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Defying Role of Specific Microorganisms on Efficiency of Organic Materials (*Olive Pomace*) on the Growth and Productivity of Olive Trees

El-Taweel, A. A.*, I. M. S. Osman, Abou-Taleb-Safia and Abou El-Wafa, M.

Department of Olive and Semi-Arid Zone Fruits, Horticulture Research Institute (HRI), Agricultural Research Center (ARC), Egypt.

*** Corresponding author**

El-Taweel, A. A.

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ABSTRACT

This study aimed to enhance the quality of olive pomace by adding microorganisms that can degrade phenolic compounds and make it suitable for cultivation. The study was conducted for three consecutive seasons (2012, 2013 and 2014) on 12-year-old Manzanillo olive trees in a private orchard located 50 km from Cairo – Alexandria Road in northwest of Egypt. The effects of two microorganisms (*Trichoderma virdi* and *Pseudomonas fluorescens*) on the growth and productivity of olive trees were evaluated. The experiment consisted of 9 treatments, each with 2 trees replicated 3 times in a randomized block design. The results showed that the treatment with compost 50 % + pomace 50 % + *Pseudomonas fluorescens* had the highest fruit set (%), yield, remaining fruits and leaf nitrogen and potassium. Meanwhile, the treatment with compost 75% + pomace 25 % + *Trichoderma virdi* + *Pseudomonas fluorescens* had the highest number of leaves and inflorescence/meter.

It could be recommended under the same conditions of this study, using compost (Comp.) 50 % + Pomce (Pom.) 50 % + *Pseudomonas fluorescens* (Pseu.) to increase the productivity of Manzanillo olive trees.

Keywords: Olive, Pomace, *Pseudomonas fluorescens* (Pseu.), Compost, microorganisms.

1. INTRODUCTION

Olive oil is a major economic product in most Mediterranean countries, with an annual production of about 3,207,000 million tons (International Olive Council, 2019). However, olive oil production also generates a large amount of olive mill waste in a short period of time, which poses a significant environmental challenge.

Seventy percent of the local olive production is used for oil manufacturing, which results in by-products that can cause serious environmental and storage problems, especially given the high volume of production (Aktas et al., 2001).

The press process and the three-phase centrifuge system account for 70 percent of the olive oil production, but they also generate a large amount of: (a) pure olive oil, (b) olive mill wastewater (OMWW) and (c) olive mill waste (pomace) (Morillo et al., 2009). The pomace (waste from olive mills) is a highly organic and chemically stable waste that can resist degradation. (Isidori et al., 2005). Therefore, it is a valuable product from both ecological and economic perspectives, as it can reduce the need for chemical fertilizers, enhance soil protection and fertility, and increase bacterial activity. Applying these materials to the soil can benefit the soil structure, nutrient availability and solubility in various ways. (MP et al., 2007).

Microorganisms play a vital role in the composting process, as they break down organic waste into substances that can nourish plants or soil. The main microorganisms involved in composting are bacteria and fungi (Alburquerqueet al., 2006), especially the fungus species (*Trichoderma viridi*) and the bacterium (Pseudomonas fluorescens). These microorganisms help to decompose soil organic matter and cycle essential nutrient elements, such as N, S and P. The microbial composition of the soil has important implications for its ecosystem functions (Reed &Martiny, 2007).

Therefore, the addition of microorganisms to organic materials (olive pomace) and compost led to an increase in productivity and crop quality.

So, the preparation of olive pomace using bacteria and fungi is important for improving fertility status and productivity. On the basis of these findings, this study aimed to assess the effects of reusing treated pomace (waste from olive presses) with *Pseudomonas fluorescens* and *Trichoderma viridi* as a biofertilizer and to determine the best application methods for increasing the yield of Manzanillo olive trees and improving soil quality in a sustainable farming system.

2. MATERIALS and METHODS

Field experiment

1.Field practices

Field experiment was conducted on a 13-years old olive trees cv. Manzanillo during 2012,2013 and 2014 on sandy clay soil in a private orchard (FiFa farm) about 50 km from Cairo-Alexandria Road in the northwest of Egypt to improve the physical and chemical properties of olive pomace by adding microorganisms specific to phenolic substances, which leads to their disposal and suitability for cultivation. Trees were planted at a distance of 5 by 5 meters (168 trees/fed.),(four dippers/tree)and grown under standard cultural practices and moderately pruned. Trees were almost uniform in growth and subjected to the same management treatments. Organic manure was applied as compost, it was applied in the second week of November in two parallel ditches of 100 x 40 x 30 cm, for length, width and depth, respectively. The ditches surrounded the tree on two sides at the end of the canopy's shade. Mineral fertilization was added during the season at a rate of 100gm. nitrogen + 270 gm potassium + 500 gm of $MgSO_4$ (9.6 % Mg) + 250 gm sulphur + 250 gm boron (Borax)/ tree.

Two strains (Pseudomonas fluorescens and Trichoderma virdi) were provided from Department of Microbiology; Soils, Water and Environment Research Institute (SWERI); ARC, Giza-Egypt to be inoculated into substrate medium (OMW).

Two strains (*Pseudomonas fluorescens* and *Trichoderma virdi*) inoculums were applied at a 5 cm immersion application (depending on the treatments) at the position of the periphery of the canopy during the period from 1st January until the end of fruit set (6 months) every two weeks. Pomace was added to treatment No.2 and treatment No.3 without microorganisms, while treatments from 4 to 9 with microorganisms. All treatments such as the soil drench application in the position of the perimeter of the canopy during the period from the first of January until the end of the fruit set (6 months) every two weeks.

2.Treatments

Fifty-four nearly uniform trees were selected for this study. The experiment included 9 treatments represented by 3 replicates (2 trees per each).

The treatments were arranged as follows.

1-Control (compost 100%).

2- Compost 75 % + Pomace25%.

3- Compost 50 % + Pomace 50%.

4- Compost 75 % + Pomace 25 % + *Trichoderma virdi*

5- Compost 75 % + Pomace 25 % +Pseudomonas fluorescens.

6- Compost 75 + Pomace 25 % + Trichoderma virdi+ Pseudomonas fluorescens.

7-Compost 50 % + Pomace 50 % + Trichoderma virdi.

8- Compost 50 % + Pomace 50 % + Pseudomonas fluorescens.

9- Compost 50 % + Pomace 50 % + Trichoderma virdi+ Pseudomonas fluorescens.

3.Measurements

3.1.Soil sampling and analyses

Soil samples were randomly collected from the zone of the root tips of the trees in the perimeter of the tree canopy at the end of each growing season. Soil sampling depth was 0-60 cm.(Fernandez Je et al., 1991).

Soils were analyzed as shown in Table (1) according to (Chapman and Pratt,1962).

3.2.Soil chemical analyses

Air-dried soil samples were ground to pass through a 2 mm sieve using a wooden mill and stored in plastic bottles prior to the chemical analysis.

Table (1). Physical and chemical analysis of the soil.

Particles size distribution		**Chemical analysis**	
Coarse sand (%)	4.93	EC dS/m (1:2.5)	4.35
Fine sand (%)	77.18	pH (1:2:5)	8.1
Silt (%)	11.9	Organic matter (%)	0.26
Texture	Loamy sand	Organic carbon (%)	0.15
Total nutrients (mg/100g)		**Total nutrients (mg/kg)**	
N	0.17	Mn	1.35
P	0.4	Zn	0.46
K	7.4	Fe	1.6

Irrigation water analysis is conducted in the Soil and Water Research Institute (SWRI), ARC as shown in Table (2).

Table (2).Chemical analysis of well's water.

E. C. W	**TSS (ppm)**	**pH**	**Cations (meq/L)**				**Anions (meq/L)**			
(mmhos)			**Mg^{++}**	**Ca^{++}**	**K^{+}**	**Na^{+}**	**SO_4**	**Cl^{-}**	**HCO_3**	**CO_3^{-}**
0.84	537.6	7.4	2	4	0.2	2.1	0.2	6.1	2	Few

3.3. Physical and chemical analysis of compost.

Table (3). Physical and chemical analysis of compost.

Parameter	Value	Parameter	Value
pH (1: 2.5)	**9.01**	Total P (%)	0.54
EC dS/m (1:2.5)	**7.89**	Total K (%)	2.64
TNOrganic carbon (%)	1.95	N NH4 (ppm)	657.9
Organic matter (%)	**28.5**	N No3 (ppm)	152.9
C/N ratio	**14.62**		

3.4.Pomace(Olive mill waste)

Pomace (OMW) was obtained from FIFA farm (km 50 of Cairo-Alexandria, Egypt)during the late fall and winter season. The raw OMW samples were generated by the three-phase olive oil extraction process for 2012,2013 and 2014 seasons. The pomace and compost were analyzed for their physical and chemical properties according to Paredes et al. (2005). Table (4) shows the results of the analysis.

Table (4):Analysis of only pomace, compost and both of them with microorganisms.

Parameter	Pomace only	Compost only	Compost + pomace
PH	6.25	9.01	8.75
EC	2.40	7.89	4.00
T.N (%)	0.79	1.95	0.93
Organic c (%)	32.71	28.5	12.13
C/N ratio	41.41	14.62	13.04
Organic matter	56.26	49.02	20.86
TotalP(%)	0.05	0.54	0.37
TotalK (%)	1.20	2.64	1.34
N-NH$_4$(ppm)	15.5	657.9	179.5
N-No$_3$(ppm)	4.44	152.9	37.70

3.5.Vegetative growth characteristics

At the end of each growing season during the first week of August, the following characteristics were measured:

3.5.1. Average shoot length (cm)

For each season of the study, five one-year-old shoots were randomly selected from each direction on each evaluated tree the average shoot length (cm) and number of leaves per shoot were measured during two studied seasons.

3.5.2. Leaves density = number of leaves / shoot and calculated in meter.

3.5.3.Leaf area (cm^2): using a planimeter device according to (**Aly, 2005**).

3.6. Flowering parameters

3.6.1.Length of inflorescence

Thirty inflorescences were randomly taken from each replicate and the length of their axis was measured.

3.6.2. No. of inf. /meter

The average number of inflorescences per shoot was recorded and calculated per meter.

3.6.3. Perfect flower (%)

Twenty inflorescences were collected at the balloon stage from the middle parts of the shoots, from each tree, following the method of **Rallo and Fernández-Escobar (1985**).

Perfect flowers (%) = Number of perfect flowers / Number of total flowers x 100

3.7.Fruit set and yield

3.7.1.Initial fruit set (%)

The percentage of fruits for each tree was recorded on twenty tagged shoots then, the set of fruits was recorded on each replicate tree after 21 days from full bloom according to the method of (**de la Rosa et al.2008**) and their percentages of No. of total flowers /tree were calculated.

3.7.2. Final fruit set and yield/ tree(kg)

The final fruit set was counted and average yield (kg) per tree was recorded at the ripe stage (olive with superficial pigmentation on more that 50% of the skin) for each replicate tree.

3.7.3. Fruits oil content (%):

It was determined as dry weight by extracting the oil from the dried fruits with soxelt fat extraction using petroleum ether as described by method in (**A.O.A.C (1995**).

3.8. Fruit physical characteristics

Thirty fruits per each tree at the ripe stage were randomly selected in three seasons from each replicate tree to determine physical and chemical characteristics of fruits in different treatments as the following:

3.8.1. Fruit length (cm), fruit diameter (cm) and fruit shape index.

In each fruit sample, fruit length and fruit width were measured. The fruit shape indexes (L/D) was also recorded according to (**Buyanovandvoronuk 1985**) as follow:

Where $I=L/D_i$

I=shape index

L=length of fruit.

D=diameter of fruit at the middle of the length.

3.8.2. Fruit weight (gm).

3.8.3. Stones and Flesh/fruit

3.8.1. Stones were extracted from the selected fruits to determine their length (cm), diameter (cm) and weight (g).

3.8.2 Flesh/fruit weight percentage was calculated according to the following equation:

Flesh weight (g) = average fruit weight-average stone weight.

Flesh/fruit (%) = Flesh weight/ average fruit weight x100 according to (**Fouad *et al.*, 1992**).

3.8.4. Leaf minerals content

At the first week of August in both seasons, 50 samples of mature leaves, from previously tagged non-fruiting shoots on each replicate, were taken from the upper third of shoot top as recommended by **(Piper, 1919)**.

Sample 200 gm. of fresh leaves were cleaned and washed several times, with tap water. The samples were air-dried and put in an electrical oven at 70°C to reach a constant weight and finally ground to be used for preparing the wet digested solution (**Piper, 1919**) which must be ready for macro nutrient analyses and calculated as percentage of dry weight.

Then, the following determinations were carried out in acidic digested solutions as reported by (**Van Shouwonburg1968**) as follow:

- Total nitrogen was determined by a modified micro-Keyldahl method as described by (**Pregl, 1968**).
- The Phosphorus concentration was determined by the colorimetric method described by (**Murphy and Riely 1962**).
- The Potassium concentration was determined by a flame photometer (**Brown and Lilleland, 1946**).

3.8.5. The experimental designed

All data were tested for the effect of treatments on analyzed parameters by one-way analysis of variance (ANOVA), according to (**Snedecor and Cochran, 1980**). Differences between treatments were compared by Duncan's multiple range tests SAS (**Stromberger et al., 1994**).

4. RESULTS AND DISSUSSION

4.1. Effect of pomace application on compost physical and chemical properties:

As Table (4) indicates the compost with pomace (OMW) resulted in lower soil pH, organic carbon, and organic matter, and higher soil EC. The waste's phenolic acids might have degraded and lowered the pH. As for the soil EC, due to the high soluble salts content. (**Fornes*et al.*,2009**). There was an increase in the availability of phosphor and potassium, while the C/N ratio decreased. Increasing K% and P% may have a beneficial effect on soil fertility. These findings are supported by previous investigations (**Alburguerque*et al.*,2011**).

4.2. Effect of compost and olive pomace wastes with/without microorganisms on some vegetative growth parameters.

As Table (5) indicates, the tested treatments significantly improved shoot growth parameters (length, number of leaves/meter and leaf area) of Manzanillo olive trees over three seasons compared to the control. The highest leaf area was achieved by Comp.50% + Pom. 50 % plus Pseu and Comp. 50 % + Pom. 50% + Tri. + Pseu., respectively. These findings agree with previous studies that reported positive effects of olive waste (OVW & OPW) on young shoot length, the number of new shoots, and leaf area of olive trees (**Abbasi et al., 2013 and El-Taweel *et al.*, 2016**).

Table (5): Effect of compost and olive pomace wastes with / without microorganisms treatments on some vegetative growth parameters of Manzanillo olive cultivar during 2012, 2013 and 2014 seasons.

Parameters / Treatments	Shoot length (cm)			No. of leaves/meter			Leaf area (cm^2)		
	2012	2013	2014	2012	2013	2014	2012	2013	2014
1- Control (compost 100 %)	11.63E	21.20B	19.27E	112.3F	122.2B	124.0CD	3.84C	3.82C	3.80D
2- *Comp. 75 % + **Pom. 25 %	14.30D	24.30A	20.93CD	115.4E	115.9C	125.3C	3.98A	3.76D	3.76E
3- Comp. 50 % + Pom. 50 %	19.57AB	18.27D	22.03C	121.3AB	120.7B	123.5D	3.98A	3.77D	3.93A
4- Comp. 75 % + Pom. 25 % + ***Tri.	19.57AB	21.63B	21.80C	119.4BC	121.8B	123.5D	3.97A	3.75D	3.95A
5- Comp. 75 % + Pom. 25 % + ****Pseu.	18.23BC	18.03D	16.97F	121.8A	121.0B	122.5DE	3.84C	3.83C	3.94A
6- Comp. 75 + Pom. 25 % + Tri. + Pseu	18.67B	19.70C	26.70A	123.0A	124.2A	128.4B	3.90B	3.88B	3.86C
7- Comp. 50 % + Pom. 50 % + Tri.	14.17D	18.13D	22.23C	117.3D	113.3D	127.4B	3.91B	3.90B	3.89B
8- Comp. 50 % + Pom. 50 % + Pseu.	20.33A	21.27B	24.67B	113.8EF	115.4C	131.8A	3.99A	3.97A	3.95A
9- Comp. 50 % + Pom. 50 % + Tri + Pseu.	17.17C	19.47CD	19.63DE	118.4CD	116.0C	121.3E	3.97A	3.95A	3.94A

Means with the same letter (s) within the same column are not significantly differ at the probability of 5 % level according to Duncan`s Multiple Range test.

* Comp: compost ** Pom.: Pomace
*** Tri.: *Trichoderma virdi* **** Pseu.: *Pseudomonas fluorescens*

4.3. Effect of compost and olive pomace wastes with/without microorganisms on flowering parameters.

The flowering characteristics of Manzanillo olive trees were significantly influenced by compost and the olive pomace with microorganisms treatments over three seasons, as shown in Table (6). The length of inf. (cm) was highest for Comp. 75% + Pom. 25% plus Tri., in the first and second seasons (2.54 and 3.04), and for the control in the third season (2.57). The No. of inf./meter was highest for Comp. 50% + Pom. 50% plus Pseu., in the first season (87.49), and for Comp.75 + Pom.25% + Tri. + Pseu./tree in the second and third seasons (81.42 and 86.39). The perfect percentage was highest for Comp.50%+ Pom.50% plus Pseu in the first and third seasons (85.24), and for Comp.75 + Pom. 25 % + Pseu./tree in the second season (72.40). These results may be due to the effect of these materials on soil properties and microbial biomass (**Haynes and Swift 1990; Noval and Rezk 2009, MP, et al.,2007**). They found that these materials can enhance soil fertility by providing nutrients and organic matter (OM), improving soil physical characteristics such as density, porosity, structure, aggregation, water retention and transmission, and altering solubility directly by retaining water (hydrophilic mature) and indirectly by changing soil pH, redox conditions, and salinity.

Table (6): Effect of compost and olive pomace with/without microorganisms treatments on flowering characteristics of Manzanillo olive cultivar during 2012, 2013 and 2014 seasons.

Parameters / Treatments	Length of Inf. (cm)			No. of inf./meter			Perfect flower (%)		
	2012	2013	2014	2012	2013	2014	2012	2013	2014
1- Control (compost 100 %)	2.20C	1.82 D	2.57 A	55.32 H	55.91 H	56.71 F	43.20I	43.51 H	45.40 G
2- *Comp. 75 % + **Pom. 25 %	2.20C	2.53F	2.71 G	55.10 H	56.33 G	52.07 G	45.31 H	49.30 G	45.85 G
3- Comp. 50 % + Pom. 50 %	1.80 G	2.54F	2.89 D	57.64 G	65.21 F	59.77 E	47.38 G	51.27 F	56.12 C
4- Comp. 75 % + Pom. 25 % + ***Tri.	2.54 A	3.04 A	3.04B	65.69 F	67.10 E	56.50 F	60.09 E	62.55 E	51.29 F
5- Comp. 75 % + Pom. 25 % + ****Pseu.	1.97F	2.96B	2.95C	69.82 D	75.52 D	63.55 D	57.20 F	72.40 A	52.89 E
6- Comp. 75 + Pom. 25 % + Tri. + Pseu	2.07E	2.90C	2.70 G	80.11 B	81.42 A	86.39 A	69.85 B	62.36 E	74.98 A
7- Comp. 50 % + Pom. 50 % + Tri.	2.26B	2.81 D	2.62 H	66.95 E	67.11 E	64.26 C	64.14 D	66.09 D	51.09 F
8- Comp. 50 % + Pom. 50 % + Pseu.	1.95F	2.61E	2.80F	87.49 A	79.21 B	56.85 F	85.24 A	67.89 C	66.45 B
9- Comp. 50 % + Pom. 50 % + Tri + Pseu.	2.13 D	2.43 G	2.84E	74.22 C	78.27 C	76.36 B	67.94 C	69.06 B	54.37 D

Means with the same letter (s) within the same column are not significantly differ at the probability of 5 % level according to Duncan`s Multiple Range test.

* Comp: compost ** Pom.: Pomace

*** Tri.: *Trichoderma virdi* **** Pseu.: *Pseudomonas fluorescens*

4.4.Effect of compost and olive pomace wastes with/without microorganisms on fruit set and yield

As shown in Table 7, the tested treatments significantly enhanced Manzanillo olive fruit, final fruit set and yield over the control trees. The highest percentages of initial fruit set (15.93, 25.92 and 18.66), final fruit set (17, 27.83 and 19.93) and yield (22.50, 24.70 and 16.87) in three seasons were achieved by Comp. 50% + Pom. 50 % plus Pseu. The fruit oil content (D. W) of Manzanillo olive trees was highest for Comp.50% + Pom.50 % plus Tri./tree in three seasons (33.91,33.20 and 32.88) compared to the control. These results agree with previous studies that reported positive effects of Pseudomonas bacterial strains on fruit set (**Abbasi et al., 2013**), of OWW on crop productivity (**Saadi et al., 2007**), and of OMW on soil microbiological activity and fertility (**Rinaldi et al., 2003; Mechri et al., 2008**). Similarly, pomace can enhance soil microbiology and fertility by mineralizing OM elements (**Mechri et al., 2008**). Moreover, composting of olive leaves and olive pomace can increase lettuce yield by 145% (**Michailides et al., 2011**).

Table (7): Effect of compost and olive pomace wastes with/without microorganisms treatments on fruit set, remaining fruits and yield of Manzanillo olive cultivar during 2012, 2013 and 2014 seasons.

Parameters	Initial fruit set (%)			Final fruit set (%)			Yield (kg/tree)			Fruit oil content (D.W)		
Treatments	2012	2013	2014	2012	2013	2014	2012	2013	2014	2012	2013	2014
1- Control (compost 100 %)	11.83 G	11.5 3I	13.28 H	11.97 G	12.8 3I	14.33 G	4.83I	7.93I	8.53 G	23.6 5G	22.8 8E	21.7 6H
2- *Comp. 75 % + **Pom. 25 %	12.65 F	15.1 5H	13.67 G	12.90 F	16.1 7H	14.33 G	9.10 H	13.8 7H	13.3 3D	25.7 4E	24.6 6C	23.8 8D
3- Comp. 50 % + Pom. 50 %	13.44 E	16.0 6G	14.85 DE	14.33 E	17.1 7G	15.63 EF	10.9 0G	15.1 0G	13.9 0C	24.8 5F	24.1 3D	23.0 4F
4- Comp. 75 % + Pom. 25 % + ***Tri.	14.62 C	17.4 7F	14.32 F	14.90 CD	18.7 3F	15.27 F	15.4 7F	16.1 7F	12.2 0E	26.8 6C	26.1 6B	25.0 3C
5- Comp. 75 % + Pom. 25 % + ****Pseu.	14.32 D	20.4 1E	14.91 D	15.30 C	21.8 3D	16.03 DE	19.8 3C	17.7 7E	10.5 7F	23.3 7H	22.1 6F	20.4 4I
6- Comp. 75 + Pom. 25 % + Tri. + Pseu	14.92 B	24.6 5B	17.60 B	16.00 B	26.4 0B	18.33 B	18.8 0D	22.0 7B	14.0 7C	30.7 5B	22.0 6F	28.5 8B
7- Comp. 50 % + Pom. 50 % + Tri.	14.48 CD	22.2 5D	16.55 C	15.33 C	23.8 3C	17.67 C	21.8 3B	20.6 7C	15.4 2B	33.9 1A	33.2 0A	32.8 8A
8- Comp. 50 % + Pom. 50 % + Pseu.	15.93 A	25.9 2A	18.66 A	17.00 A	27.8 3A	19.93 A	22.5 0A	24.7 0A	16.8 7A	26.1 3D	24.0 6D	23.2 3E
9- Comp. 50 % + Pom. 50 % + Tri + Pseu.	13.58 E	23.0 5C	14.66 E	14.67 DE	20.6 7E	16.33 D	17.8 3E	18.9 0D	14.1 3C	25.8 5E	24.0 8D	22.0 4G

Means with the same letter (s) within the same column are not significantly differ at the probability of 5 % level according to Duncan`s Multiple Range test.

* Comp: compost ** Pom.: Pomace

*** Tri.: *Trichoderma virdi* **** Pseu.: *Pseudomonas fluorescens*

4.5. Effect of compost and olive pomace wastes with/without microorganisms on leaf mineral contents.

As Table (8) shows, the olive pomace wastes with microorganisms treatments significantly influenced macro elements (N, P and K %) concentration in Manzanillo olive leaves over three seasons. The highest nitrogen percent (1.963) in the first season and (1.68 and 1.75) in the second and third seasons was achieved by Comp. 50 % + Pom. 50 % + Pseu. The highest phosphorus content (0.190, 0.307 and 0.363) in three seasons was achieved by Comp.75% + Pom.25%. The highest potassium percent (1.94 and 1.97) in the first and second seasons and (1.97) in the third season was achieved by Comp. 50%+Pom.50% plus Pseu./tree and Comp. 50% +Pom. 50%, respectively. The findings discussed are in line with studies reported by (**Bardi and Malusa, 2012)**, which demonstrated the positive impact of microorganisms on nutrient absorption. Additionally, other research has shown that bio fertilizers can affect soil properties by containing various strains of symbiotic associative diazotrophes, solubilizing microorganisms of phosphate, and silicate dissolving microorganisms (**Baradiand Malusa, 2012**). Potassium may play a role in the synthesis of chlorophyll pigments, which may explain the positive effect observed on nutrient content in leaves. Furthermore, **Zewail et al., (2015)** found that Pseudomonas fluorescens is capable of producing IAA and cytokine, has N_2-fixing capacity, and M3 has phosphate-solubilizing capacity. The improvement of nutrient content in leaves may be due to the effective role of microorganisms in enhancing the absorption, translocation, and accumulation of mineral contents in leaves (**Hikal, 2000**). Furthermore, **Cruz-Méndez et al.,(2021)** discovered that microbes play significant roles in nutrient cycling, biodegradation/biological degradation, and can have both positive and negative responses to temperature, making them an important component of climate change models.

Table (8): Effect of compost and olive pomace wastes with/without microorganisms treatments on leaf mineral contents of Manzanillo olive cultivar during 2012, 2013 and 2014 seasons.

Parameters / Treatments	N (%)			P (%)			K (%)		
	2012	2013	2014	2012	2013	2014	2012	2013	2014
1- Control (compost 100 %)	1.44EF	1.33H	1.21E	0.150B	0.210G	0.240E	1.45H	1.52E	1.47G
2- *Comp. 75 % + **Pom. 25 %	1.57D	1.43F	1.23E	0.190A	0.307A	0.363A	1.65F	1.86BC	2.38B
3- Comp. 50 % + Pom. 50 %	1.96A	1.64C	1.48BC	0.200A	0.290AB	0.350AB	1.53G	1.89B	2.44A
4- Comp. 75 % + Pom. 25 % + ***Tri.	1.73B	1.53E	1.44C	0.147B	0.250DE	0.380A	1.82B	1.81CD	2.22E
5- Comp. 75 % + Pom. 25 % + ****Pseu.	1.47E	1.35G	1.32D	0.150B	0.277BC	0.323BC	1.68E	1.84B-D	2.27D
6- Comp. 75 + Pom. 25 % + Tri. + Pseu	1.47E	1.66B	1.75A	0.120C	0.223FG	0.247DE	1.72D	1.79D	2.19F
7- Comp. 50 % + Pom. 50 % + Tri.	1.42F	1.44F	1.53B	0.120C	0.263CD	0.283CD	1.75C	1.81CD	2.27D
8- Comp. 50 % + Pom. 50 % + Pseu.	1.44EF	1.68A	1.75A	0.120C	0.237EF	0.290C	1.94A	1.97A	2.34C
9- Comp. 50 % + Pom. 50 % + Tri + Pseu.	1.62C	1.56D	1.48BC	0.110C	0.290AB	0.307C	1.73D	1.80CD	2.28D

Means with the same letter (s) within the same column are not significantly differ at the probability of 5 % level according to Duncan`s Multiple Range test.

* Comp: compost ** Pom.: Pomace

*** Tri.: *Trichoderma virdi* **** Pseu.: *Pseudomonas fluorescens*

4.6. Effect of compost and olive pomace wastes with / without microorganisms on fruit physical characteristics.

According to **Khalil et al., (2020)**, *Phanerocheatechysosporium* has shown great potential in removing phenolic compounds and breaking down lignin and cellulose in olive pomace. On the other hand, *Trichoderma viridi,* a fungus species, plays a crucial role in assisting plants in coping with various environmental stresses, including drought and nutritional stress. Moreover, *Trichoderma viridi* is known for producing enzymes, particularly cellulose, which are essential for plant growth and development (**Reino et al., 2008**).

Table (9): Effect of compost and olive pomace wastes with/without microorganisms treatments on fruit length, fruit diameter, shape index and weight (g) of Manzanillo olive cultivar during 2012, 2013 and 2014 seasons.

Parameters / Treatments	Fruit length (cm)			Fruit diameter (cm)			Fruit shape index			Fruit weight (g)		
	2012	2013	2014	2012	2013	2014	2012	2013	2014	2012	2013	2014
1- Control (compost 100 %)	2.48BC	2.36C	2.15B	1.90G	1.85G	1.80BC	1.31A	1.28B	1.19B	3.78E	4.12E	3.79F
2- *Comp. 75 % + **Pom. 25 %	2.63A	2.66A	2.15B	2.07CD	2.01F	1.76D	1.27B	1.32A	1.22A	3.65F	4.19D	3.74F
3- Comp. 50 % + Pom. 50 %	2.57AB	2.50B	2.16B	2.11BC	2.09D	1.86A	1.22D	1.20C	1.16C	4.00C	4.36C	3.96E
4- Comp. 75 % + Pom. 25 % + ***Tri.	2.45C	2.30D	2.11CD	2.13B	2.15C	1.86A	1.15F	1.07E	1.13D	4.03C	4.48B	4.03D
5- Comp. 75 % + Pom. 25 % + ****Pseu.	2.46C	2.25E	2.17B	2.00F	2.19B	1.82BC	1.23CD	1.03F	1.19B	3.84E	4.01F	3.81F
6- Comp. 75 + Pom. 25 % + Tri. + Pseu	2.36D	2.28D	2.08D	1.98F	2.08D	1.81BC	1.19E	1.10D	1.15CD	3.92D	4.07EF	3.92E
7- Comp. 50 % + Pom. 50 % + Tri.	2.52BC	2.21F	2.10D	2.23A	2.25A	1.83AB	1.13F	0.98G	1.15CD	4.36B	4.40C	4.54B
8- Comp. 50 % + Pom. 50 % + Pseu.	2.55B	2.18G	2.08D	2.02EF	2.05E	1.80BC	1.26B	1.06E	1.16CD	4.29B	4.36C	4.29C
9- Comp. 50 % + Pom. 50 % + Tri + Pseu.	2.55B	2.26E	2.25A	2.05DE	1.88G	1.82BC	1.24C	1.20C	1.24A	4.61A	4.68A	4.61A

Means with the same letter (s) within the same column are not significantly differ at the probability of 5 % level according to Duncan`s Multiple Range test.

* Comp: compost ** Pom.: Pomace

*** Tri.: *Trichoderma virdi* **** Pseu.: *Pseudomonas fluorescens*

4.7. The effect on Stone length, diameter and weight

Table (10) shows that the tested treatments significantly enhanced stone length, diameter and weight of Manzanillo olive trees over three seasons. The stone length was highest for Comp. 50 % + Pom. 50 % + Tri. + Pseu./tree in all three seasons (1.66; 1.67 and 1.68 cm). The stone diameter was highest for Comp. 50 % + Pom. 50 % + Tri., + Pseu./tree in the first season (1,000 cm), for Comp.50 + Pom.50% + Pseu., in the second and third seasons (0.937 and 0.943 cm). The stone weight was highest for Comp.50 + Pom.50% + Pseu., in all three seasons compared to the control.

Table (10): Effect of compost and olive pomace wastes with/ without microorganisms treatments on stone length, diameter (cm) and weight (g) of Manzanillo olive cultivar during 2012, 2013 and 2014 seasons.

Parameters	**Stone length (cm)**			**Stone diameter (cm)**			**Stone weight (g)**		
Treatments	2012	2013	2014	2012	2013	2014	2012	2013	2014
1- Control (compost 100 %)	1.60C	1.63BC	1.65AB	0.833D	0.843CD	0.823E	0.842B	0.841C	0.841BC
2- *Comp. 75 % + **Pom. 25 %	1.65AB	1.64A-C	1.63BC	0.917BC	0.887BC	0.867CD	0.874A	0.876A	0.876A
3- Comp. 50 % + Pom. 50 %	1.62BC	1.62C	1.62CD	0.963AB	0.907AB	0.950A	0.844B	0.848BC	0.854BC
4- Comp. 75 % + Pom. 25 % + ***Tri.	1.60C	1.63BC	1.60E	0.967AB	0.903AB	0.850DE	0.877A	0.880A	0.878A
5- Comp. 75 % + Pom. 25 % + ****Pseu.	1.62BC	1.66AB	1.64A-C	0.950AB	0.880B-D	0.900BC	0.841B	0.843C	0.843BC
6- Comp. 75 + Pom. 25 % + Tri. + Pseu	1.63A-C	1.68A	1.63BC	0.873CD	0.837D	0.917AB	0.880A	0.882A	0.881A
7- Comp. 50 % + Pom. 50 % + Tri.	1.63A-C	1.65A-C	1.61DE	0.960AB	0.890B	0.883B-D	0.867A	0.863AB	0.838C
8- Comp. 50 % + Pom. 50 % + Pseu.	1.60C	1.62C	1.65AB	0.950AB	0.897AB	0.943A	0.830B	0.836C	0.844BC
9- Comp. 50 % + Pom. 50 % + Tri + Pseu.	1.66A	1.67A	1.68A	1.000A	0.937A	0.890BC	0.867A	0.867AB	0.862AB

Means with the same letter (s) within the same column are not significantly differ at the probability of 5 % level according to Duncan`s Multiple Range test.

* Comp: compost ** Pom.: Pomace

*** Tri.: *Trichoderma virdi* **** Pseu.: *Pseudomonas fluorescens*

4.8. The effect on flesh weight, flesh/fruit and flesh/stone ratio

The data presented in Table (11) showed that Comp. 50%+Pom.50% +Tri.+ Pseu./tree gave the heaviest flesh weight and flesh/fruit (3.75,3.81 & 3.75 and 81.13&81.47and81.24) during three seasons. As for flesh/stone ratio, Comp.50%+pom.50%+Pseu., and Comp. 50%+Pom.50% +Tri. + Pseu. /tree gave higher recorded (4.32&4.40) in the first season and second seasons. While in the third one the superior values (4.35) were recorded with the application of Comp. 50%+Pom.50% +Tri. /tree.

Table 11 displays data indicating that the use of Comp. 50% + Pom. 50% + Tri. + Pseu./tree resulted in the heaviest flesh weight and flesh/fruit ratios during all three seasons, with values of 3.75, 3.81, and 3.75 and 81.13, 81.47, and 81.24, respectively. Regarding the flesh/stone ratio, Comp. 50% + Pom. 50% + Pseu., and Comp. 50% + Pom. 50% + Tri. + Pseu./tree produced higher ratios of 4.32 and 4.40 in the first and second seasons, respectively. In the third season, the application of Comp. 50% + Pom. 50% + Tri./tree resulted in the superior ratio of 4.35. These results indicate that the application of Comp. 50% + Pom. 50% + Tri. + Pseu./tree may have a positive impact on the quality of the fruit, specifically in terms of flesh weight and flesh/fruit ratios, while the flesh/stone ratio may be improved by the application of Comp. 50% + Pom. 50% + Pseu., and Comp. 50% + Pom. 50% + Tri. + Pseu./tree.

Table (11): Effect of compost and olive pomace wastes with / without microorganisms treatments on flesh weight (g), flesh/fruit weight (%) and flesh/stone ratio of Manzanillo olive cultivar during 2012, 2013 and 2014 seasons.

Parameters / Treatments	Flesh weight (g)			Flesh/fruit (%)			Flesh/stone ratio		
	2012	2013	2014	2012	2013	2014	2012	2013	2014
1- Control (compost 100 %)	2.94 E	3.28 D	2.95F	77.63 D	79.59 C	77.74 CD	3.49 CD	3.90 BC	3.51 CD
2- *Comp. 75 % + **Pom. 25 %	2.77F	3.31 D	2.86 G	75.97 E	79.09 C	76.55 E	3.17 D	3.78 C	3.27 D
3- Comp. 50 % + Pom. 50 %	3.16 C	3.51 C	3.11 CD	78.89 C	80.56 B	78.42 C	3.74 BC	4.14 AB	3.64 C
4- Comp. 75 % + Pom. 25 % + ***Tri.	3.16 C	3.60 B	3.16 C	78.25 CD	80.35 B	78.22 C	3.60 C	4.09 AB	3.59 CD
5- Comp. 75 % + Pom. 25 % + ****Pseu.	2.99 DE	3.17 E	2.97 EF	78.07 D	78.99 CD	77.86 CD	3.56 C	3.76 C	3.52 CD
6- Comp. 75 + Pom. 25 % + Tri. + Pseu	3.04 D	3.19 E	3.04 DE	77.48 D	78.34 D	77.45 D	3.45 CD	3.62 C	3.44 CD
7- Comp. 50 % + Pom. 50 % + Tri.	3.49 B	3.53 BC	3.71 A	80.07 B	80.38 B	81.54 A	4.03 AB	4.10 AB	4.43 A
8- Comp. 50 % + Pom. 50 % + Pseu.	3.46 B	3.53 BC	3.45 B	80.63 AB	80.85 AB	80.31 B	4.17 A	4.23 A	4.08 B
9- Comp. 50 % + Pom. 50 % + Tri + Pseu.	3.75 A	3.81 A	3.75 A	81.13 A	81.47 A	81.24 A	4.32 A	4.40 A	4.35 AB

Means with the same letter (s) within the same column are not significantly differ at the probability of 5 % level according to Duncan`s Multiple Range test.
* Comp: compost ** Pom.: Pomace
*** Tri.: *Trichoderma virdi* **** Pseu.: *Pseudomonas fluorescens*

5. Conclusion

Adding microorganisms to organic materials such as olive pomace and compost has been shown to enhance crop productivity and quality. Based on the results, it is recommended to use Compost. 50% + pomace. 50% + *Pseudomonas fluorescence*, as it produced the best outcomes in terms of enhancing the productivity of Manzanillo olive trees. Therefore, this approach may be a suitable option for farmers and growers seeking to improve the quality and yield of their crops.

Conflicts of Interest/ Competing interest

All authors declare that they have no conflicts of interest.

Data availability statement

All data sets collected and analyzed during the current study are available from the corresponding author on reasonable request.

List of Abbreviations

ANOVA Analysis of variance
ARC Agricultural Research Center
HRI Horticulture Research Institute
OM Organic matter
OMWW Olive mill wastewater
SWERI Soils, Water and Environment Research Institute
A.O.A.C Association of Official Agricultural Chemists (A.O.A.C.)
D.W Dry weight
FIFA El-FIFA farm which is located at Km 48 of Cairo Alexandria Road in the north of Egypt.
IAA Indole acetic acid
OMW Olive mill waste
OWW Olive waste water

6- REFERENCE

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