) recorded the highest metabolic quotient ($3.143 \mu\text{g C g}^{-1} \text{ soil}$). Larger doses of fungal mycorrhiza could have a decisive role in improving metabolic quotient together with the Iraqi organic wastes. The reason may be that the Iraqi poultry wastes contain a high percentage of carbon and nitrogen which support the metabolic quotient. Larger doses of Mycorrhiza (40 gm. soil^{-1}), which contains more spores compared to the recommendation (20 gm. soil^{-1}), led to an increase in fungal community and consequently, an increase in biomass and an increase in metabolism, as well as the use of poultry wastes led to an effective response to the biomass in the soil and this is consistent with Jackeline et al. (2018).

Table (5) Effect of adding Mycorrhiza and Italian, Iraqi poultry wastes on the metabolic quotient in soil ($\mu\text{g C g}^{-1} \text{ soil}$)

Bio fertilization (A)	Organic wastes Levels (B)					Bio fertilizer average
	0	P1	P2	I1	I2	
0	2.177	2.317	2.563	2.317	3.12	2.49
M1	2.207	2.283	2.867	2.343	3.15	2.57
M2	2.223	2.283	2.883	2.93	3.16	2.69
Fertilization average	2.202	2.327	2.771	2.35	3.143	
L.S.D	AB=1.045		B=0.603		A=0.46	

Microbial quotient in soil (%)

Microbial quotient, which is a sensitive measure of the carbon quality and it can reflect the soil storage capacity of carbon, Mycorrhiza significantly affected the microbial metabolism. The treatment (M2) recorded the highest increase in microbial quotient (2.374 %). The increase in fungal spores could lead to an increase in the fungal community which led to a clear improvement in the microbial quotient of the soil and the storage capacity of carbon as they respire slowly while storing more carbon. The results also indicate that the addition of organic fertilizer had a significant effect on the microbial quotient, the combination (M2I2) recorded the highest increase in microbial quotient (3.3%).

Table (6) Effect of adding Mycorrhiza and Italian, d Iraqi poultry wastes on the soil microbial equation for soil microorganisms(%)

Bio fertilization (A)	Organic wastes Levels (B)					Bio fertilizer average
	0	P1	P2	I1	I2	
0	0.7	0.97	1.2	1.34	1.35	1.112
M1	1.35	1.82	2.2	2.07	2.87	2.062
M	1.54	1.84	2.77	2.42	3.3	2.374
Fertilization average	1.196	1.543	2.056	1.94	2.506	
L.S.D	AB=2.4		B=1.4		A=1.1	

The percentage of organic carbon in the soil

The results in a table (7) indicated that the addition of Mycorrhiza significantly affected the percentage of organic carbon in the soil, where the treatment (M2) gave the highest increase (3.49%). The results also indicated that the addition of poultry wastes to the soil had a significant effect on the percentage of organic carbon, where treatment (i2) gave the highest percentage of carbon. The combination of (M2i2) gave the highest increase (5.753%). This may be due to the fact that when adding higher doses of mycorrhizal fungi (40 g.soil-1) could increase the number of fungal spores that increase the numbers of fungi that increase organic carbon through their secretions of glomalin and thus increase carbon storage.

Table (7) Effect of adding Mycorrhiza and Italian, Iraqi poultry wastes on soil organic carbon percentage (%)

Bio fertilization (A)	Organic wastes Levels (B)					Bio fertilizer average
	0	P1	P2	I1	I2	
0	1.767	1.01	3.273	3.947	4.997	2.99
M1	1.767	2.107	3.35	4.417	5.047	3.33
M2	1.783	2.153	3.383	4.423	5.753	3.49
Fertilization average	1.772	2.09	3.336	4.262	5.266	
L.S.D	AB=0.568		B=0.328		A=0.254	

The total yield

The results in Table (8) indicated that the addition of Mycorrhizal fungi significantly affected the plant yield, where the treatment (M2) resulted from the highest increase (2180 kg / 1000 m2). The results indicated that organic wastes had a significant effect on the total yield of eggplant. The treatment (i2) gave the highest increase in the yield (2588 kg / 1000 m2). the reason why the Iraqi poultry wastes (12 tons/ha) with Mycorrhiza (40 g / soil) improved plant yield might be due to the role of organic matter in improving the physical and chemical properties of soil and increasing the availability of nutrients and the activity of

microorganisms that leads to an increase in the vegetative growth and thus increasing the number of fruits, and this is consistent with what was mentioned by Abdelrazzag (2002) about the positive role of Mycorrhiza in enhancing plant yield.

Table (8) Effect of adding Mycorrhiza and Italian, Iraqi poultry wastes on the total yield (kg / 1000 m²)

Bio fertilization (A)	Organic wastes Levels (B)					Bio fertilizer average
	0	P1	P2	I1	I2	
0	742	875	892	1406	1394	1062
M1	1078	1591	1983	2289	3106	2009
M2	1413	1675	2075	2475	3264	2180
Fertilization average	1078	1380	1650	2056	2588	
L.S.D	AB=263.8		B=152.3		A=118.0	

Conclusion

The addition of mycorrhiza alone led to a significant increase in the microbial activity, mycorrhizal infection, organic carbon, plant height and eggplant yield, but we did not notice a significant increase in the rest of the studied parameters. The addition of local poultry wastes and Italian poultry wastes increased the eggplant yield and the soil nutrients as long as microbial activity. The use of local organic wastes (poultry) with mycorrhizal inoculation at 40 g spores made the best contribution to improving soil parameters and plant growth of eggplant.

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