

## Research Article

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## Detoxification of Olive-mill Solid Waste and its Probable Application as Organic Fertilizer

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### Abstract

Despite the phytotoxicity of olive-mill solid waste (OMSW) due to its high polyphenols content, OMSW have fertilizer characteristics, which make it a potential source for organic fertilization. Composting of OMSW treatment process was conducted in this study to eliminate the phytotoxicity and solve the environmental impact of this waste. Recycling of OMSW was carried out via composting of six batches of trials using equal proportions of OMSW, cow manure (C) and wheat straw (W). The treatment process was performed at two time intervals (two and five months), after each one, the recipient species (*Vicia faba* L.) was planted. The results showed the efficiency of composting in reducing OMSW original toxicity after two months than five months. The germination percentage and the plumule and radicle lengths of *V. faba* showed a significant improvement when the OMSW was composted with C at different proportions before using as soil amendments. Besides, the total biomass was noticeably increased at the high concentration of C-OMSW. Similarly, the total pigments concentration in *V. faba* was increased by using various composts after two months, where the highest pigment content was observed at 40% W-OMSW treatment.

The maximum uptake of potassium and sodium was recorded through the application of W-OMSW compost to soil after two months. Furthermore, the C-OMSW composts showed the highest concentration of nitrogen, calcium, iron and manganese. However, the C-W-OMSW composts recorded the highest concentration of phosphorous, magnesium and copper. Finally, this study developed a low cost treatment that will enable the growers to convert OMSW into a natural nontoxic compost rich with essential nutrients which have positive effects on plants growth.

**Keywords:** Olive-mill solid waste; *Vicia faba*; Biomass; Pigment; Nutrient uptake

### Introduction

Extraction of olive oil is a common industry in the Mediterranean region, producing a large amount of olive mill wastes in short periods. The traditional systems of olive oil extraction generate beside the olive oil (20%), two wastes: solid (30%) and aqueous wastes (50%) [1]. The two byproducts represent a serious environmental problem in Mediterranean countries where most of the world's olive oil is produced [2].

Olive-mill solid wastes (OMSW) contain high levels of organic matter and mineral nutrients mainly nitrogen, phosphorous, potassium and magnesium. The organic fraction of the OMSW contains phenolic substances, some nitrogenous compounds, organic acids, sugars, tannins, pectins, carotenoids, and almost all of the water-soluble ingredients of the olives. This make OMSW an important resource for beneficial application in various fields such as sorbent material for heavy metals [3,4], drinking water treatment [5,6]. Besides, its utilization in preparation of activated carbon [7], feeding animals, fuel and charcoal industry [8,9]. It is used as organic fertilizer in degraded agricultural soils after a composting operation or as fuel in a cogeneration system. Thus application of OMSW in agricultural practice in Mediterranean agro ecosystems representing a topic of great interest [10-13].

Various management technologies have been applied for treating olive-mills wastes. Among these are the physical processes, thermal processes, physico-chemical processes and biological processes. Biological processes utilize the microorganisms to decompose the biodegradable chemical compounds in OMSW. Composting is one of the main technologies for recycling OMSW and converting it into a fertilizer [14,15]. From both ecological and economical points of view,

detoxification of OMSW using composting has several advantages over the other treatment ways.

The current study was conducted to evaluate the possibility of biological detoxification of OMSW and its potential use as a soil conditioner and amendment agent. To achieve this aim, cow manure and wheat straw were used as biological materials to compost OMSW. Both of them act as a catalyst to eliminate the toxicity of OMSW at different time intervals. As well, the most suitable time to the beginning of agriculture after the fermentation of both cow manure (C) and wheat straw (W) with OMSW was estimated.

### Materials and Methods

#### Plant materials

Olive-mill solid waste (OMSW) was obtained from three traditional press olive-mills in El-Dabaa region (180 km west of Alexandria). As well, cow manure and wheat straw were purchased from number of cattle farms at El-Behira governorate (50 km south of Alexandria, Egypt). *V. faba* L. (broad bean) as a test or recipient species was obtained from the National Research Center at Dokki, Giza.

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## Analysis of soil

Soil samples from the natural sites where the alleged materials (OMSW) are not deposited were used to execute the growth experiment. The samples were analyzed for some of their chemical and physical properties according to Allen [16].

## Analysis of olive-mill solid waste

Three composite samples of OMSW were ground in Wiley Mill to pass 1.0 mm mesh. One part was used for the determination of the concentration of N, P, K, Na, Ca, Mg, Fe, Mn, Cu, Zn and of the total ash, total phenols and lipids, while the other parts were used for the determination of organic matter, electric conductivity, pH and moisture content. All these procedures were according to Allen [16].

## Composting of olive-mill waste

Six piles were prepared by mixing OMSW with cow manure (C) and wheat straw (W) in two groups for testing composting at two time intervals (two and five months). Two Kgs of six treatments (C: W: OMSW; C: OMSW; W: OMSW; OMSW; C and W) at the same ratios on dry weight base with anaerobic- aerobic conditions and occasional stirring were prepared. The treatments were incubated under normal laboratory conditions with day temperature ranging from 19-22°C and night temperature from 12-14°C. After that, all treatments were moisturized by 2 liters of water one time in two months treatments and twice in five months treatments.

## Growth experiment

Growth experiment was performed to test the potential effects of various prepared composts on germination efficiency, growth rate, total dry weight, nutrient content and uptake of the recipient species in sandy clay loam soil under 80% field capacity water regime.

## Implantation

After two months, the 6 composts were air-dried and ground in Wiley Mill to pass 1.0 mm mesh. A sandy clay loam soil was mixed with each treatment of the six composts at different proportions (10, 20, 30 and 40%) in three replicates of each proportion for each tested time interval. One treatment was run as control contains untreated soil. After mixing, the amount of soil with various composts needed was transferred to 20-cm diameter, 2-L plastic pots. Ten seeds of *V. faba* were planted in each pot during March and July. The experiment was carried out under normal laboratory conditions with day temperature ranging from 19-22°C and night temperature from 12-14°C. The plants were watered every two days on the average with normal tap water. The amount of water loss corresponding to average soil-plant- evapotranspiration was calculated over 24-hours for three replicates. Seedlings of *V. faba* were harvested after two weeks from planting.

## Germination efficiency and dry matter accumulation

A certain number of morphologically homogenous *V. faba* seedlings from each treatment were excavated and washed carefully by running water to remove adhering soil particles, then by distilled water. Germination percentage (GP), plumule (PL) and radicle (RL) lengths were recorded. The plant materials were dried at 70°C till constant weight to determine the total dry weight.

Germination percentage (GP) was calculated for each treatment at different proportions according to the general equation:

Germination percentage=(treatment/total number of germinated seeds)×100

Where: Treatment: Number of germinated seeds for each treatment.

## Pigment content

The photosynthetic pigments chlorophyll a (Chl a), b (Chl b) and carotenoids (Carot) were extracted and determined using the spectrophotometric method described by Metzner [17]. The values are expressed as (mg g<sup>-1</sup> fresh wt.).

## Nutrient content and uptake

The Oven-dried plant samples were ground in a Wiley Mill to pass 1.0 mm screen. These samples were chemically analyzed to determine their contents of macronutrient elements (N, P, K, Na, Ca, and Mg) and micronutrient elements (Fe, Mn, and Cu) according to Allen [16]. Concentrations of all nutrients were expressed on a dry weight basis and the total nutrient uptake at each treatment was calculated according to the following relation:

$$\text{Nutrient uptake} = \frac{(\text{Nutrient concentration} \times \text{Total dry weight})}{\text{Number of days}}$$

The values of macronutrient elements expressed as (mg day<sup>-1</sup>).

The values of micronutrient elements expressed as (µg day<sup>-1</sup>).

## Statistical analysis of data

All chemical and physical analyses were performed thrice. Data concerning the effect of different concentrations of all treatments on measured parameters of the recipient species were subjected to standard analysis of variance (ANOVA) and t-test using the COSTAT 2.00 statistical analysis software manufactured by Co-Hort Software Company. Differences between treatment means were considered statistically significant at P < 0.05.

## Results

### Soil analysis

The routine analyses for soil applied in the current study are presented in Table 1. In general, the results show that the applied soil was a sandy clay loam soil. The soil texture was represented by 24% clay, 58% sand, 18% silt. Hence, this type of soil comprises considerable

Parameter	Range
<b>Physical properties</b>	
Texture	Sandy clay loam
Clay <sup>b</sup>	24.00-26.90
Sand <sup>b</sup>	58.00-61.28
Silt <sup>b</sup>	18.00-19.23
<b>Chemical properties</b>	
Electrical conductivity <sup>a</sup>	2.72-2.82
Organic matter <sup>b</sup>	9.08-10.00
pH	7.82-9.01
Free carbon <sup>b</sup>	1.50-1.53
N <sup>c</sup>	1.100-1.104
P <sup>c</sup>	0.520-0.527
K <sup>c</sup>	3.50-3.98
Ca <sup>c</sup>	15.40-17.34
Mg <sup>c</sup>	1.40-1.42
Cl <sup>c</sup>	15.25-16.45
CO <sub>3</sub> <sup>c</sup>	35.00-37.21
SO <sub>4</sub> <sup>c</sup>	18.70-19.38

**Table 1:** Variation in some physical and chemical characteristics of the soil used for the pot experiment Considering that, a: dsm<sup>-1</sup>, b: % and c: mg g<sup>-1</sup>.

content of organic matter (10%). The applied soil presents a slightly basic or alkaline pH (7.8). The electrical conductivity (EC) which indicates the total soluble salts in the soil was recorded 2.72 dS m<sup>-1</sup>. Concerning, the essential elements the nitrogen, phosphorous, potassium contents were 1.1, 0.5 and 3.5 mg g<sup>-1</sup>, respectively.

### Main physical and chemical characteristics of olive-mill solid waste (OMSW)

The main physical and chemical characteristics of OaMSW applied in this study are presented in Table 2. It is essential to emphasize that the applied OMSW is very rich in organic matter 73.24% comprising relatively large amount of lignin, cellulose and hemicellulose, lipids, and carbohydrates as well as phenolics. The applied OMSW contains 4.66% phenolic compounds (total phenols). The electrical conductivity (EC) which indicates the total soluble salts in the OMSW was recorded 6 dS m<sup>-1</sup>. The applied OMSW presents a moisture content about 66.2% and an acidic matter (pH: 5.40). OMSW possesses considerable amounts of mineral nutrients such as nitrogen (12.50 mg g<sup>-1</sup>), phosphorous (1.30 mg g<sup>-1</sup>), potassium (21.14 mg g<sup>-1</sup>), sodium (1.95 mg g<sup>-1</sup>), calcium (10.19 mg g<sup>-1</sup>), and magnesium (1.20 mg g<sup>-1</sup>) and a wide range of micronutrients such as iron (815 µg g<sup>-1</sup>), copper (15.0 µg g<sup>-1</sup>), manganese (42.0 µg g<sup>-1</sup>) and zinc (16.0 µg g<sup>-1</sup>).

## Germination and Growth Efficiency

### Germination percentage (GP)

Figures 1 and 2 showed the results concerned with the effect of various composts on the germination percentage of *V. faba* seeds at two composting time intervals (two and five months). These results showed that, the germination percentage of *V. faba* in control was 100% at two and five months.

After two months of incubation time, the germination percentage in OMSW compost treatment showed gradual decrease upon increasing the OMSW proportion (Figure 1). The lowest germination percentage was observed as 50% at 40% OMSW compared to control. Similar trend was observed after five months where the germination percentage was also decreasing upon increasing proportion of OMSW. The lowest germination percentage was observed as 10 and 20% at 30%

and 40% OMSW, respectively compared to control. By comparing the two incubation time, the germination percentage decreased significantly ( $p < 0.5$ ) from the second incubation time (5 months) to the first incubation time (2 months).

The germination percentage in wheat straw (W) compost treatment decreased to 80% compared to control and no seed germination showed at 40% W for the first incubation period. The longer incubation time to five months presented significant decrease in the germination percentage compared to two months. Where, the germination percentage decreased to 30% at 20% W treatment compared to control and no seed germination noticed at 30% and 40% W for the second incubation period.

When the cow manure (C) compost was used, the germination percentage was 100% almost similar to control after two months except for the treatment proportion of 40%. While, after five months the germination percentage decreased by 10% to 30% compared to control at 40 and 10% treatment, respectively then gradually increased. The two incubation periods gave the best values for seed germination compared to other treatments.

Using of wheat straw-olive-mill solid waste (W-OMSW) compost decreased the germination percentage gradually upon increasing W-OMSW proportion after two months. The lowest germination percentage was observed as 50% at 40% W-OMSW compared to control. Also, the second incubation time presented gradual decrease in the germination percentage. Where, the germination percentage was decreased upon increasing the proportion of W-OMSW by 20%-30% compared to control at 10, 20 and 30% treatment, respectively. No seed germination was observed at 40% proportion level.

After the two months of incubation, the germination percentage in cow manure-olive-mill solid waste (C-OMSW) compost with its different proportions achieved close values to control. On the other hand, the germination percentage was reduced by 40%- 70% compared to control at 10%, 20% and 40% proportion after the five months. No seed germination was observed at 30% proportion. The longer the compost incubation time, the more decrease in the germination percentage.

The cow manure-wheat straw-olive-mill solid waste (C-W-OMSW) compost slightly decreased the germination percentage by 10%-20% upon increasing the proportion of C-W-OMSW compared to control after the first incubation time. Whereas, the second incubation time exhibited a gradual decrease in the germination percentage through different proportions of C-W-OMSW compared to control. At 40% C-W-OMSW, the germination percentage decreased significantly to 10% compared to control. Thus, the second incubation time offered an increase in the germination percentage compared to first incubation time.

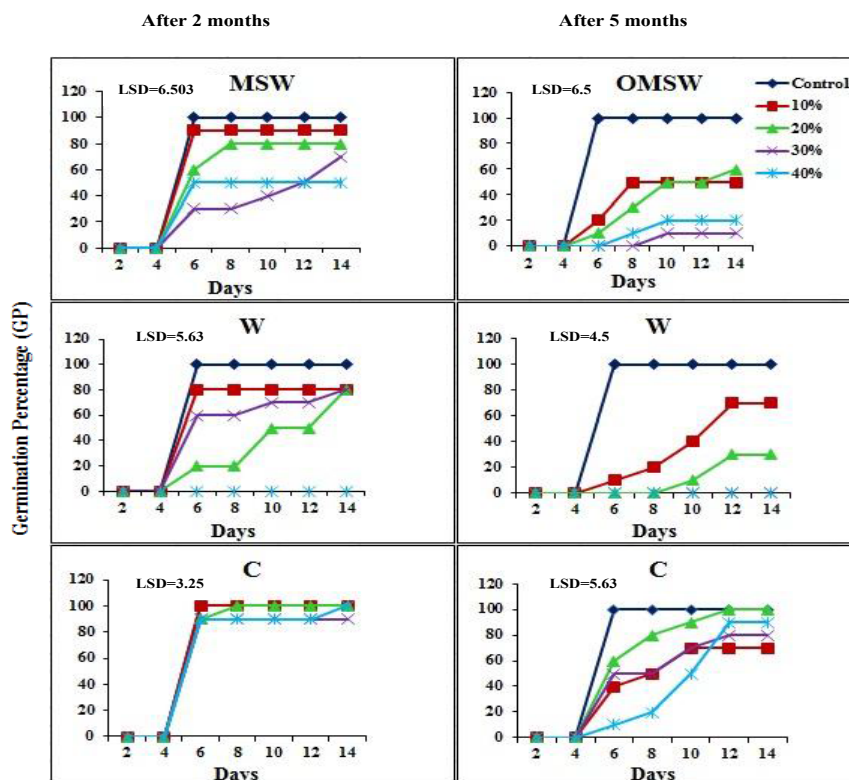
### Plumule (PL) and radicle (RL) lengths

The effects of different composts on plumule and radicle lengths of *V. faba* at two time intervals are illustrated in Figures 3 and 4. The plumule and radicle lengths of *V. faba* were reduced in control from 25.46, 12.5 cm at two months to 13.91, 10.4 cm at five months, respectively.

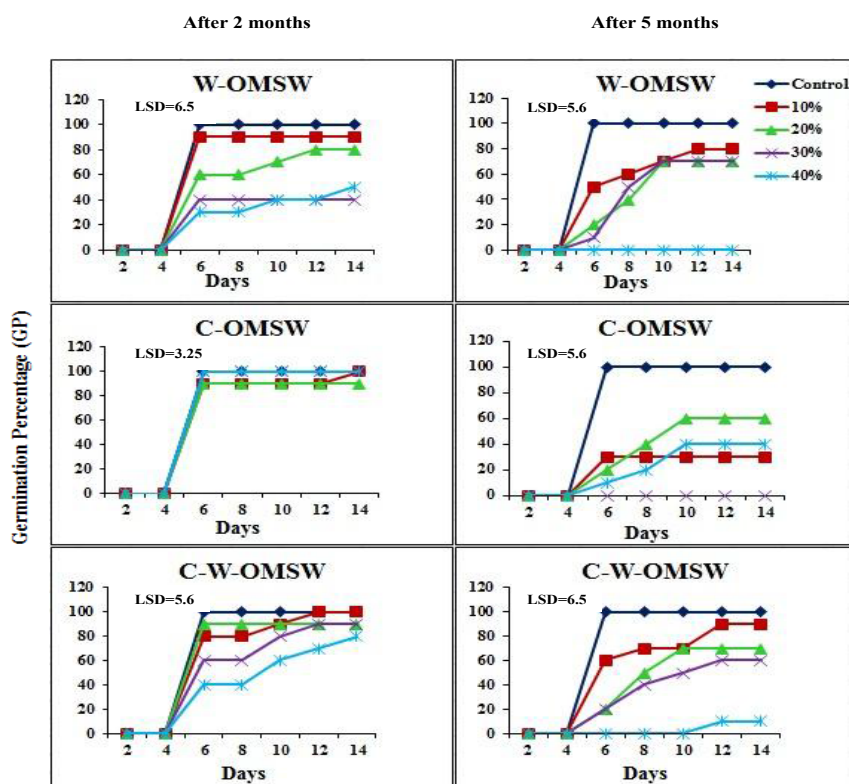
Generally, in OMSW compost the plumule length was reduced at all proportion levels and the radicle length was decreased at most of proportion levels at two months (Figure 3). The results demonstrated that the plumule lengths decreased to 24.8, 25.15, 8.63 and 12.1 cm at

Parameter	Range
Dry matter	33.5-34.3
Moisture <sup>b</sup>	66.2-69.1
Organic matter <sup>b</sup>	73.2-75.9
Electrical conductivity <sup>a</sup>	5.83-6.31
PH	5.40-5.71
Lipid <sup>b</sup>	23.4-24.2
Total phenols <sup>b</sup>	4.66-5.71
Nutrients elements	
N <sup>c</sup>	12.50-13.90
P <sup>c</sup>	1.30-1.70
K <sup>c</sup>	21.14-22.92
Na <sup>c</sup>	1.95-2.08
Ca <sup>c</sup>	10.19-11.41
Mg <sup>c</sup>	1.20-1.32
Fe <sup>d</sup>	815-893
Cu <sup>d</sup>	15.0-15.10
Mn <sup>d</sup>	42.0-43.23
Zn <sup>d</sup>	16.0-16.40

**Table 2:** The main physical and chemical characteristics of olive-mill solid wastes (OMSW) collected at year-2011 harvest season from three olive-mills at Dabaa area-western coastal region of Egypt. Considering that, a: dsm<sup>-1</sup>, b: %, c: mg g<sup>-1</sup> and d: µg g<sup>-1</sup>.

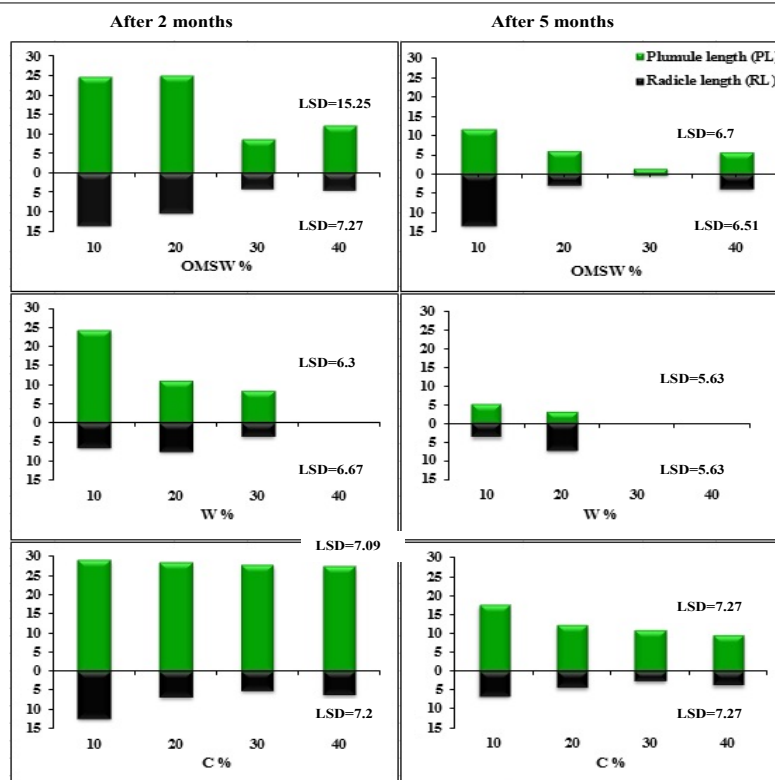


**Figure 1:** Effect of olive-mill solid waste (OMSW), wheat straw (W) and cow manure (C) composts at two time intervals on germination percentage of *Vicia faba* L.

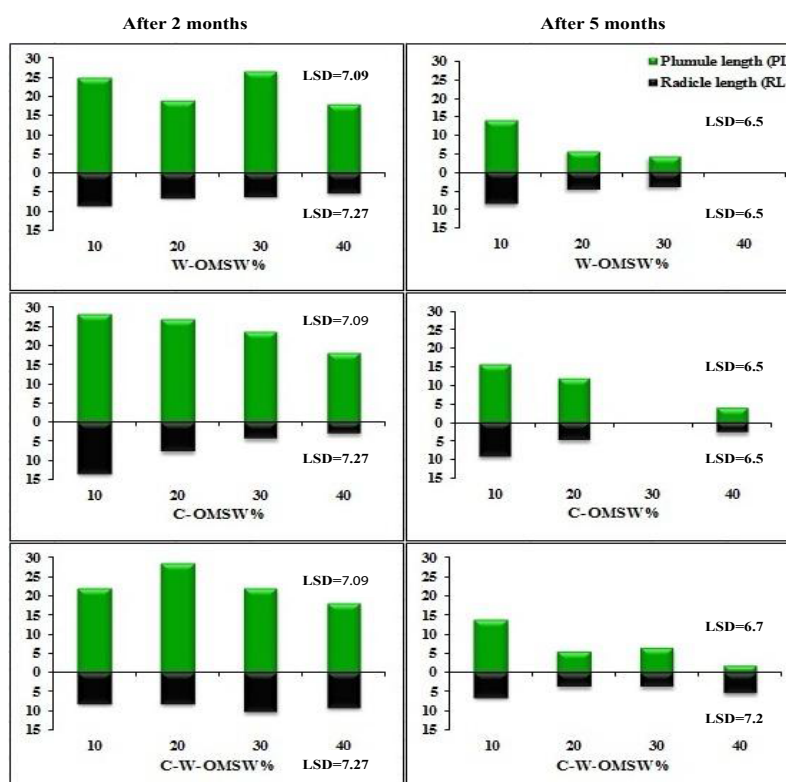


**Figure 2:** Effect of wheat straw-olive-mill solid waste (W-OMSW), cow manure-olive-mill solid waste (C-OMSW) and cow manure-wheat straw-olive-mill solid waste (C-W-OMSW) composts at two time intervals on germination percentage of *Vicia faba* L.





**Figure 3:** Effect of olive-mill solid waste (OMSW), wheat straw (W) and cow manure (C) composts at two time intervals on Plumule and Radicle length of *Vicia faba* L. Considering the control values 25.46 and 12.1 cm at two months and 13.91 and 10.5 at five months, respectively.



**Figure 4:** Effect of wheat straw-olive-mill solid waste (W-OMSW), cow manure olive-mill solid waste (C-OMSW) and cow manure-wheat straw-olive-mill solid waste (C-W-OMSW) composts at two time intervals on Plumule and Radicle length of *Vicia faba* L. Considering the control values 25.46 and 12.1 cm at two months and 13.91 and 10.5 at five months, respectively.

10%, 20%, 30%, and 40% OMSW proportions, respectively. In addition, the radicle lengths increased to 13.22 cm at 10% OMSW then decreased to 10.18, 3.9 and 4.2 cm at 20%, 30%, and 40% OMSW, respectively for the first time interval. After composting for five months, the plumule lengths in OMSW treatment decreased to 11.6, 6.16, 1.5, and 5.75 cm at 10%, 20%, 30%, and 40% proportion, respectively. Besides, the radicle length increased to 13.5 cm at 10% OMSW then decreased to 3.16, 0.3 and 4 cm at 20%, 30%, and 40% OMSW, respectively compared to control value.

In W compost the plumule and radicle lengths of *V. faba* were reduced at the different proportions after composting for two months. The plumule and radicle lengths were 24.21, 10.9, 8.45 cm and 6.56, 7.54, 3.7 cm at 10%, 20% and 30% W, respectively. For the second time interval, the plumule and radicle lengths in W treatment were decreased to 5.07, 3 cm and 3.78, 7.33 cm at 10%, 20% W, respectively.

The plumule length of *V. faba* grown in C compost increased to 29.07, 28.44, 27.67 and 27.25 cm at 10%, 20%, 30%, and 40% C proportions, respectively. In addition, the radicle length increased to 12.56 at 10% C then decreased to 7, 5.28 and 6.45 cm at 20%, 30%, and 40% C after the first time interval, respectively. Whereas, after the second time interval, the plumule length was increased to 17.42 cm at 10% C then decreased to 12.1, 10.75, 9.5 cm at 20%, 30%, 40% C, respectively. Besides, the radicle length was decreased to 6.64, 4.42, 2.75, 3.87 cm at 10%, 20%, 30%, 40% C, respectively.

The plumule length of *V. faba* increased by 30% W-OMSW amendment to 26.5 cm after composting for two months. Conversely, it was decreased to 24.9, 18.88, 17.8 cm at 10%, 20%, 40% W-OMSW, respectively. In addition, the radicle length decreased to 8.5, 6.5, 6.25 and 5.4 cm at 10%, 20%, 30%, and 40% W-OMSW composts, respectively. The effect of longer incubation time (5 months) on the plumule length showed an increase to 14.25 cm at 10% W-OMSW then a decrease to 5.88, 4.21 cm at 20%, and 30% W-OMSW, respectively. There was a direct correlation between the increase of W-OMSW proportion and the radicle length. Since the radicle length was decreased to 8.56, 4.64, 4 cm upon increasing the proportion from 10% to 20% to 30% W-OMSW, respectively.

The C-OMSW compost increased the plumule length of *V. faba* to 28.45 and 27.11 cm at 10% and 20% C-OMSW then decreased to 23.65, 18.3 cm at 30%, 40% C-OMSW, respectively. In addition, the radicle length increased to 13.45 cm at 10% C-OMSW then decreased to 7.77, 4.45, 3.1 cm at 20%, 30%, 40% C-OMSW, respectively after the first time interval. An increase in the plumule length to 15.56 cm at 10% C-OMSW then a decrease to 11.92, 4 cm at 20%, 40% C-OMSW, respectively were observed after 5 months. Moreover, the radicle length decreased to 9.16, 4.83, 2.75 cm at 10%, 20%, 40% C-OMSW, respectively.

The C-W-OMSW compost increased the plumule length of *V. faba* to 28.83 cm at 20% C-W-OMSW. On the contrary, it decreased the plumule length to 22.3, 22, 18.12 cm at 10%, 30%, 40% C-W-OMSW, respectively. In addition, the radicle length increased to 8.15, 8.17, 10.4, 9.18 cm at 10% 20%, 30%, 40% C-W-OMSW after composting for two months, respectively. The decrease in plumule length by C-W-OMSW application after composting for five months were 13.5, 5.32, 6.33, 1.8 cm at 10% 20%, 40% C-W-OMSW, respectively. Similarly, the radicle length was decreased to 6.5, 3.77, 3.83, 5.5 cm at 10% 20%, 40% C-W-OMSW, respectively.

## Total dry weight

The effect of different composts on the total dry weight (g) of *V. faba* is represented in Figures 5 and 6. Generally speaking, the application of different composts after composting for two months was showed slight decrease in the total dry weight of *V. faba*. After the first incubation time, the total dry weight showed a significant increase from 2.61 g in control to 5.4 and 5.9 g at 10% C-W-OMSW and 40% C-OMSW proportion, respectively. On the other hand, the total dry weight showed a significant decrease at 40% OMSW and 30% C, 10% W-OMSW with values range from 1.4 to 1.5 g. whereas, the other different composts didn't showed any significant variation in their values. The application of different composts after composting for five months was showed increasing in the total dry weight of *V. faba* at most of composts. Maximum total dry weight was observed at 30% W-OMSW proportion as 3.37 g compared to control of 1.41 g. The minimum total dry weight of *V. faba* was observed at 30% OMSW as 0.44 g. No seed germination was observed at 40% W after composting for two months. Similarly, 30%, 40%W, 40%W-OMSW and 30% C-OMSW after composting for five have not showed seed germination.

## Pigment fractions

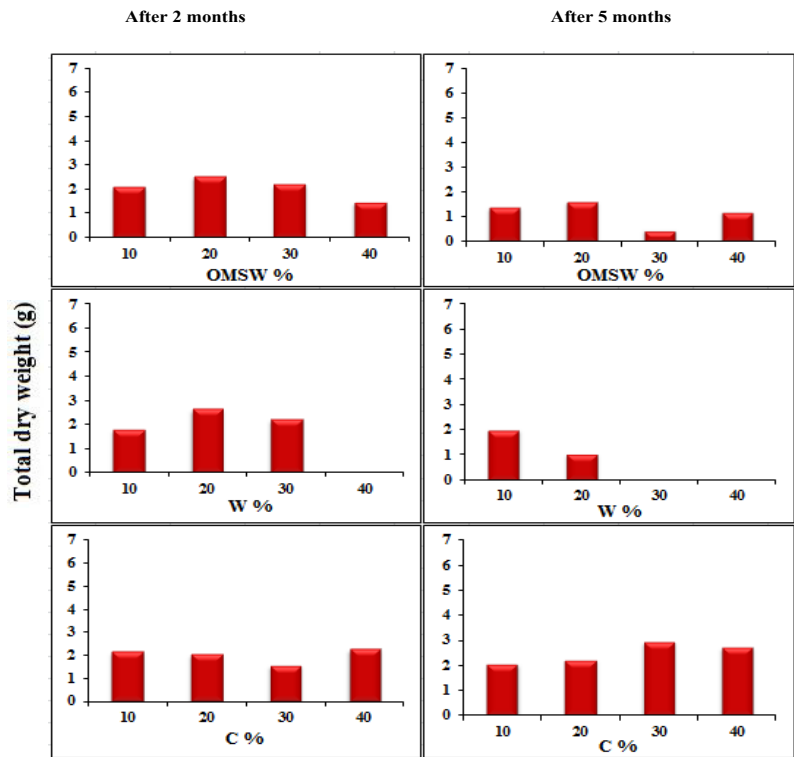
The effect of different composts on total pigment fractions concentrations in *V. faba* at two time intervals is illustrated in Figures 7 and 8. The total pigment fractions concentrations in *V. faba* decreased in control samples from 5.663 to 3.821 mg g<sup>-1</sup> fresh wt. after composting for two and five months, respectively.

Application of 10% OMSW compost reduced the concentration of total pigments in *V. faba* to 3.615 mg g<sup>-1</sup> fresh wt. compared to control. Raising the OMSW compost concentration to 20%, 30% and 40% caused an increase in total pigment concentration to 6.178, 5.756, 6.107 mg g<sup>-1</sup> fresh wt., respectively after composting for two months. The total pigments concentration increased to 4.171, 4.083 mg g<sup>-1</sup> fresh wt. at 10%, 20% OMSW, respectively then decreased to 1.297 mg g<sup>-1</sup> fresh wt. at 40% OMSW for the second time interval.

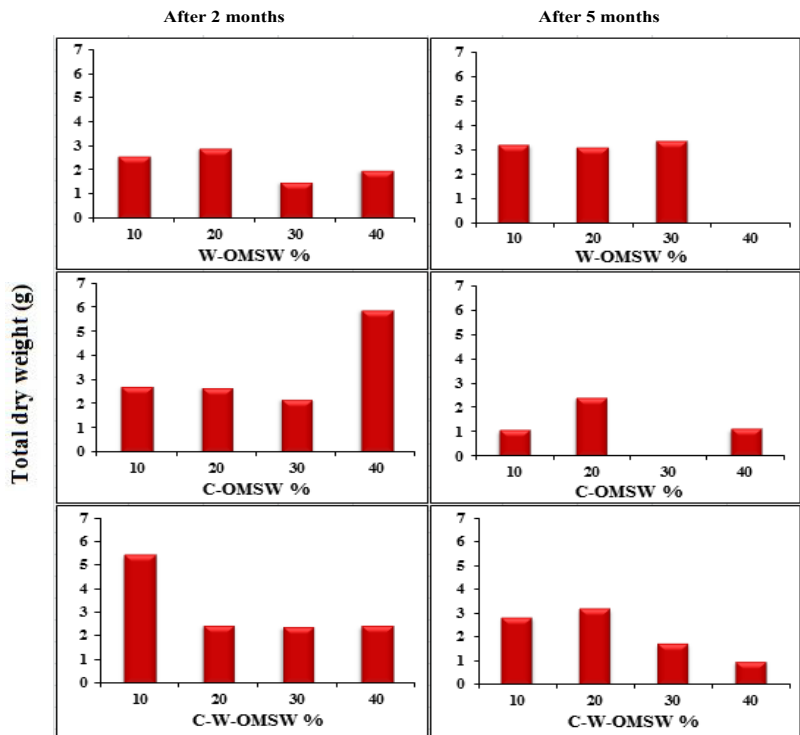
There was a decrease in total pigments concentration in *V. faba* to 4.820 mg g<sup>-1</sup> fresh wt. when 10% W compost was used compared to control. Then, it was increased to 5.631, 5.442 mg g<sup>-1</sup> fresh wt. at 20%, 30% W, respectively after the first time interval. After the second incubation time, the total pigments concentration increased to 4.568 mg g<sup>-1</sup> fresh wt. at 10% W then decreased to 3.156 mg g<sup>-1</sup> fresh wt. at 20% OMSW.

Using of C compost increased the total pigments concentration in *V. faba* to 6.389 mg g<sup>-1</sup> fresh wt. at 10% C then it was decreased to 5.319 and 5.098 mg g<sup>-1</sup> fresh wt. at 20% and 30% C, respectively. The total pigments content improved to 5.982 mg g<sup>-1</sup> fresh wt. at 40% C after the first incubation time. After five months following different concentration of C application, the total pigments concentrations increased to 6.752, 6.334, 5.597 and 5.793 mg g<sup>-1</sup> fresh wt. at 10%, 20%, 30% and 40% C, respectively.

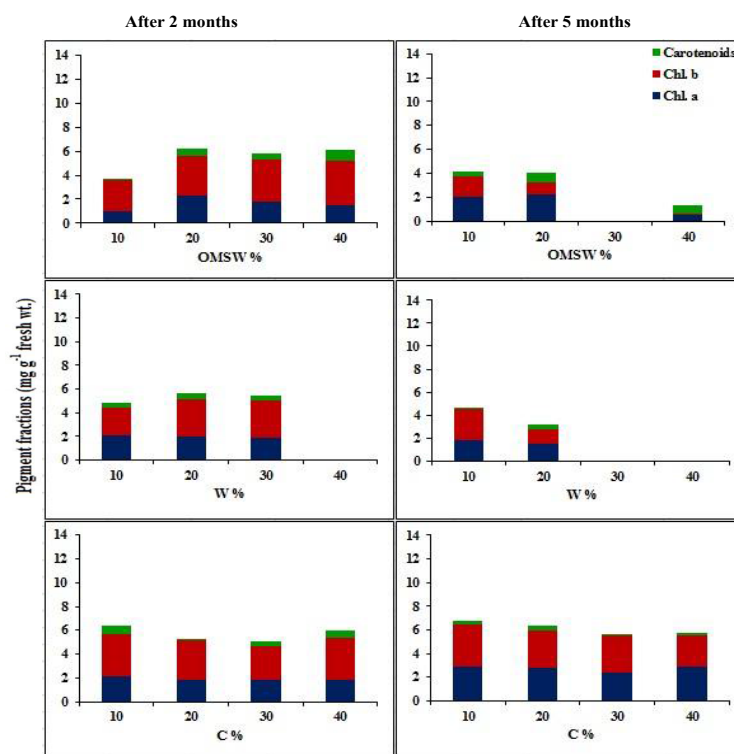
Generally, the application of W-OMSW, C-OMSW and C-W-OMSW composts at different proportions increased the total pigments concentration in *V. faba* after two incubation times compared to control. A gradual increase in the total pigments content was observed which is dependent on increasing W-OMSW proportions, where the pigments content increased from 6.211 to 13.087 and from 6.178 to 7.295 mg g<sup>-1</sup> fresh wt. after two and five months, respectively. The total pigments content increased from 7.024 to 8.646 and from 4.200 to 6.588 mg g<sup>-1</sup> fresh wt. due to application of C-OMSW after two and five



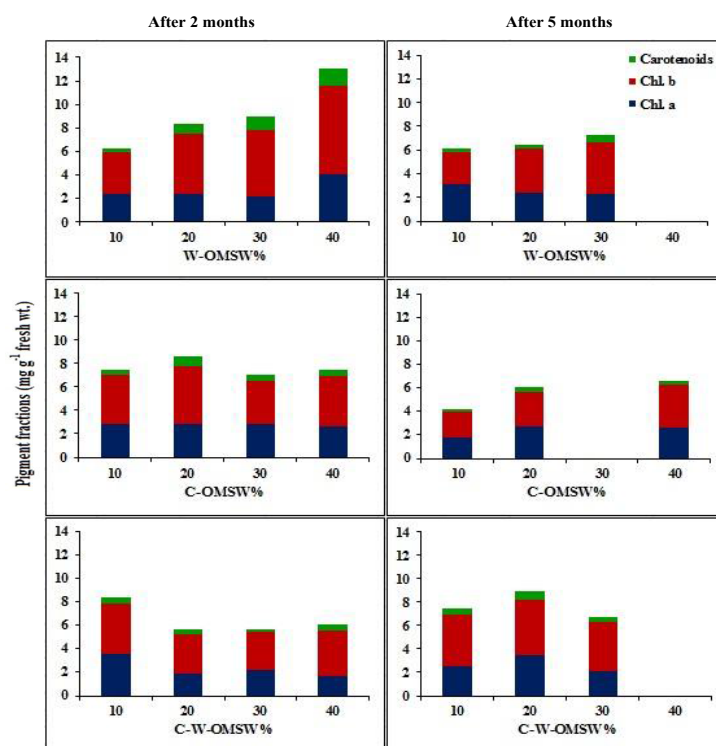
**Figure 5:** Effect of olive-mill solid waste (OMSW), wheat straw (W) and cow manure (C) composts at two time intervals (2 and 5 months) on the total dry weight of *Vicia faba* L. Considering the control values 2.61 g and 1.41 g at two and five months, respectively.



**Figure 6:** Effect of wheat straw-olive-mill solid waste (W-OMSW), cow manure olive-mill solid waste (C-OMSW) and cow manure-wheat straw-olive-mill solid waste (C-W-OMSW) composts at two time intervals on the total dry weight of *Vicia faba* L. Considering the control values 2.61 g and 1.41 g after two and five months, respectively.



**Figure 7:** Effect of olive-mill solid waste (OMSW), wheat straw (W) and cow manure (C) composts at two time intervals on pigment fractions concentrations (mg g<sup>-1</sup> fresh wt.) of *Vicia faba* L. Considering the control values 5.663 and 3.821 mg g<sup>-1</sup> fresh wt. at two and five months, respectively.



**Figure 8:** Effect of wheat straw-olive-mill solid waste (W-OMSW), cow manure olive-mill solid waste (C-OMSW) and cow manure-wheat straw-olive-mill solid waste (C-W-OMSW) composts at two time intervals on pigment fractions concentrations (mg g<sup>-1</sup> fresh wt.) of *Vicia faba* L. Considering the control values 5.663 and 3.821 mg g<sup>-1</sup> fresh wt. at two and five months, respectively.



months, respectively. In case of C-W-OMSW different amendments, the total pigments content increased from 5.676 to 8.419 and from 6.735 to 8.955 mg g<sup>-1</sup> fresh wt. after the first and two time intervals, respectively compared to control.

In a summary, after two month of incubation, the maximum total pigment concentration achieved at 30% and 40%W-OMSW treatments, while the minimum concentration was recorded at 10% OMSW compared to control. After longer incubation time of five months, total pigment concentration attained its highest value in 20% C-W-OMSW treatment, while the lowest concentration was obtained at 40% OMSW treatment.

### Nutrient uptake

Uptake levels of plant nutrient are expressed on a dry weight basis analyzed per day. Nutrients uptake for large quantities are reported in mg day<sup>-1</sup> where nutrients uptake for smaller quantities are reported in µg day<sup>-1</sup>.

**Macronutrients:** Effect of different composts on the uptake of some macronutrients mainly N, P, K, Na, Ca, and Mg in *V. faba* seedlings at two incubation time are presented in Figure 9. The nitrogen uptake was observed as 0.438 and 0.105 mg day<sup>-1</sup> for the control after the first and second incubation time, respectively. The maximum nitrogen uptake after two months of incubation was recorded as 0.425 mg day<sup>-1</sup> when the W compost used compared to control. The minimum uptake of nitrogen showed as 0.176 mg day<sup>-1</sup> with W-OMSW compost. After five months, the highest 0.062 mg day<sup>-1</sup> and the lowest 0.017 mg day<sup>-1</sup> were observed by adding C-OMSW and W composts, respectively. Generally, the nitrogen uptake similarly decreased through the application of different composts compared to control after the two incubation times. By comparing the two incubation times, nitrogen uptake achieved higher values after two months than after five months of incubation.

The control values of the phosphorous uptake were recorded as 0.145 and 0.002 mg day<sup>-1</sup> after two and five months of incubation, respectively. Uptake of phosphorous was ranged from 0.009 to 0.132 mg day<sup>-1</sup> by applying C-OMSW and W composts after the first incubation time. The phosphorous uptake increased to 0.008 mg day<sup>-1</sup> when the C compost used after the second incubation time. Similarly to control, the lowest phosphorous uptake was observed as 0.002 mg day<sup>-1</sup> via adding the C-OMSW compost. Generally speaking, the phosphorous uptake was observed in higher values after two months than after five months of incubation.

Potassium uptake of 0.07 and 0.41 mg day<sup>-1</sup> were recorded for control after two and five months, respectively. The element showed similar uptake value to control (0.07 mg day<sup>-1</sup>) when W-OMSW compost applied after two months. The uptake of the nutrient was decreased to 0.05 mg day<sup>-1</sup> by the application of C compost after two months. On the other hand, the uptake of potassium was ranged from 0.03 to 0.09 mg day<sup>-1</sup> via the application of different composts after five months of incubation.

Sodium uptake was observed as 0.188 and 1.112 mg day<sup>-1</sup> for control after two and five months of incubation period, respectively. The element uptake increased to 0.195 mg day<sup>-1</sup> by using W-OMSW compost compared to control after two months. The minimum uptake (0.135 mg day<sup>-1</sup>) achieved by adding C compost. Sodium uptake increased to 0.228 mg day<sup>-1</sup> by using C-OMSW compost compared to control after five months. The sodium uptake noticeably decreased to 0.076 mg day<sup>-1</sup> by adding OMSW compost.

Uptake of calcium was recorded as 0.352 and 0.138 mg day<sup>-1</sup> for control after first and second incubation time, respectively. The lowest (0.056 mg day<sup>-1</sup>) and the highest (0.202 mg day<sup>-1</sup>) values of calcium uptake were observed after the first time of incubation by applying OMSW and C composts, respectively. Uptake of this nutrient was increased to 0.17 mg day<sup>-1</sup> through the application of C compost after the second incubation time. The calcium uptake was considerably decreased to 0.03 mg day<sup>-1</sup> by adding W compost after five months of incubation.

The uptake values of magnesium for control were recorded as 0.014 and 0.007 mg day<sup>-1</sup> after the two and five months of incubation, respectively. After two months, the nutrient uptake through using different composts was approximately similar to control where the uptake was varied from 0.01 to 0.014 mg day<sup>-1</sup>. On the other hand, the element uptake with C-OMSW addition increased to 0.017 mg day<sup>-1</sup> compared to control after five months. The minimum Mg uptake 0.005 mg day<sup>-1</sup> was observed by adding OMSW compost after the second incubation time.

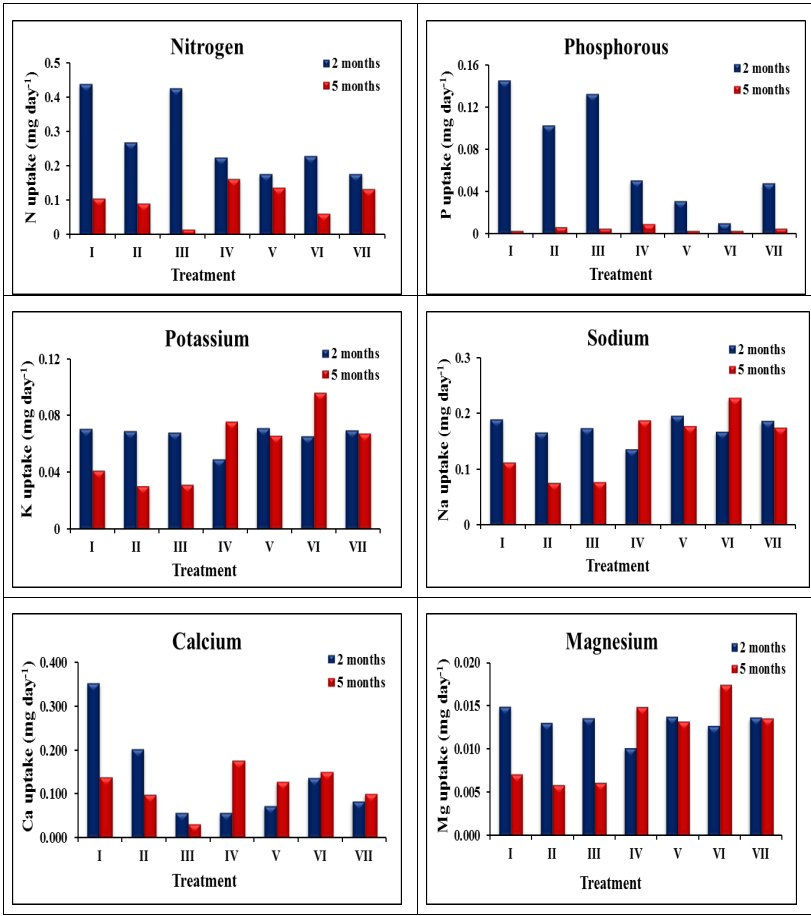
**Micronutrients:** Effect of different composts on the uptake of some micronutrients such as Fe, Mn, Cu in *V. faba* seedlings at two incubation time are illustrated in Figure 10. The uptake values of iron were recorded as 419.46 and 132.54 µg day<sup>-1</sup> for control after the first and second incubation time, respectively. The iron uptake was decreased in the range of 123.17 to 292.05 µg day<sup>-1</sup> compared to control through the addition of different composts after two months. On the other hand, an increase in the element uptake was observed at C treatment to 195.27 µg day<sup>-1</sup> compared to control after five months. The minimum uptake of iron was recorded at W treatment as 42.03 µg day<sup>-1</sup> after five months. By comparing the two incubation periods, the uptake of iron recorded higher values after two months than after five months at almost treatments.

The results indicated that, manganese uptake for control was 16.77 and 5.94 µg day<sup>-1</sup> after two and five months, respectively. Generally, this element uptake for various amendments was evidently decreased compared to control in the range of 4.38 to 11.47 µg day<sup>-1</sup> following two months of incubation. The five months experiment reported that, the values of Mn uptake were also decreased by using different composts in the range of 1.31 to 5.35 µg day<sup>-1</sup>. Also, the uptake of manganese was generally decreased compared to control of five months when the various composts were used. By comparing two incubation times, the first incubation time recorded higher values of Mn uptake than the second incubation time in different treatments.

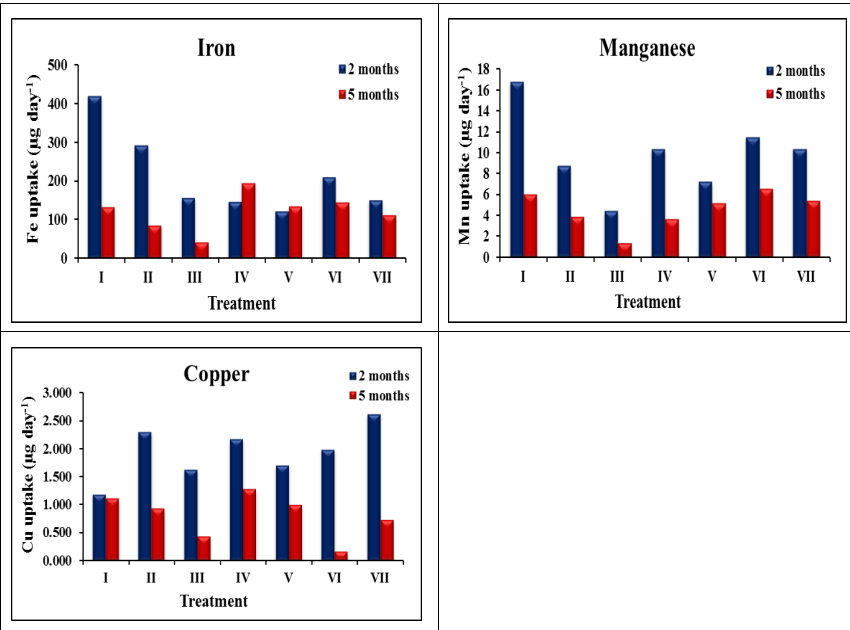
Copper uptake was observed as 1.176 and 1.098 µg day<sup>-1</sup> for control after two and five months of incubation period, respectively. By comparing to control of two months, the Copper uptake was increased by C-W-OMSW compost added to 2.621 µg day<sup>-1</sup>. In contrast, the lowest Copper uptake was observed as 1.629 µg day<sup>-1</sup> by adding W compost after two months of incubation. The copper uptake for C treatments was increased to 1.262 µg day<sup>-1</sup> compared to control of five months of incubation. The lowest Copper uptake after five months was noticeably observed as 0.155 µg day<sup>-1</sup> by adding C-OMSW compost. By comparing two incubation times, short incubation time recorded higher values of copper uptake than the longer incubation time when different composts used.

### Discussion

The annual OMW (olive-mill waste) production of Mediterranean olive-growing countries was estimated to ranging from 7 million



**Figure 9:** Effect of different composts at two time intervals on the uptake (mg day<sup>-1</sup>) of some macronutrient in *Vicia faba* L. I: Control, II: olive -mill solid waste, III: wheat straw, IV: cow manure, V: wheat straw-olive-mill solid waste, VI: cow manure olive-mill solid waste and VII: cow manure-wheat straw-olive-mill solid waste.



**Figure 10:** Effect of different composts at two time intervals on the uptake (µg day<sup>-1</sup>) of some micronutrient in *Vicia faba* L. I: Control, II: olive -mill solid waste, III: wheat straw, IV: cow manure, V: wheat straw-olive-mill solid waste, VI: cow manure olive-mill solid waste and VII: cow manure-wheat straw-olive-mill solid waste.

to over 30 million m<sup>3</sup>. OMSW (olive-mill solid waste) disposal from olive oil mills is a major environmental issue in several olive growing countries in the world. OMSW contains high toxic levels of organic matter concentration (94%) such as poly-phenols, giving it an elevated polluting load. The efforts to find a solution to the OMW problem are more than 50 years old [18].

Composting of OMSW as a soil amendment represents a promising agricultural practice. OMSW must be applied to soil only after appropriate decomposition processes, addition of lignocellulose residues to obtain adequate physical conditions for composting, due to their sticky texture [19,20] and phenol degrading bacteria that reduced phytotoxicity scale. The OMSW composts supported the growth of plants and enhance the microbial community of the soil. The composting process brought about the total disappearance of phytotoxicity found in raw materials.

The addition of OMWAE (olive-Mill waste aqueous extract) to the seeds exhibited a severe reduction to the germination parameters while the addition of OMWCP (olive-Mill waste crude powder) to the soil at least one month before sowing may be favorable for dry matter accumulation, metabolite synthesis and nutrient uptake taking in consideration the concentration and time of application [21].

The affiliated aim in the current laboratory-scale study is to assess the suitability as a vermicomposting substrate of exhausted OMSW either on its own or mixed with cow manure (C) and/or municipal biosolids as wheat straw (W).

### Germination percentage (GP)

The unamended OMSW showed concentration dependent decrease in germination percentage (GP) of *V. faba* seeds upon increasing its concentrations compared to other bio-treatments: W-OMSW (Wheat straw-olive-mill solid waste), C-OMSW (Cow manure-olive-mill solid waste) and C-W-OMSW (Wheat straw-Cow manure-olive-mill solid waste). This may be due to the presence of phytotoxic phenolic compounds. It is known that, OMSW inhibits seeds germination of different plant species [22,23]. Some plant extracts or residues may inhibit germination, emergence and subsequent growth of other plants by exuding toxic substances. These substances are called allelochemicals [24]. When these allelochemicals are taken up by germinating seeds of the same or of other plant species there may be some degrees of germination and emergence inhibition or growth injury [25].

Recent work showed that olive waste compost suppressed *Fusarium oxysporum* f. sp. *Melonis* and *Verticillium dhaliae* [26,27]. In the present study, After the first time interval (composting for 2 months), there was insignificant difference between the germination rates in W-OMSW compost treatment at various concentration levels compared to unamended OMSW treatment. That may be due to the addition of wheat straw which reduces the moisture content and increase the free air space. Compost should be kept moist, but not saturated. If the materials are too wet, they will compact and restrict the airflow through the pile. Therefore, this leads to anaerobic conditions, which slows down the degradation process and causes foul odors. The high rate of *V. faba* germination in different concentration levels of C-OMSW compost treatments suggests that compost is not phytotoxic. Additionally, such stimulation was supported by data of another study in which the toxicity test of chicken manure and solid fraction of olive-mill residues compost extracts were not toxic to plants but also stimulate seed germination and seedling growth rates compared to control [28].

After composting for five months the germination percentage of *V. faba* was improved to be very close to control at 10% C-W-OMSW and 10%, 20% W-OMSW mixtures [29]. showed that the materials, such as leaves, hay or straw, provide channels to oxygen inflow into and through the compost. This system also keeps the accumulation compost from settling and compacting for long time. The inhibition capability of C-OMSW compost may be explained by the formation of toxic metabolites by some microbial communities that developed in the piles during the longer period of the composting process [30].

Similar studies reported the toxic effects of phenolic compounds on higher plants [31-34] and it has been suggested that such effects arise from alterations of water uptake, or of the metabolism of auxins and/or other phytohormones [34-36]. A clear relationship between phenolic compounds and abscisic acid (ABA) on the inhibition of seed germination in lettuce was suggested Li [37]. In particular, it was shown that mono-hydroxy phenolic compounds increased while polyphenols decreased the inhibitory action of ABA both on seedling growth and seed germinability. These phenolics reduce the seed germination percentage [38,39], due to their interference with indol acetic acid metabolism, or synthesis of protein and ion uptake by the plant.

### Plumule (PL) and radicle (RL) lengths

Bioassays of germination, radicle growth and coleoptile growth are used to test the allelopathic potential of crop species [40]. The elongation of radicle or plumule can be in conjugation with GP. Growth bioassays are more often sensitive than germination bioassays [41,42] observed that seed germination has been regarded as a less sensitive method than hypocotyl and radicle length when used as a bioassay for the evaluation of phytotoxicity.

The current study illustrated that, there is a great variation in the influence of different composts brought by composting for 2 and 5 months on plumule (PL) and radicle (RL) lengths of *V. faba*. The reduction may be due to phytotoxic activity of phytochemicals present in aqueous extracts of OMSW [43] found that the elongation of radicle and epicotyl was reduced in all treatments of *Acacia auriculiformis* extract proportional to concentration levels. Effects of *Acacia auriculiformis* extract much more pronounced on epicotyl than radicle elongation. Fag and Stewart [44] suggested that the inhibitory effect was related to the presence of allelochemicals including tannins, wax, flavonoids and phenolic acids.

While the raw OMSW negatively affect PL and RL, all compost treatments stimulated them. This demonstrated the presence of phytotoxic activity of phytochemical exist in unamended OMSW in addition to the absence of phytotoxicity in the prepared compost. Certainly, in C-OMSW compost bio-treatment after two months of incubation, PL and RL exhibited an increase than other treatments at two and five months of incubation. When the time interval increased, the PL and RL of *V. faba* decreased. That may be due to the formation of new intermediate compounds or occurrence of new reactions in piles via increasing the incubation time than two months. Also, toxic symptoms seemed before harvesting after the second time interval such as black spotting on most leaves and rout or stampede of *V. faba* plant, as well as few plants withered away. These symptoms were explained by the appearance of black and weak radicle due to the phytotoxic substances in soil.

### Total dry weight

Experimental findings have confirmed the effect of different composts on total biomass (TB) of *V. faba*. Generally, the total biomass

was showed a significant decrease upon increasing the unamended OMSW after two times of incubation. This result may be due to the high phenolic compounds content in OMSW. It is well known that, phenolic compounds are major contributors to the toxicity and the antibacterial activity of OMW. They limit its microbial degradability. The application of dry olive-mill residues decreased the dry weight of tomato, soybean and alfalfa plants due to its phenolic content Phenols in seeds have also been proposed as germination inhibitors. The presence of phenols causes the inhibition of germination of *Atriplex Triangularis* and *Pinus laricio* seeds.

The application of 40% C-OMSW amendment showed notable increase in the total biomass of examined species after two months of incubation. Similar results were found by Al-Harhi [21] who reported that, the application of composted OMSW showed notable increase in total dry weight of *Citrullus colocynthis*, *Lepidium sativum* and *Trigonella foenum-graecum*. Also, the biomass yields of maize fertilized with OMW compost at 60 and 90 tones ha<sup>-1</sup> were similar to and higher than those receiving inorganic fertilizer observed that OMW compost application in the soil caused positive effects on physical, chemical, and biological properties of the soil. This practice could be suggested as an application for premeditating the soil in the present study and overcome the inhibition problem of untreated OMW disposal practices.

In the present study, the application of different composts increased the total biomass of *Vicia faba* after the first time of incubation than the second time. This result supports the difference in the mechanism of phytotoxic compounds degradation and raises a question about the potential interactions that occur after a period more than two months.

### Total pigment concentrations

The variation in plant pigment content (chlorophyll and carotenoids) is seemed to be attributed to the variation in the environmental conditions and the developmental stage of plants of plants influences the type and degree of reaction to the environmental factors such as irradiance and nutrients [45]. It was obvious from the current study that, the application of composted OMSW increased the total pigment content of *V. faba* after two time intervals compared with uncomposted OMSW treatment and control plants. All prepared composts act as organic fertilizers which play an important role in plant growth, as they are source of plant nutrients. Moreover, they improve soil properties and promote water use efficiency by plants. This is quite expected to enhance photosynthesis.

After two months, the highest pigments content was observed at 40% W-OMSW treatment. The significant increase in total pigment content may be attributed to the low moisture level caused by the presence of wheat straw. On the similar trend, El-Quesni and Shehata [46,47] stated that, the low moisture level was the most effective treatment for promoting the synthesis and accumulation of photosynthetic pigments. The recoded data revealed the positive and active effect of 20% C-W-OMSW amendment on the total pigment content after five months of incubation. The increase in the total pigment content of *V. faba* might be due to the favorable effect of nitrogen and other organic elements which are present in their optimum quantities [48]. It may be due to the decrease in the polyphenols concentrations to an optimum level in this compost. Also, the presence of potassium, magnesium, Iron and copper in their optimum quantities in the compost stimulated the biosynthesis of pigment [49].

### Nutrient concentrations and uptake

The plant nutrients taken up by crops during the growing season

may come from many sources, including soil reserves, added fertilizer or manure, and crop residues. Nutrients such as N, P and K are required in large quantities, while others are required in very small quantities. Plants influence each other by means of exudates [24], leachates from residues incorporated in the growing medium [50-52] or residues in natural, undisturbed condition [53]. In addition to inhibiting effect, on growth and development, nutrient uptake also is affected. Previous assertions had suggested that allelochemicals inhibit seed germination by blocking of nutrient reserve and cell division thereby caused significant reduction in the growth of plumule and radicle of many crops [54,55]. Furthermore, most of the nutrients contained in the OMW composts supported adequately plant growth, even in short-term crops [56] and that the olive mill waste compost increased markedly the shoot growth of the salt-tolerant sugar beet [57].

Nitrogen (N), phosphorus (P) and calcium (Ca) are critical components which control several metabolic processes required for plant growth. The present study showed that, the application of untreated OMSW and treated OMSW to the soil after two and five months of incubation reduced the elements content and uptake. Several phenolic acids interfere with mineral uptake and cause a subsequent reduction in tissue concentration of nutrients. Plants phenolics are capable of interfere with the uptake of nutrients, and can also affect the rates of nutrient cycling [58]. Earlier works have also reported similar action of chemical compounds enhancing P uptake process. These findings are in agreement with that of Bhowmik and Doll [41] which reported that P concentration was greater with common ragweed (*Ambrosia artimisiifolia*) and velvetleaf (*Abutilon theophrasti*) residues in soybean. Ca uptake was significantly inhibited by  $5 \times 10^{-4}$  M and  $10^{-3}$  M of caffeic acid and  $5 \times 10^{-4}$  M of protocatechuic acid [59].

Mostly, there was a general trend of increase in the concentration of nitrogen, phosphorus and potassium in response to OMW application up to 30% concentration in *Citrullus colocynthis* and *Lepidium sativum*. On the other hand, *Trigonella foenum-graecum* attained its maximum concentration of the studied elements at 20% concentration. Thereafter, the concentration linearly decreased in the three investigated species. In most cases, the reduction in dry weight of the three examined species in the present study was parallel to the reduction in ion uptake Al-Harhi and Alsaadawi [21,59] found that cowpea seedling growth, chlorophyll a, total chlorophyll, chlorophyll a/b ratio, and the uptake of N, P, K, Fe, and Mo were significantly reduced by most of the test concentrations of some phenolic acids. However, chlorophyll b content and the Mg uptake were not significantly affected by all the phenolic acid concentrations. The data also revealed that, the reduction in dry weight was parallel to the reduction in chlorophyll content and ion uptake, and the reduction in chlorophyll was also parallel to the reduction in ion uptake. These data indicate that one mechanism of growth inhibition by this allelochemical may be an alteration of chlorophyll content and nutrient balance.

Alternatively, the present study indicated that potassium (K), sodium (Na) and magnesium (Mg) concentration achieved high values via applying compost incubated for two months. Specifically, C-OMSW compost caused high K and Mg while W-OMSW compost caused the high values of Na. on the other hand, C-OMSW compost caused high Na and Mg while W-OMSW compost caused the high values of K after composting for five months. Abdalla [60] found that the content of K increased with the application of biofertilizer treatments. The interference with nutrient uptake and subsequent reduction in nutrient accumulation is one of the most effective mechanisms of phenolic compounds action [61,62]. Moreover, allelochemicals can alter the rate



at which ions are absorbed by plants. A reduction in both macro- and micronutrients are encountered in the presence of phenolic acids [63].

The current study indicated that iron (Fe), manganese (Mn) and copper (Cu) concentration and uptake achieved high values via applying compost incubated for two months. Specifically, C-OMSW compost caused high Fe and Mn while C-W-OMSW compost caused the high values of Cu. on the other hand, C-OMSW compost caused high Fe and Mn content while C-W-OMSW compost caused the high values of Cu content after composting for five months. The uptake achieved high values of Fe, Mn and Cu via applying W-OMSW compost incubated for five months.

Some characteristics of OMW are favorable for agriculture, since this it is rich in organic matter, nitrogen, phosphorous, potassium and magnesium [64,65]. The organic fraction of the OMW contains a complex consortium of phenolic substances, some nitrogenous compounds (especially amino acids), organic acids, sugars, tannins, pectins, carotenoids, polyphenols and almost all of the water soluble constituents of the olives [66,67]. The inorganic fraction contains chloride, sulfate, and phosphoric salts of potassium as well as calcium, iron, magnesium, sodium, copper, and other trace elements in various chemical forms. The inorganic constituents at the concentration levels found in OMW are not toxic; quite the reverse, they may potentially serve as good sources of plant nutrients and thereby rendering this effluent potentially suitable for recycling as a soil amendment [65]. Additionally, in organic and sustainable farming, the nutritional value of OMW as well as its potential herbicidal activity [68], and ability to induce suppression against soil-borne plant pathogens are of extra value [69].

In summary, The W-OMSW composts recorded the highest concentration of Na element in *V. faba* after composting for two months. The C-OMSW composts recorded the highest concentration of the following nutrients: N, K, Ca, Mg, Fe and Mn. The C-W-OMSW composts recorded the highest concentration of both P and Cu. Moreover, the highest uptake of two nutrients: K and Na were recorded through the application of W-OMSW compost after two months. In addition, the C-OMSW composts showed the highest uptake of N, Ca, Fe and Mn. The C-W-OMSW composts recorded the highest uptake of P, Mg and Cu. After composting for five months, the W-OMSW composts recorded the highest concentration of the following N, Na, Ca, Fe, Mn and Cu in *V. faba*. The C-OMSW composts recorded the highest concentration of K. The highest P and Mg concentration recorded in the C-W-OMSW compost. Furthermore, the W-OMSW composts recorded the highest uptake of N and Cu after five months. The C-OMSW composts recorded the highest uptake of the following nutrients: K, Na, Ca, Mg, Fe, Mn. Furthermore, the C-W-OMSW composts recorded the highest uptake of P.

Soil amendment using compost has several distinct advantages over the addition of fertilizers to soil. There is significant loss of nutrients from soil after fertilization due to leaching. Compost retains nutrients in the soil and releases them on demand by the plants due to its high cation exchange capacity, maintains the water-holding capacity of soil, thereby helping the plants to be more drought resistant.

## Conclusion

The management of OMSW constitutes a long-term and particularly unsolved problem, because of their high organic load, their particular physicochemical composition, the potentially toxic

attributes, the intense of short time interval of production and the high cost investment requirements.

None of the detoxification techniques on an individual basis allow the problem of disposal of OMSW to be solved to a complete and exhaustive extent, effectively and in an ecologically satisfactory way. Existing treatment methods solve partially the problem.

The present work presents a bio-economic analysis of the OMSW treatment using composting with certain biological materials. Where, the composting process brought about the total disappearance of phytotoxicity found in raw materials.

The introduction of the proposed new integrated composts reduces dramatically the environmental damage and provides a profitable alternative to the olive mills due to utilization of all by-products.

The suggested treatments can reverse the effect of large concentrations of OMSW in the following order: C-OMSW, W-OMSW then C-W-OMSW amendment.

The mean composting period can be considered as satisfactory after two months, taking into account that the whole experiment of the OMSW composting is new. So that, the longer time interval of the composting has been considered, longer incubation may have affected the concentration of both toxic substances (polyphenols, organic acids) as well as the benefit substances like organic matter and mineral content.

Thus at the end we must discover what was happened during the composting time intervals more than the two months. Further studies that we are planning now including the understanding of microbial actions that may take place during longer incubation time.

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