

## The effects of fermented olive mill wastewater mixtures on the growth and development of bean (*phaseolus vulgaris*)

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### ARTICLE INFO

#### Keywords:

Olive mill wastewater  
Plant growth  
Biological life  
Antibacterial activity

### ABSTRACT

The extraction of olive oil generates considerable volumes of an agricultural by-product, olive mill wastewater (OMWW), which is damaging to the environment and biological life. To investigate the effect of fermented OMWW on number of pods, pod length, pod weight, dry weight of plant, chlorophyll *a*, chlorophyll *b*, and total chlorophyll of common bean, plants were grown and irrigated with various percentages of fermented OMWW (0%, 4%, 6%, 8%, 10%, and 100%) in a greenhouse. The study found that irrigating agricultural land with a water mix containing a specified quantity of OMWW enhanced physicochemical parameters and influenced plant characteristics. The plant characteristics were substantially affected when wastewater irrigated agricultural areas with water polluted with OMWW at concentrations no more than 4%. According to the findings of this study, utilizing 4% OMWW resulted in the best bean plant growth response, with an increase in growth indices and photosynthetic pigments.

### 1. Introduction

Olive mill wastewater (OMWW) is the liquid by-product obtained from three-phase (solid, aqueous, oil) olive oil extraction systems. Up to 30 million m<sup>3</sup> OMWW is generated annually in the Mediterranean region [1].

Typically, the olive extraction process generates olive oil (20–32%), aqueous part (40–50%), and biosolids production (30%) [2]. Most nations in the Mediterranean region, where 97.5% of the world's oil is produced, rely heavily on olive oil production for socioeconomic growth [3].

OMWW is a serious issue since it comprises a large amount of water as well as both organic substances and nutritious components. OMWW consists of vegetation water, tissues of olive fruits, and process water used in different stages of the extraction. Therefore, it has a high biological and chemical oxygen demand, high concentrations of soluble phenolic compounds (Total Phenols; TP), oil residues, as well as the residual solid content (Total Suspended Solids) and, therefore, it cannot be processed by conventional sewage treatment plants [4].

In Jordan, OMWW is the most polluting activity in terms of discharge volume, chemical content, and/or strength, and if discharged

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<https://doi.org/10.1016/j.wri.2023.100201>

Received 29 March 2022; Received in revised form 24 December 2022; Accepted 17 January 2023

Available online 26 January 2023

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into municipal sewers or surface waterways, it will harm aquatic life as well as humans and the environment due to its elevated organic content phytotoxicity and antimicrobial properties [5]. When put to the soil, these wastes have a high fertilizer value due to their high organic matter concentration and some mineral nutrient content [6]. Additionally, OMWW soil amendment is well-known for its antibacterial activity and ability to reduce diseases in plants [7]. As a result, more focus has been made on discovering the best methods for distributing OMWW on agricultural lands and recycling both organic substances and nutritious components in the soil crop system [8].

Several studies have shown that a controlled distribution of OMWW over soil layer might be used as a composting approach [9–11]. It has also been suggested that OMWW might be utilized as a low-cost biofertilizer [12]. Food and clean water are the two most fundamental requirements for survival. As a result, offering both items to customers is the top priority and the major problem for the global economy. A tremendous increase in global population, along with urban growth, has contributed to a significant increase in water and food usage, while available resources have become increasingly scarce. The gap between supply and demand for water is getting wider and has reached such alarming levels that it is facing a danger to human presence in some regions of the world. When accessible water supplies are limited, reusing OMWW effluent is an appealing alternative since it may reduce strain on water supplies while also providing an extra water resource.

Agriculture consumes a lot of water, contributing for more over 70% of overall water use in other areas like Jordan [13]. Improved water planning in agriculture is critical to lowering usage, minimizing network losses, and reducing evaporation. One of most common way for dealing with the challenges connected with that whole effluent is to apply it directly to cultivated soils as biofertilizer [3]. Utilizing its beneficial components would provide a different, economically viable, and environmentally friendly strategy to managing OMWW. Organic matter, nutrients, and water are all significant components of OMWW. Organic matter is crucial for a number of soil features, including soil structure, ion binding, pH buffering, and water retention. When used at the right application rates, OMW can serve as a natural fertilizer and soil amendment [14].

Several studies were conducted regarding the use of treated water as an organic fertilizer.

[15–18]. The application of OMWW fertilizer to the soil improves the chemical and physicochemical properties of the soil (Blankes et al., 2002). On the other hand, the results of experiments conducted on some plants that used OMWW bio-fertilizer to increase agricultural productivity showed that the yield gained using bio-fertilizer was higher than that obtained using conventional inorganic fertilizer. [16]; Tsagaraki et al., 2007). In contrast to previous studies [19,20], reported that OMWW application has a negative impact on cultivated plants.

The impacts of fatty acids and phenolics found in OMWW may change the bioavailability of nutrients within living cells, affecting plant development [21]. According to Ref. [22] the most beneficial results from adding OMWW were observed when OMWW was treated in a repeated-batch method.

The purpose of using OMWW to irrigate crop plants is to safely dispose of effluents that may be harmful to the environment and human health, as well as to maximize fertilizer value. The quality of irrigation water is critical for plant growth since it can change various physical soil properties such as TDS, EC, temperature, turbidity, hardness, and sedimentation, as well as chemical properties such as alkalinity and acidity.

The primary goal of this comparative research was to assess the suitability and effects of reusing OMWW at different concentrations on the growth performance of green bean. The fermented olive wastewater came from different manufacturers; therefore, it was not exclusive to one species production process. Green bean (*Phaseolus vulgaris*) plants were chosen for this study because they are a fast-growing plant that can be grown in the home since they are easy to nurture and provide a good crop for garden space required.

## 2. Materials and methods

The soil used in this study was fine-loamy, mixed, thermic, and Calcic Paleargid calcareous soil with a low microbial activity collected from the surface (0–20 cm) of the Research Station at Al-Huson University College of Al-Balqa Applied University in northern Jordan (32°27'N, 35°27'E). The soil was sieved using a 2 mm sieve after it had been air-dried. The soil was evaluated for main physicochemical parameters, while particle size distribution was assessed using a hydrometer [23]; soil electrical conductivity (EC) was measured on 1:1 soil: water extracts. The organic matter content was assessed using the Walkley-Black technique, and accessible phosphorus by sodium bicarbonate extraction. It should be noted that this soil never really taken OMWW before. Experiments were conducted at the greenhouse of Al-Huson University College.

OMWW was fermented with a liquid culture through each biologically active phenol degrading bacterium, *Enterobacter asburiae* and *Pseudomonas aeruginosa*. In comparison to unfermented OMWW which has a higher level of total nitrogen, soluble nutrients (nitrogen and potassium), and lower COD, BOD, and total phenolic content [24].

Eighteen pots were filled with dry soil, in these pots, 10 kg of dry soil was added, along with five seeds of *Phaseolus vulgaris* were planted at a depth of 5–7 cm in each pot.

As a source of irrigation water, the settled OMWW with and without dilution with potable water was applied to the soil. A greenhouse pot experiment used a randomized complete block design with three replications to examine the following treatments.

1. %100 OMWW, pure OMWW.
2. OMWW that has been diluted with 480 water: 20 OMWW (4% OMWW, V/V)
3. OMWW that has been diluted with 470 water: 30 OMWW (6% OMWW, V/V)
4. OMWW that has been diluted with 460 water: 40 OMWW (8% OMWW, V/V)
5. OMWW that has been diluted with 470 water: 50 OMWW (10% OMWW, V/V)

The physiological and chemical analyses of irrigated water at various concentrations are shown in Table 1.

All pots were irrigated to saturation with tap water on the first day, and then 1 L per pot was irrigated for two weeks until a vigorous shoot formed. Following the two-week period, each sub-group was watered with its treatment. Finally, once the plants had ceased growing, measurements of dry weight of plant, number of pods, weight of pods, and length of pods, total chlorophyll, chlorophyll *a* and *b* were taken.

The number of pods, length of the pods, fresh and dry weight were obtained at the end of experiment. The plant was covered in aluminium foil and placed in an 80 °C oven for 24 h. Weighing was done with a single pan electrical balance. The pigment analyses, chlorophyll *a*, chlorophyll *b*, and total chlorophyll, were calculated after 15 days of germination in varied levels of effluent [25].

Chemical oxygen demand (COD), 5-day biochemical oxygen demand (BOD<sub>5</sub>), pH, phenol, ammonia, phosphate, and electrical conductivity were all determined in the collected samples (EC). All these analyses were carried out in accordance with the Standard Methods for the Examination of Water and Wastewater (1995). All treatments were repeated three times, and the findings were statistically analysed as the averages of the three replicates.

### 3. Results and discussion

The fermented OMWW included a high concentration of total suspended solids (TSS) (Table 1). Turbidity, phenol, phosphorus, EC, BOD and COD were found to be the most abundant in 100% OMWW. The effect of irrigation with different percentages of OMWW on the number of pods in common bean (*Phaseolus vulgaris* L) was demonstrated in Fig. 1. In practice, OMWW dilution occurred when water encountered OMWW that had previously been put to the soil for irrigation.

Plants irrigated with varied percentages of OMWW significantly produced more pods per plant, ranging from 2.3 to 12.7% more than the control ( $p \leq 0.05$ ) with the highest results obtained when irrigated with 4% OMWW. Irrigation with different percentage of OMWW to bean plants (*Phaseolus vulgaris* L) increased pod length significantly, ranging from 5 to 23% more than the control ( $p \leq 0.05$ ) with the highest results obtained when irrigated with 4% OMWW (Fig. 2).

The effect of irrigation with fermented OMWW had a significant effect on pod weight. OMWW 4% and 6% scored the highest shoot fresh weight of pod (5.78 and 5.69 g, respectively) and the lowest (5.16 g) under control Treatment (Fig. 3). Irrigation with fermented OMWW showed a significant effect on plant dry weight (Fig. 4). Plant dry weight was the highest (9.26 g) at OMWW 4% and the lowest (7.66 g) at OMWW 0% (control). The increase percentage of plant dry weight was 20.9%, 9.7%, 3.3%, 2.59%, respectively, at 4% OMWW, 6% OMWW, 8% OMWW, 10% OMWW compared with the plant dry weight (7.66 g) at tap water.

Figs. 5–7 depicts the effect of fermented OMWW on photosynthetic properties. Fermented olive mill wastewater irrigation at 4 and 6% significantly increased chlorophyll *a* (Fig. 5). With 4% and 6% fermented OMWW irrigation, chlorophyll *a* was increased by about 31% and 15%, respectively. The response of chlorophyll *b* to fermented OMWW irrigation was opposite to the effects on chlorophyll *a* (Fig. 6). The results of irrigation with fermented OMWW at different percentage showed a significant effect in total chlorophyll content in the early growth stages (Fig. 7). Significant differences ( $p < 0.05$ ) indicated that irrigation with fermented OMWW 4% scored the highest total chlorophyll content (9.2) followed by 8.22, 8.15, 7.79, 7.70 under water treatments of OMWW 6%, OMWW 8%, OMWW 10% and OMWW 0%, respectively.

Fig. 8 shows the effect of irrigation with different Percentage OMWW on *Phaseolus vulgaris* growth performance with the highest results obtained when irrigated with 4% OMWW except for chlorophyll *b*. Irrigated with tap water and planted or growing in clay exclusively, the percentage in Fig. 8 reflects the relative growth - rise or reduction of certain plant characteristics when compared to other irrigated fluids and different growing types of soils.

The findings of this study correspond with those of [22]; who found that when OMWW was treated in a repeated-batch technique, the seed biomass, spike number, and kernel weight rose in the treated plants [16]. found that fresh OMWW may be transformed utilizing composting technology to boost soil fertility and plant productivity. According to Ref. [3] plants irrigated with OMWW have higher biomass, spike number, plant growth, and comparable or even higher dry production than plants watered with water. According to Ref. [26]; The catalytic treatment of olive oil mill wastewater with MnO<sub>2</sub> can result in a quality change that increases its ability as a soil modification while lowering environmental risk, which has a significant impact on wheat grain yield by increasing the number of kernels per unit area, spike density, and kernel weight [27]. observed that the use of table olive waste as irrigation water throughout the growing of four Mediterranean plants: tomato, strawberry, cucumber, and pepper boosted growth of plants, fruit size, and total overall output by at least 20% against dipping and spraying. Powdery mildew illness was also decreased in strawberry plants. According to Ref. [28]; OMWW strengthened plants and increased their development and health. Indeed, treated plants performed much better in the following areas: (i) plant height (146.2 and 130.5%), and (ii) average stem diameter. According to Ref. [29]; treated

**Table 1**  
Physiochemical analysis of fermented Olive Mill Wastewater at various concentration.

Treatments		TSS mg/ L	Turbidity NTU	Phenol g/ L	Phosphorus mg/ L	EC mS/ cm	BOD mg/ L	COD mg/ L
Fermented olive mill wastewater (OMW)	100% OMWW	1300	3.5	70.8	115	21	3005	6400
480 water: 20 OMWW	4% OMWW	52	0.14	2.8	5	0.9	120	250
470 water: 30 OMWW	6% OMWW	78	0.21	4.3	7	1.3	180	384
460 water: 40 OMWW	8% OMWW	104	0.28	5.7	9	1.7	240	512
450 water: 50 OMWW	10% OMWW	130	0.35	7.1	12	2.1	300	640

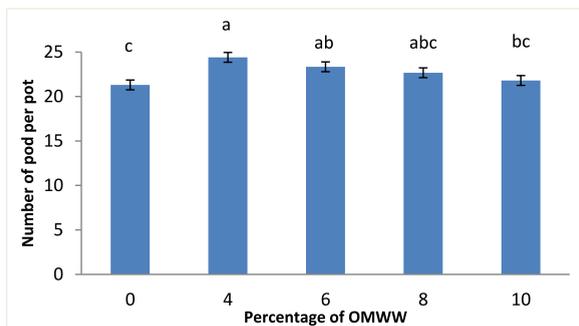


Fig. 1. Effect of irrigation with different Percentage OMWW on Number of Pods.

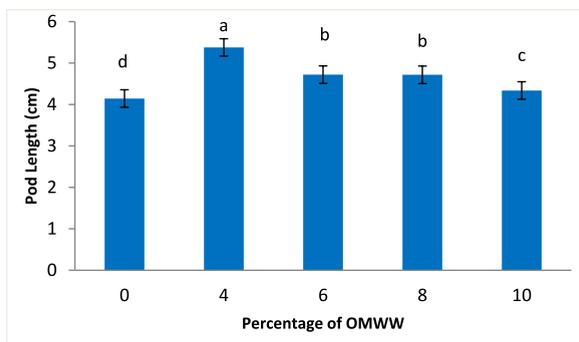


Fig. 2. Effect of irrigation with different Percentage OMWW on Length of Pods.

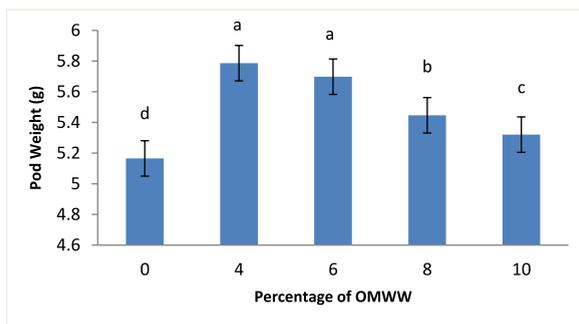


Fig. 3. Effect of irrigation with different Percentage OMWW on Weight of Pods.

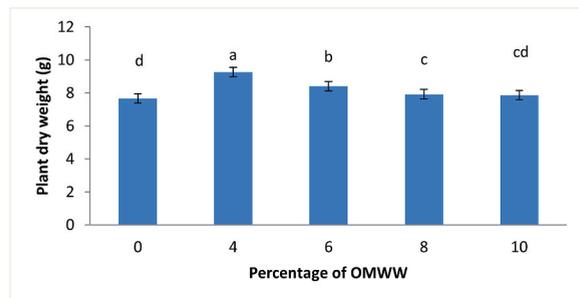


Fig. 4. Effect of irrigation with different Percentage OMWW on Dry Weight of Pods.

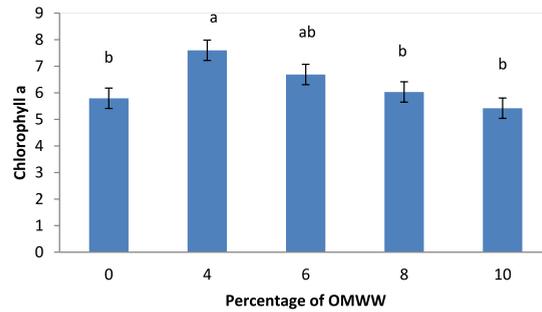


Fig. 5. Effect of irrigation with different Percentage OMWW on chlorophyll a.

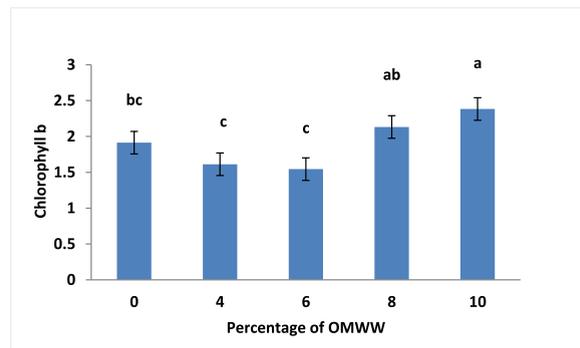


Fig. 6. Effect of irrigation with different Percentage OMWW on chlorophyll b.

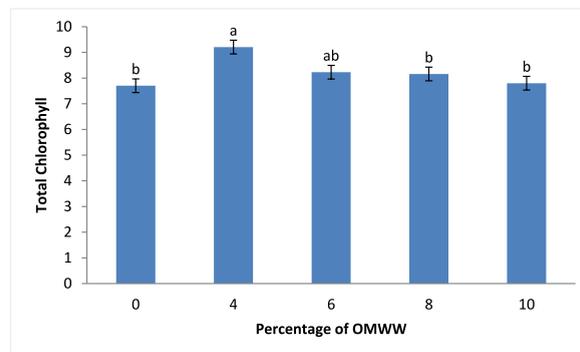


Fig. 7. Effect of irrigation with different Percentage OMWW on total chlorophyll.

plants utilizing OMWW by various methods increased plant growth [30]. discovered that smaller dosages of OMWW had a favorable impact on plant development, while higher concentrations had a harmful effect. In contrast to Ref. [20]; the results reveal that the plants grown reduced after OMWW spreading due to a lower tillering index, the plant did not yield new leaves but a greater number of secondary shoots, which limited the leaf area index, and the durum wheat grain yield reduced from the first to the last years in comparison to total dry matter yield. Moreover [21], discovered that the effects of fatty acids and phenolics in OMWW may alter the access of nutrients inside bio membranes, influencing plant growth. According to Ref. [31], when effluent concentrations increased, so did chlorophyll content; similarly, chlorophyll content was higher than in contaminated soil. The plant characteristics were substantially affected and can be explained by the fact that OMWW is a salt-free liquid with fungicide properties, which will improve plant number, length, etc. As a result, OMWW may be used as a fertilizer to increase plant strength and production, as well as a fertilizer. Many scientists have established that this OMWW has a high fertilizer value when applied to the soil. Because olive mill effluent includes a high concentration of organic matter in a big volume of water, it has the potential to be used as a fertilizer, particularly for soils and crops. OMWW holds significant levels of organic content (30–200 kg COD m<sup>-3</sup>), particularly aromatics, which, when combined with its high C/N ratio and low pH, impede biological breakdown operations.

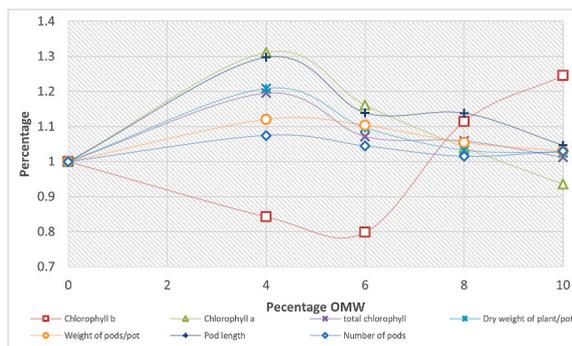


Fig. 8. Effect of fermented olive oil mill wastewater (OMWW) mixed with tap water at various ratios on *Phaseolus vulgaris* growth performance.

OMWW boosted soil carbon content while maintaining a very low specific respiration. The change, on the other hand, had a favorable effect on soil-specific respiration. Fungi, the most significant organisms degrading lignin and polyphenols, were boosted by the OMWW. As a result, this fungal thrived on soil changed with OMWW, where the pH and C/N ratio were similarly favorable [32]. The primary concept in OMWW reuse was that most of the mineral elements (K, P, N) produced from the soils during cultivation may be reused by employing treated olive wastewater as a natural nutrient, soil amendments, and a favorable irrigation supply of water [33]. According to Ref. [34]; antibacterial substances discovered in OMWW included tyrosol, hydroxytyrosol, oleuropeine, 3–4 dihydroxyphenyl acetic acid, and 4-hydroxybenzoic acid. Implementation on certain plants, though, may inhibit seedling growth and early plant development in a range of plants (Cavallaro et al., 2014), and might even promote leaf and fruit abscission owing to plant sensitivity to the amount of OMWW supplied [20].

The work will result in increasing volumes of higher-quality olive oil and more efficient and cost-effective oil production in dry nations like Jordan. Every year, OMWW will be used as a source of irrigation, with varying amounts deemed supplementary irrigation, to produce higher-quality oil at a lesser cost. Researchers and farmers will need to collaborate to establish the ideal harvest period, development of olive oil yield, and quality of olive oil in order to further improve olive oil quality and production efficiency.

#### 4. Conclusion

The study found that irrigating agricultural land with a water mix containing a specified quantity of OMWW enhanced physico-chemical parameters and influenced plant characteristics. The plant characteristics were substantially affected when wastewater irrigated agricultural areas with water polluted with OMWW at concentrations no more than 4%.

Except for chlorophyll *b*, 4% OMWW irrigation yielded the greatest results in all pots. With 2–8% more pods per plant, 5–30% more pod length, 3–11% more pod weight, 2.5–21% more pod dry weight, 4.1–31% more chlorophyll, up to 8% more OMWW, and 1–20% more total chlorophyll, the greatest outcomes were attained.

#### Authors' contributions

Zaydoun Abu Salem, He planned for the cultivation of the crops. Kamel Alzboon, He selected the suitable area for cultivation, selected the suitable seeds. Jalal A. Al-Tabbal, He was entering the data on excel and continuing the statistical analysis results. Participate in writing the manuscript especially discussion and conclusion. He also formatting the paper at your journal style. He also participates in English editing of the manuscript. Participate in writing the manuscript especially discussion and conclusion. Moawiya A. Haddad and Nabeel M. Bani-Hani, Followed the planting of seeds until the plants became fully developed.

#### Funding

“The author(s) thanks UN for funding this research through Perez-Guerrero Trust Fund for South - South Cooperation.”. Also special thanks for Al-Balqa Applied University, Al-Huson College for making this work possible.

#### Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

#### Data availability

Data will be made available on request.

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