

Growth, Water Relation and Physiological Responses of Eggplant (*Solanum melongena* L.) under Different Olive Mill Waste Water Levels

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Abstract

Eggplant (*Solanum melongena* L.) is an important traditional crop, cultivated worldwide. Olive mill wastewater (OMW) is an important olive oil extraction byproduct due to its high concentration of different valuable compounds. Three eggplant cultivars were evaluated under different concentrations of olive mill wastewater. Seedlings were treated with four OMW levels (control, 25% OMW, 50% OMW and 100% OMW). Eggplant cultivars expressed different physiological responses when irrigated with OMW at different levels. Many physiological parameters such as growth, stomatal conductance, transpiration, relative water content, relative growth rate, stem diameter and chlorophyll content were examined. It has been found that many of these responses were adversely affected when plants were irrigated with OMW. Unexpectedly, the water status of eggplants was not affected by the different levels of OMW as plants maintained transpiration rate similar to the control values. Our findings clearly suggest that the physiological responses of eggplant to OMW vary among cultivars. However, the ability of plants to absorb water were not affected by OMW. We recommend that the sensitivity of the eggplant cultivars to OMW be taken into consideration before irrigating eggplants with OMW.

Keywords: *Solanum melongena*; Olive mill wastewater; Phenolic compounds; transpiration, diffusive resistant, Plant relative growth rate; Net assimilation rate.

1. Introduction

Demand for food is much more than ever before. The food production chain including agriculture and industry leads to accumulation of wastes that need to be assimilated or safely disposed with minimum environmental impacts. Olive mill wastewater (OMW) is an example of olive oil extraction byproduct that can be of beneficial for some uses due to the high concentration of different compounds. Olive (*Olea europea* L.) trees are drought tolerant ones commonly grown in Mediterranean region as viable trees that do not require much effort (Brito *et al.*, 2019, Torres-Ruiz *et al.*, 2013). Olive oil marketing is an expanding worldwide industry especially in the olive-growing regions due to the high demand for its fruits and oil and the associated economical value (Khdair and Abu-Rumman, 2020, Brito *et al.*, 2019). The production of olive oil in Mediterranean countries represents 95-98% of the entire worldwide total production (Arvaniti *et al.*, 2012).

Processing of olive fruits to extract olive oil generates two types of waste which are a large quantity of solid waste called pomace (Khdair and Abu-Rumman, 2020) or olive cake and OMW that is generating a main ecological problem (Khdair and Abu-Rumman, 2020, Arvaniti *et al.*, 2012). Olive pressing industry produces 1,000 metric tons of this OMW per harvesting season (annually) in

Mediterranean countries in a short period of time (October to late January) (Khdair and Abu-Rumman, 2020, Paraskeva and Diamadopoulos, 2006). The amount of OMW differs according to the extraction method, with a range of 50 and 80 m³ ha⁻¹ using pressure or centrifuge extraction techniques (Belaqziz *et al.*, 2008, Hanifi and El Hadrami, 2009).

OMW is a dark brown color acidic waste, highly saline, and rich in organic matter and minerals (Arvaniti *et al.*, 2012, Comegna *et al.*, 2022, Rusan and Malkawi, 2016, Lopes *et al.*, 2009). The organic matter composition differs in its phenolics, sugars, polysaccharides, proteins, lignins and fatty acids content according to type of olive, treatment procedure, the level of fruit maturity, time of harvest, and processing method (Arvaniti *et al.*, 2012, De Marco *et al.*, 2007).

The ecological problem of OMW arises from the high concentration of phenolic, tannins and flavonoids compounds that negatively impacts plant growth and microorganisms if approaching toxicity levels (Comegna *et al.*, 2022, Arvaniti *et al.*, 2012, Isidori *et al.*, 2005). In addition, OMW is characterized with unpleasant odor after anaerobic digestion (Arvaniti *et al.*, 2012) and represents a threat to pollute surface and groundwater sources, with an adverse impact on the environment aesthetic value (Khatib *et al.*, 2009, Arvaniti *et al.*, 2012). Therefore,

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some European countries prohibit discharging of this wastewater in nature (Hanifi and El Hadrami, 2009).

The uncontrolled disposal of OMW on soil leads to reduction in soil water retention ability and infiltration rate, increases soil hydrophobicity, adversely affects soil acidity, salinity, nitrogen immobilization, and nutrient leakage (Khdair and Abu-Rumman, 2020, Sierra *et al.*, 2007). Different methods have been proposed for OMW treatment based on evaporation ponds, thermal concentration, and different physicochemical and biological treatments (Martinez *et al.*, 1994). Unfortunately, most of these methods are costly, and still the end by-products need to be disposed and uploaded to an environment system (Khdair and Abu-Rumman, 2020, Arvaniti *et al.*, 2012, Paredes *et al.*, 1999). However, some researchers reported advantages and benefits associated with OMW where incorporating OMW with soil changes soil physicochemical and microbiological properties, reduces evapotranspiration in arid and semi-arid zones and improves soil water balance, amends soil structure, decreases soil erosion, increases infiltration rate and water holding capacity with no negative impacts on soil properties (Kurtz *et al.*, 2021, Khdair and Abu-Rumman, 2020, Rusan and Malkawi, 2016, Rusan *et al.*, 2015, Rigane *et al.*, 2015, Ayoub *et al.*, 2014, Wahsha *et al.*, 2014, Barbera *et al.*, 2013). Furthermore, OMW can replenish soil with macronutrients (nitrogen, phosphorus and potassium) along with organic matter that is beneficial to soils (Comegna *et al.*, 2022, Khdair and Abu-Rumman, 2020, Rusan and Malkawi, 2016, Rusan *et al.*, 2015). Therefore, there is a potential for OMW to be exploited in agricultural as source for nutrient, soil conditioner, and a water conservation technique.

Eggplant (*Solanum melongena* L.) is a vegetable crop belonging to *solanaceae* family. Eggplant is a traditional crop mainly in Asia, Southern Europe and the Mediterranean countries. It is the fifth most economically important solanaceous crop after potato, tomato, pepper, and tobacco (Taher *et al.*, 2017). In 2008, about 1.96 million ha were devoted to cultivating eggplant worldwide (FAO, 2010). The large annual quantities of OMW production accompanied with both its climatic hazards and its nutritive values to plants nominated this product for more investigation. The aim of this work was to investigate the growth rate, water relations, and physiological responses of three eggplant cultivars under different concentrations of the olive mill waste water applications.

2. Materials and Methods

2.1. Study location

This study was conducted in a greenhouse at the Hashemite University, Zarqa, 32°05' N Latitude and 36°06' E Longitudes. Greenhouse day temperatures were in the range of 20-35°C, and mean midday photosynthetic photon flux density (PPFD) was 365 $\mu\text{mol}\cdot\text{s}^{-1}\cdot\text{m}^2$ measured by a quantum sensor (LI_250A; LICOR.)

2.2. Plant Material and Experimental Design

Seeds of three eggplants (*S. melogena* L.) cultivars (Blacky, Hi-Tech, Denmark; Pearly F1, Hi-Tech, Denmark; and Classic, Harris moran, China) were used for this experiment. Seeds were germinated in trays containing

peatmoss (KEKKILA, European Union). After 30 days of seeding date, seedlings were transplanted into 5 L pots containing autoclaved mixture of fumigated media composed of peatmoss: perlite: soil (1:1:1v/v). Cloth screens were placed in the bottom of the pots to prevent soil loss and allow drainage of extra irrigation water. Transplanted seedlings were kept well irrigated in the greenhouse for a month. Plants were fertilized using Nutri-Leaf 60, USA (20N-20P- 20K) at a rate of 5g/L of irrigation water once a week after transplantation.

The plants were gradually irrigated with OMW solution until the designated treatment levels were reached in order to acclimatize the plants and avoid shock. Four levels of irrigation water mixed with OMW were used (control, 25% OMW, 50% OMW and 100% OMW). Uniform plants from each cultivar were assigned randomly to one of four irrigation treatments and allowed to grow for 65 days. The experimental design was completely randomized block design with five replications. Each block contained 12 plants (3 cultivars x 4 irrigation solution). An extra 24 plants (8 per cultivar) were used to determine the initial plant growth characteristics before initiating salinity treatment application.

2.3. Initial Seedling Traits

Initial plant growth characteristics were collected for each cultivar using the extra plants maintained for this purpose. The harvested plants were separated into leaves, stems and roots. The collected baseline data included leaf area, leaf dry weight, stem dry weight and root dry weights. Leaf area (cm^2) was measured using a portable leaf area meter (LI-3000A; LICOR; Lincoln, Nebr. USA). Roots were washed by tap water to remove soil mixture. Oven dry weights of leaves, stems, and roots were determined after drying at 65 °C to a constant weight (data not shown).

2.4. Treatments preparation

2.4.1. Determination of total phenol content in the OMW

Total phenol content (TPC) was determined according to Slinkard and Singleton, (1977) with Folin-Ciocalteu reagent using Gallic acid as a standard. Briefly, 1 ml of approximately diluted samples and a standard solution of Gallic acid were added to a 25 ml volumetric flask containing 9 ml of distilled water. Distilled water was used as blank reagent; one ml of Folin-Ciocalteu phenol reagent was added to the mixture and were shaken for 5 min. Then, 10 ml of 7% Na_2CO_3 solution were mixed and then allowed to stand for 2 h. Next, the absorbance was measured at 760 nm using spectrophotometer. The samples TPC was averaged to 0.753 $\text{mg}\cdot\text{l}^{-1}$, and that was considered as stock OMW solution (100%). The remaining treatments were prepared from the stock solution and diluted to (50% and 25%) and that was equivalent to 0.377 and 0.188 $\text{mg}\cdot\text{l}^{-1}$, respectively.

2.4.2. OMW solution characteristics

Electrical conductivity (EC) using EC meter (Milwaukee SP500), TPC and pH were determined for the OMW irrigation solutions (Table 1).

Table 1: Electrical conductivity, pH and TPC of OMW irrigation solutions.

Parameters	OMW irrigation solution		
	100%	50%	25%
pH	5.5	5.8	5.8
Electrical Conductivity (EC) (dS. m ⁻¹)	1.870	1.325	1.092
TPC (mg.l ⁻¹)	0.753	0.377	0.188

On the same day when the initial data were recorded, the five blocks were irrigated with OMW treatment (25%, to prevent shock, except for the control that was watered with tap water only. The OMW concentration was then gradually increased for two weeks until each desired OMW treatment level was achieved (control, 25% OMW, 50% OMW and 100% OMW). Once all the experimental plants had started receiving their designated treatments levels, all treatments were irrigated manually every two days to the field capacity for the total duration of 65 days.

2.5. Physiological Traits

Chlorophyll Concentration Index (CCI) was determined biweekly by averaging two midday readings of each plant. The SPAD was measured on the two youngest fully-expanded mature healthy leaves (LI-250A OPTICSCIENCES CCM-200). Transpiration and stomatal conductance (g_s) were measured biweekly using a steady state porometer (LI-1600; LICOR; Lincoln, Nebr.). The height of each plant was measured biweekly from soil surface to the top of the plant.

2.6. Final Harvest

All plants were harvested after 65 days of initiating the experiment. Harvested plants were washed, air dried on filter paper, separated into leaves, stems and roots. Leaf area (cm²) was determined using a portable leaf area meter (LI-3000A; LICOR; Lincoln, Nebr. USA). Stem diameters were measured using an electronic 0-150 mm digital caliper (Swiss). The oven dry weights of plant leaves, stems and roots were determined after the dry weight was stabilized at 65 °C. Leaf discs from two youngest fully expanded mature leaves of all plants were used to determine Relative Water Content (RWC).

RWC was calculated using the equation (FW-DW/TW-DW) (100), where FW is the fresh weight, DW represents

fresh weight sample oven dried at 68 °C and TW represents the turgid (saturated) weight of sample, which was immersed overnight in distilled water (Bsoul *et al.*, 2006). Relative growth rates were calculated using the equation of Gutschick and Kay, (1995): $RGR = (\ln W2 - \ln W1) / (T2 - T1)$, where W2 was the final dry weight at day 65 (T2), and W1 was the initial DW determined from initial data harvest on day 1 (T1). Net assimilation rates (NAR) were calculated as: $NAR = M2 - M1 / T2 - T1 \times \log L2 - \log L1 / L2 - L1$, where M2 was the final dry weight at day 65 (T2), and M1 was the initial DW determined from the initial weight recorded on day one of the experiment (T1). Leaf area ratio (cm².g⁻¹) was calculated as $SLA = \text{leaf area} / \text{leaf dry weight}$ where SLA represents Specific Leaf Area. Specific stem length (cm.g⁻¹) was calculated as $SSL = \text{stem height} / \text{stem dry weight}$.

2.7. Statistical Analysis

Statistical analysis was performed using SAS 9.1 software for Windows (2003). Significant differences between values of all parameters were determined at $P \leq 0.05$ using Proc GLM, PDIF option, ANOVA and Duncan's Multiple Range Tests were used.

3. Results

There was no significant interaction observed between cultivars and treatments. Stem dry weight had no significant differences among cultivars or treatments. Pearly cultivar had the lowest leaf dry weight, shoot dry weight, plant dry weight, plant height and stem diameter (Table 2). In addition, Pearly had the lowest relative growth rate, but the three cultivars had similar NAR (Table 3). Olive mill wastewater has reduced most of the growth parameters of the eggplants when compared to the control (Table 2). Classic cultivar had the thickest stem diameter (5.02 mm). However, the stem diameter had no significant differences among OMW treatments (Table 2). Noticeably, irrigation with high olive mill wastewater concentration (100%) had no effect on the plant height (Table 2) and NAR (Table 3). Olive mill wastewater treatments had no significant effect on the RWC of eggplants cultivars (Table 3). The CCI values were highest for classic cultivar and were negatively affected by all the OMW concentrations as compared to the control (Table 3).

Table 2. Plant biomass dry weights, shoot to root ratio, plant height, and stem diameter of three eggplant cultivars (Blacky, Pearly, and Classic) subjected to four OMW treatments (control, 25%, 50%, and 100%) for 65 days.

Cultivar	Stem DW (g)	Leaf DW (g)	Shoot DW (g)	Root DW (g)	Plant DW (g)	Shoot/ Root (g)	Plant height (cm)	Stem diameter (mm)
Blacky	0.84 ^{zA}	0.84 ^{AB}	1.77 ^{AB}	0.74 ^A	2.32 ^{AB}	3.97 ^A	16.55 ^{AB}	4.43 ^{AB}
Pearly	0.65 ^A	0.52 ^B	1.18 ^B	0.43 ^A	1.61 ^B	4.76 ^A	14.13 ^B	4.21 ^B
Classic	0.87 ^A	1.02 ^A	1.90 ^A	0.84 ^A	2.72 ^A	3.23 ^A	18.85 ^A	5.02 ^A
Treatment								
Control	0.96 ^A	1.52 ^A	2.51 ^A	1.16 ^A	3.53 ^A	3.21 ^{AB}	17.67 ^{AB}	4.78 ^A
25%	0.73 ^A	0.80 ^B	1.54 ^B	0.57 ^B	2.09 ^B	5.65 ^A	14.35 ^B	4.56 ^A
50%	0.68 ^A	0.57 ^{BC}	1.25 ^B	0.51 ^B	1.77 ^B	4.34 ^{AB}	16.05 ^{AB}	4.37 ^A
100%	0.77 ^A	0.29 ^C	1.16 ^B	0.45 ^B	1.48 ^B	2.74 ^B	17.97 ^A	4.51 ^A
P-value								
Cultivar	0.296	0.032	0.033	0.111	0.039	0.036	0.005	0.0423
Treatment	0.371	0.0001	0.0003	0.0065	0.0004	0.0003	0.0937	0.7277

^zMeans (n = 5) within columns followed by the same letter were not statistically different. Means were assessed at $P \leq 0.05$ using Proc GLM, PDIFF option.

Table 3. Plant net assimilation rate (NAR), relative growth rate (RGR), leaf relative water content (RWC), and chlorophyll content index (CCI), of three eggplant cultivars (Blacky, Pearly, and Classic) subjected to four olive mill wastewater treatments (control, 25%, 50%, and 100%) for 65 days.

Cultivar	NAR (mg·cm ⁻² ·d ⁻¹)	RGR (g·g ⁻¹ ·d ⁻¹)	RWC %	CCI
Blacky	0.090 ^A	0.016 ^{AB}	0.36 ^A	8.12 ^B
Pearly	0.067 ^A	0.014 ^B	0.40 ^A	8.96 ^{AB}
Classic	0.099 ^A	0.019 ^A	0.39 ^A	13.30 ^A
Treatment				
Control	0.115 ^A	0.021 ^A	0.37 ^A	15.65 ^A
25%	0.069 ^{AB}	0.016 ^B	0.40 ^A	9.11 ^B
50%	0.066 ^B	0.015 ^B	0.35 ^A	8.08 ^B
100%	0.105 ^{AB}	0.013 ^B	0.42 ^A	7.67 ^B
P-value				
Cultivar	0.0539	0.0227	0.5218	0.0179
Treatment	0.0092	0.0042	0.3289	0.002

^zMeans (n = 5) within columns followed by the same letter were not statistically different. Means were assessed at $P \leq 0.05$ using Proc GLM, PDIFF option.

The three cultivars had similar LA, LAR, and LWR values with no significant differences among them (Table 4). On the other hand, Pearly had recorded the highest SLA. However, LA, SLA, LAR and LWR were significantly impacted by OMW treatments. It is obvious that high OMW concentration (100%) had significantly reduced the eggplants LA, SLA, LAR and LWR (Table 4).

Table 4. Leaf area (LA), specific leaf area (SLA), leaf area ratio (LAR), and leaf weight ratio (LWR) of three eggplant cultivars (Blacky, Pearly, and Classic) subjected to four olive mill wastewater treatments (control, 25%, 50%, and 100%) for 65 days.

Cultivar	LA (cm ²)	SLA (cm ² ·mg ⁻¹)	LAR (cm ² ·mg ⁻¹)	LWR (g·g ⁻¹)
Blacky	180.09 ^A	225.49 ^B	68.62 ^A	0.31 ^A
Pearly	148.58 ^A	271.77 ^A	96.70 ^A	0.33 ^A
Classic	191.20 ^A	194.23 ^B	77.48 ^A	0.38 ^A
Treatment				
Control	298.00 ^A	225.31 ^B	87.71 ^A	0.40 ^A
25%	193.07 ^B	264.01 ^A	109.34 ^A	0.41 ^A
50%	145.11 ^B	249.38 ^A	89.58 ^A	0.36 ^A
100%	56.96 ^C	183.29 ^B	37.10 ^B	0.19 ^B
P-value				
Cultivar	0.4992	0.0062	0.1965	0.2826
Treatment	0.0001	0.0268	0.0016	0.0002

^zMeans (n = 5) within columns followed by the same letter were not statistically different. Means were assessed at $P \leq 0.05$ using Proc GLM, PDIFF option.

The biweekly data showed that eggplant CCI continued to decrease throughout the experiment progress under all OMW concentration as compared to the control (Fig 1A). On the other hand, Classic cultivar had the highest chlorophyll content (Fig 1B).

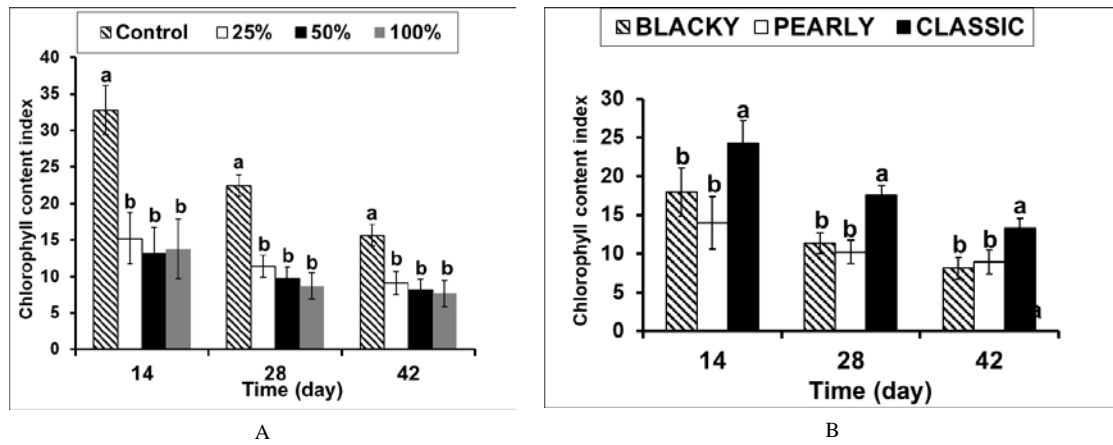


Figure 1: Chlorophyll content index: Biweekly CCI values of three eggplant cultivars (Blacky, Pearly, and Classic) subjected to four olive mill wastewater treatments (control, 25%, 50%, and 100%). Data are means \pm SE of 5 replicates. Means among the neighbored columns marked with the same letter were not significantly different at the $P \leq 0.05$ using Proc GLM, PDIF option.

Eggplant transpiration rate was not affected by the increased OMW concentration in the experiment. However, transpiration rate was increased throughout the

experiment (Fig 2A). After 42 days, the Classic cultivar started to show less transpiration rate than Blacky and Pearly cultivars (Fig 2B).

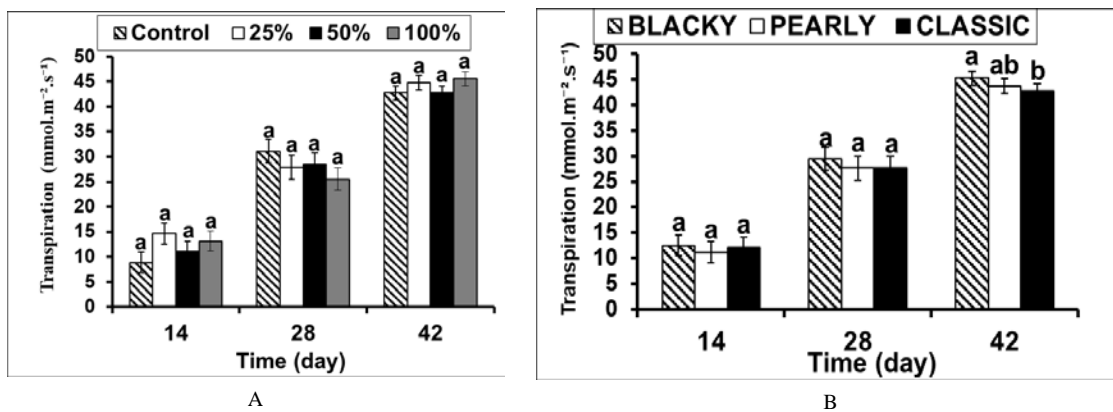


Figure 2: Transpiration: Biweekly transpiration values of three eggplant cultivars (Blacky, Pearly, and Classic) subjected to four olive mill wastewater treatments (control, 25%, 50%, and 100%) Data are means \pm SE of 5 replicates. Means among the neighbored columns marked with the same letter were not significantly different at the $P \leq 0.05$ using Proc GLM, PDIF option.

The three eggplants had similar stomatal diffusive resistance under the different OMW concentration, but

they all had high stomatal diffusive resistance values after 28 days (4 weeks) (Fig 3B).

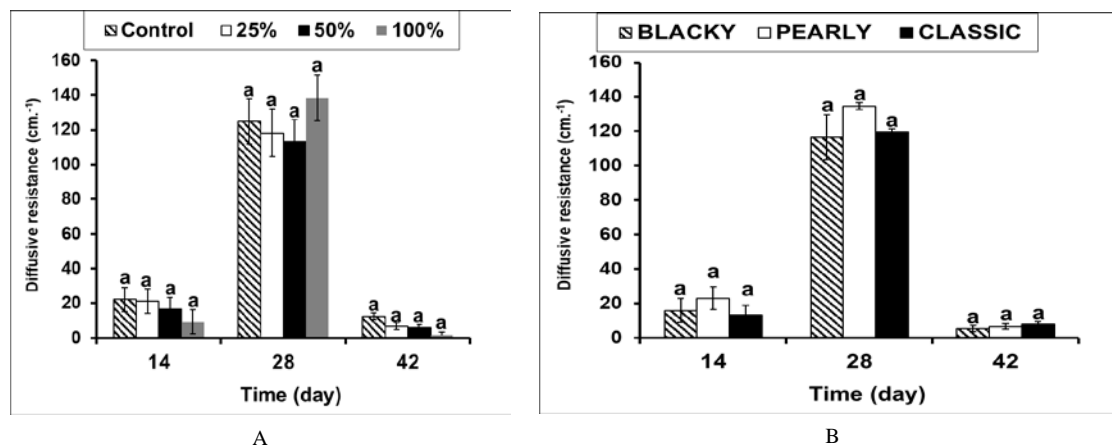


Figure 3: Diffusive resistance: Biweekly Diffusive resistance values of three eggplant cultivars (Blacky, Pearly, and Classic) subjected to four olive mill wastewater treatments (control, 25%, 50%, and 100%). Data are means \pm SE of 5 replicates. Means among the neighbored columns marked with the same letter were not significantly different at the $P \leq 0.05$ using Proc GLM, PDIF option.

4. Discussion

Our data obviously reported that olive mill wastewater negatively impacted the growth of the eggplants, specially at high concentration. However, some cultivars such as Pearly might be affected more than the other cultivars. Rusan *et al.*, (2016) reported that plant growth was reduced when irrigated with untreated OMW. Our data is consistent with Asfi *et al.* (2012) who reported that plant biomass was more affected than plant height under OMW treatments. In addition, negative effect of OMW treatment on dry biomass production was reported in many other plants (Mekki *et al.*, 2006; Ouzounidou *et al.*, 2008; Ouzounidou *et al.*, 2010).

However, OMW had no effect on the eggplant water status at all concentration levels as the RWC was not affected and maintained values similar to the control at all OMW treatments concentrations. Moreover, the three cultivars had similar RWC. Our data might suggest that the OMW treatments had no effect on the absorption and the hydraulic conductance of water, specially when we refer to the shoot/root ratio (Table 2) which showed that the plants maintained good root system even when irrigated with 100% OMW. In addition, the plants transpiration rate and the stomatal diffusive resistance maintained similar readings with the control throughout the experiment (Fig. 2 and Fig. 3). Chartzoulakis *et al.* (2010) reported that the application of raw OMW had no effect on plants stomatal conductance and showed similar values to control plants.

Our data might suggest that the impaired growth of the eggplants in our experiment was not a consequence of water deficit. Bsoul *et al.* (2022) mentioned that water availability is a factor that usually reduces NAR and growth. Based on the results of CCI data (Table 3), we believe that the RGR and NAR are directly impacted by chlorophyll content. Fitter and Hay (2002) reported that RGR represents the extent to which a plant invests its photosynthesis in current growth and improves its ability for upcoming photosynthesis. High OMW concentration (100%) had significantly reduced the eggplants LA, SLA, LAR and LWR (Table 4). Ben-Rouina *et al.* (1999) also reported that the increased levels of OMW irrigation were phytotoxic, and plants might die. The variation of cultivars growth indices (LA, SLA, LAR and LWR) could be attributed to the variable sensitivity among these cultivars as reported in previous research (Montemurro *et al.*, 2011). Specifically, these growth indices are associated with the leaf photosynthetic mesophyll cells (Benincasa *et al.*, 2003).

In conclusion, many physiological parameters were adversely affected when the plants were irrigated with OMW. On the other hand, the water status of the eggplants was not affected by the different levels of OMW as the plants maintained transpiration rate similar to the control values. In addition, our findings clearly suggest that the physiological responses of eggplant to OMW vary among cultivars, and the reason behind that was not the effect of OMW on the ability of the plants to absorb water. We recommend that before irrigating eggplants with OMW it is important that the sensitivity of the cultivar to OMW be taken into consideration. Moreover, the unobservable advantages of irrigating *S. melongena* with OMW for some physiological parameters do not necessarily make a

recommendation for not investigating OMW as a source for bioactive molecules that support plant growth and development. In addition, OMW is rich in nutrients such as nitrogen, phosphorus, potassium, carbon, sodium (Santos *et al.*, 2019), anthocyanins, flavinoids, and many other useful compounds (Sciubba *et al.*, 2020). Therefore, we suggest that further experiments with some manipulations can deliver potential applicable findings. For example, diluting the OMW below 25% is expected to maintain the nutritional value of OMW without arresting plant growth and development. Furthermore, investigating the physiological responses of different crops to different OMW might be needed primarily before heading to investigate cultivars responses. For example, Santos *et al.*, (2019) investigated different crop growth and development in relation to olive wastewater solution applications.

5. Author Contributions

Conceptualization, Emad Y. Bsoul
 Methodology, Shorouq Jaradat and Emad Y. Bsoul
 Validation, Emad Y. Bsoul and Shorouq Jaradat
 Formal analysis, Emad Y. Bsoul and Salman Al-Kofahi
 Investigation, Emad Y. Bsoul and Shorouq Jaradat
 Resources, Emad Y. Bsoul and Salman Al-Kofahi
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 Visualization, Emad Y. Bsoul
 Supervision, Emad Y. Bsoul
 Project administration, Emad Y. Bsoul
 All authors have read and agreed to the published version of the manuscript.

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7. Data Availability Statement

Data supporting reported results are available with the corresponding author.

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Conflict of Interests

The authors declare that there is no conflict of interests.

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