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Article

Olive Performance under the Soil Application of Humic Acid and the Spraying of Titanium and Zinc Nanoparticles under Soil Salinity Stress

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Abstract: Salinity is a major social, economic, and environmental menace in climates with low rainfall and high evapotranspiration, and it influences plant growth and causes restriction to crop production in the world. Additionally, under salinity stress, numerous physiological processes such as photosynthesis, biomass accumulation, and photosynthate transfer are also harshly lessened, and it also limits the absorption of adequate water by plants and leads to a dimension in plant water status. Therefore, the current study was conducted to investigate the soil application of humic acid (HA) at 0, 0.5, 1 and 2 kg/tree alone or in combination with the foliar spraying of 0 mg ZnO_2 + 0 mg TiO_2, 200 mg ZnO_2 + 60 mg TiO_2 and/or 300 mg ZnO_2 + 80 mg TiO_2 through the two successive seasons 2022 and 2023. The results demonstrated that the use of HA alone or in combination with the spraying of TiO_2 and ZnO_2 greatly improved the leaf chlorophyll, flower number, fruit set percentages, fruit yields in kg or in ton per hectare, fruit weight, fruit size, and fruit firmness. Additionally, the same used treatments greatly improved the fruit content from TSS and oil percentages and also the leaf mineral content from N, P and K, while they minimized the fruit drop percentage and fruit moisture content as compared to control. The most positive influence was observed with the soil implementation of 2 kg HA combined with 300 mg ZnO_2 + 80 mg TiO_2 in the two experimental seasons.

Keywords: *Olea europaea*; yield; biostimulants; nano fertilizers; oil percentages

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1. Introduction

Salinity stress is the most minatory stress that has an extreme impact on plant growth and progress, reducing the plant performance, productivity and physiochemical characteristics in desert and semi-desert areas [1–4] by reducing the absorption of essential nutrients such as Ca^{+2} and K^+ [5]. Moreover, salinity affects physiological and metabolic processes by reducing water and essential nutrient absorption through plant roots and increasing the rates of Na^+ and Cl^- ions, which can reach toxic concentrations and inhibit photosynthesis and growth [6–9].

Applying humic substances to saline soils can ameliorate sodium leaching and minimize each exchangeable sodium percentage and soil salinity [10]. Humic substances markedly increased plant growth by raising the rates of respiration, photosynthesis, oxygen and phosphorus absorption and improving the root cell development [11,12]. Additionally, HA has an important role in stimulating plant development, and it can encourage the plant's primary and secondary metabolism related to the tolerance of abiotic stress, consequently resulting in improving the plant growth [13,14]. Humic acid applied externally increased the dry weight of the shoots and roots [15], strengthened the cell membrane, maintained water absorption under osmotic stress, enhanced potassium absorption, enhanced protein and hormone synthesis, and alleviated root cell prolongation [16].

It was documented that TiO_2 NPs are helpful for the physiological, morphological, and biochemical parameters of different crops [17]. Despite being a scarce nutrient, titanium (Ti) is used as a biostimulant in plant cultivation, where it improves and speeds up biochemical processes that lead to crop growth [18]. Moreover, it is considered advantageous for plant growth, especially in raising the photosynthesis process by enhancing pollen development, iron ion activity, and plant nutrient absorption [19]. Additionally, by enhancing enzyme activity, the process of photosynthesis, nutrient intake, and stress tolerance against conditions like cold and drought, which can have a detrimental impact on crop output and quality, the utilization of Ti by little amounts via roots or leaves has improved crop performance [20]. Additionally, the usage of Ti positively affects numerous crop phonological processes including root elongation, vegetative growth, development, and resilience to biotic and abiotic stresses, which result in improving the crop properties [21].

Zinc (Zn) is a crucial element that has a paramount effect in organizing many physiological plant processes such as the synthesis of gibberellin, auxin, cytokinin, and abscisic acid, as well as the synthesis of chlorophyll, chloroplast progress, and stability of cell membrane and its structure [22]. Additionally, ZnO_2 NPs contribute to the enhancement of various crops' growth traits, photosynthesis, and yield, as well as the efficiency and nutrient content of edible plant portions and the synthesis of sugar and protein [23,24]. Additionally, Zn NPs can improve soil fertility, plant growth and crop productivity as well as relieve undesirable stresses [25–27]. It has been documented that ZnO_2 NPs can mitigate stress in mango trees [28] by improving the chlorophyll pigments, and balance of elements in cells and preserving the cell membrane solidity. Therefore, the present study was conducted to investigate the effect of the addition of HA to soil solely or in combination with the spraying of TiO_2 and ZnO_2 nano particles on improving the performance of olive trees under soil salinity stress.

2. Materials and Methods

The current study was conducted during 2022 and 2023 on ten-year-old Picual olive trees planted in the Wady El Natron region, located at a latitude of 0.371345 and longitude 30.360996 at Beheira Governorate, Egypt, at a distance of 4 × 4 m in sandy soil under a drip irrigation system. The physical and chemical characteristics of the experimental soil are shown in Table 1 [29].

To perform this experiment, seventy-two trees similar in size and growth strength were chosen and subjected to the used agricultural practices followed in the orchard. The trees were fertilized with humic acid (HA), 100% water-soluble humic acid (Qingdao Hibong Industrial Technology Co., Ltd., Qingdao City, Shandong, China), at 0, 0.5, 1 and 2 kg/tree in March 2022 and 2023 seasons, where it was added to the soil around the trees and after then covered well with the soil of the experiment. After that, the olive trees were sprayed with nanoparticles from titanium (TiO_2) at 0, 60, and 80 mg/L Ti and Zinc (ZnO_2) at 0, 200 and 300 mg/L in April (start of the season), mid-May (full bloom) and three weeks later, comparing to untreated trees (control). The design of the experiment is a Split Plot that contains two factors: the main factor is a soil application with humic acid and the submain factor is the foliar spraying of nano fertilizers (nano zinc and nano titanium). The control treatment is zero humic acid and zero ZnO_2 + zero TiO_2. The abovementioned applied treatments were investigated by studying their influence on the following parameters.

Table 1. Chemical and physical properties of the experimental soil before and after the addition of humic acid.

Parameter	**Sample**				
	Mechanical Analysis			Macronutrients	
	Before	After		Before	After
Soil depth	0–60 cm	0–60 cm	N	83 ppm	105 ppm
Sand	95.7%	95.7%	P	8.6 ppm	10.6 ppm
Silt	2%	2%	K	104 ppm	223 ppm
Clay	2.3%	2.3%	Micronutrients		
Textural class	Sand	Sand	Fe	1.63 ppm	1.88 ppm
pH	8.52	7.95	Zn	1.58 ppm	1.83 ppm
EC	4.12 ds/m	3.4 ds/m	Mn	3.54 ppm	3.64 ppm
			Cu	0.37 ppm	0.67 ppm
	Soluble Cations			Soluble anions	
Na^+	16.75 Meq/L	11.43 Meq/L	Cl^-	20.5 Meq/L	14.5 Meq/L
K^+	9.14 Meq/L	10.44 Meq/L	HCO_3^-	12.4 Meq/L	10.4 Meq/L
Ca^+	8.0 Meq/L	6.8 Meq/L	CO_3^{2-}	0.0 Meq/L	0.0 Meq/L
Mg^+	7.2 Meq/L	4.5 Meq/L	SO_4^{2-}	8.19 Meq/L	9.19 Meq/L

2.1. Leaf Total Chlorophyll (SPAD)

It was measured in the fresh leaves by a Minolta chlorophyll meter (SPAD-502; Konica Minolta, Osaka, Japan) by taking 10 readings from the mature leaves in the middle part of the shoots around the trees. The flower number per m^2 was accounted for.

2.2. Flower Number, Fruit Set and Fruit Drop Percentages

To account for the fruit set and fruit drop percentages, five branches from each side of each replicate (tree) were chosen and labelled carefully, accounting for the number of flowers, and then the fruit set % was calculated according to Equation (1).

$$\text{Fruit set \%} = \frac{\text{No.of fruitlets}}{\text{No.of perfect flowers}} \times 100 \tag{1}$$

Fruit drop (%) was estimated by calculating the difference between the number of set fruits and the dropped fruits using Equation (2).

$$\text{Fruit drop (\%)} = \frac{\text{No.of dropped fruits}}{\text{No.of set fruits}} \times 100 \tag{2}$$

2.3. Fruit Yield

In October (2022–2023), the yield of each tree was estimated as fruit weight in kg and was then estimated for hectares in a ton by multiplying the yield of each tree * number of trees.

2.4. Fruit Quality Attributes

Forty fruits from each tree/replicate were collected immediately after harvesting and transported to the lab to determine the fruits' physical and chemical characteristics.

2.4.1. Fruit Physical Characteristics

Fruit fresh weight, flesh weight, and seed weight were estimated by calculating the average weight of 40 fruits from each tree/replicate. Average fruit length and diameter were measured using a Digital Vernier Caliper (Suzhou Sunrix Precision Tools Co., Ltd., Suzhou, Jiangsu, China). Fruit firmness was estimated by using a Magness and Taylor pressure tester with a 7/18-inch plunger (mod. FT 02 (0-2 Lb., Via Reale, 63-48011

Alfonsine, Italy). The fruit moisture content was determined by measuring the fresh weight of 50 fruits, and they were dried until a constant weight, and the moisture content was the difference between the two fresh and dry weights of fruits.

2.4.2. Fruit Chemical Characteristics

Total soluble solids from the fresh-cut olive fruits were measured using a handheld digital refractometer (ATAGO CO., LTD., Tokyo, Japan).

Oil content: Samples from the flesh fruit were dried and then ground, and 2 g was weighed, filtered and placed in the Soxhlet apparatus using petroleum ether [30]. The oil percentage was calculated using Equation (3):

$$\text{Oil}\ \% = \frac{\text{weight of extracted oil}}{\text{weight of sample}} \times 100 \quad (3)$$

2.5. Leaf Minerals Status

After harvesting the fruits in the 2022 and 2023 seasons, 40 leaves from the middle part of the shoots were harvested from each tree/replicate. The leaves were washed very well with tap water and then distilled water. They were dried at 70 °C until constant weight and then ground and digested using H_2SO_4 and H_2O_2 until the solution became clear. The nitrogen content (N) was determined using the micro Kjeldahl method [31]. The phosphorus content (P) was measured using the vanadomolybdo method [32]. The potassium content (K) was determined using a flame photometer [33].

2.6. Statistical Analysis

The results were obtained using statistical analysis with Split Plot Design using CoHort Software 6.311 (Pacific Grove, CA, USA), and the least significant difference (LSD) at 0.05% was used to compare the means of treatments [34].

3. Results

3.1. Leaf Total Chlorophyll, Flower Number and Fruit Set Percentage

The soil application of HA combined with the folia spraying of TiO_2 and ZnO_2 greatly increased the leaf chlorophyll content compared to the control. Additionally, the soil application of HA at 2 kg per tree combined with 300 mg ZnO_2 + 80 mg TiO_2 gave the highest increments (27.24 and 32.1%) in the first and second seasons (Table 2). It was also improved by the application of 2 kg HA combined with 200 mg ZnO_2 + 60 mg TiO_2 (27 and 29.94%) as well as by 1 kg HA combined with 300 mg ZnO_2 + 80 mg TiO_2 (25.51 and 28.34%) in the first and second seasons. The flower number was notably increased by the soil implementation of 2 kg HA combined with the spraying of 300 mg ZnO_2 + 80 mg TiO_2 (31.44 and 32.72%) or 200 mg ZnO_2 + 60 mg TiO_2 (29.75 and 28.85%) compared with control. Moreover, it was also enhanced using 1 kg HA combined with 300 mg ZnO_2 + 80 mg TiO_2 (29.44 and 31.05%) compared to control. Additionally, the highest fruit set percentages were markedly better by the use of 2 kg HA in combination with the spraying of 300 mg ZnO_2 + 80 mg TiO_2 (38.59 and 42.34%) and with 200 mg ZnO_2 + 60 mg TiO_2 (31.33 and 27.54%) and also by 2kg HA combined with 300 mg ZnO_2 + 80 mg TiO_2 (29.87 and 36.13) compared to untreated trees.

Table 2. The combined application of HA soil application with the spraying of TiO_2 and ZnO_2 nanoparticles on the leaf total chlorophyll, flower number and fruit set percentages of olive during the 2022 and 2023 seasons.

Treatments		Leaf Chlorophyll (SPAD)		Flower Number		Fruit Set %	
HA	Fertilizers	2022	2023	2022	2023	2022	2023
0 (Control)	0 mg ZnO_2 + 0 mg TiO_2 (Control)	54.75d ±2.99	55.00f ±1.82	785.00c ±55.07	832.50b ±69.94	3.31d ±0.25	3.50d ±0.14
	200 mg ZnO_2 + 60 mg TiO_2	61.00c ±4.08	61.75e ±4.64	795.00c ±45.09	846.50b ±46.71	3.45cd ±0.13	3.62d ±0.2
	300 mg ZnO_2 + 80 mg TiO_2	63.75bc ±3.30	65.00de ±2.16	822.50c ±33.04	875.00b ±55.68	3.59cd ±0.16	3.57d ±0.1
0.5 kg	0 mg ZnO_2 + 0 mg TiO_2	65.50b ±1.73	68.00d ±2.16	845.00c ±73.26	892.50b ±42.72	3.60cd ±0.1	3.69d ±0.14
	200 mg ZnO_2+ 60 mg TiO_2	65.50b ±1.00	69.00cd ±2.94	903.75bc ±18.87	927.50b ±17.08	3.55cd ±0.1	3.78d ±0.4
	300 mg ZnO_2 + 80 mg TiO_2	66.25b ±2.63	74.00bc ±2.71	997.50ab ±59.09	1112.50a ±85.39	3.60cd ±0.16	4.09d ±0.2
1 kg	0 mg ZnO_2 + 0 mg TiO_2	71.75a ±2.06	74.25bc ±2.87	1052.50a ±61.85	1137.50a ±62.91	3.62cd ±0.12	4.00d ±0.14
	200 mg ZnO_2 + 60 mgTiO_2	73.75a ±2.22	73.50bc ±1.29	1087.50a ±85.39	1165.00a ±44.35	3.91cd ±0.28	4.81c ±0.5
	300 mg ZnO_2 + 80 mg TiO_2	73.50a ±1.29	76.75ab ±0.96	1112.50a ±103.08	1207.50a ±57.37	4.72b ±0.22	5.48b ±0.2
2 kg	0 mg ZnO_2 + 0 mg TiO_2	73.00a ±2.16	74.75b ±0.22	1060.00a ±77.89	1167.50a ±106.89	3.95c ±0.21	3.80d ±0.3
	200 mg ZnO_2 + 60 mg TiO_2	75.00a ±0.82	78.50ab ±2.65	1117.50a ±103.72	1170.00a ±67.82	4.82b ±0.32	4.83c ±0.4
	300 mg ZnO_2 + 80 mg TiO_2	75.25a ±2.22	81.00a ±1.15	1145.00a ±42.03	1237.50a ±75	5.39a ±0.60	6.07a ±0.3
$LSD_{0.05}$		3.08	3.97	109.08	98.96	0.38	0.42

Means marked with the same letters do not differ significantly at 0.05.

3.2. Fruit Drop Percentage, and Fruit Yield in kg or in Ton

The soil application of 2 kg HA combined with 300 mg ZnO_2 + 80 mg TiO_2 (5.18 and 4.20%) and 200 mg ZnO_2 + 60 mg TiO_2 (3.74 and 2.67%) and the soil application of 2 kg per tree HA with 300 mg ZnO_2 + 80 mg TiO_2 (3.13 and 3.04%) significantly reduced the fruit drop percentages compared to the control (Table 3). Fruit yields in kg per tree and in ton per hectare were considerably increased by the use of combined application of 2 kg HA with the spraying of 300 mg ZnO_2 + 80 mg TiO_2 (28.11 and 29.79%) or with 200 mg ZnO_2 + 60 mg TiO_2 (18.32 and 21.43%) in the two seasons.

Table 3. The combined application of HA soil application with the spraying of TiO_2 and ZnO_2 nanoparticles on fruit drop percentages, fruit yield in kg per tree or in ton per hectare of olive during the 2022 and 2023 seasons.

Treatments		Fruit Drop %		Fruit Yield (kg/Tree)		Yield (Ton/H)	
HA	**Fertilizers**	**2022**	**2023**	**2022**	**2023**	**2022**	**2023**
0 (Control)	0 mg ZnO_2 + 0 mg TiO_2 (Control)	97.64a ±0.28	95.90a ±0.69	39.00e ±2.58	41.25d ±1.5	23.40e ±1.55	24.75d ±0.90
	200 mg ZnO_2 + 60 mg TiO_2	97.41ab ±0.59	95.16b ±0.58	40.00de ±1.63	42.50d ±2.08	24.00de ±0.98	25.50d ±1.25
	300 mg ZnO_2 + 80 mg TiO_2	96.33bc ±0.43	94.44b–d ±0.34	41.25c–e ±1.50	43.00cd ±2.58	24.75c–e ±0.90	25.80cd ±1.55
0.5 kg	0 mg ZnO_2 + 0 mg TiO_2	96.60a–c ±0.67	94.92bc ±0.62	43.00b–e ±0.42	44.25cd ±1.70	25.65b–e ±0.75	26.55cd ±1.02
	200 mg ZnO_2+ 60 mg TiO_2	96.59a–c ±0.32	94.89bc ±0.41	43.50b–e ±1.29	45.75cd ±1.71	26.10b–e ±0.77	27.45cd ±1.02
	300 mg ZnO_2 + 80 mg TiO_2	95.51cd ±0.31	93.38e ±0.41	44.00b–e ±0.82	51.00b ±2.94	26.40b–e ±0.49	30.60b ±1.77
1 kg	0 mg ZnO_2 + 0 mg TiO_2	95.50cd ±0.50	94.40b–d ±0.17	42.75b–e ±1.26	47.50bc ±2.08	25.80b–e ±0.49	28.50bc ±1.70
	200 mg ZnO_2 + 60 mgTiO_2	94.58de ±0.48	93.67de ±0.1	45.00b–d ±1.15	50.75b 1.71	27.00b–d ±0.69	30.45b ±1.02
	300 mg ZnO_2 + 80 mg TiO_2	94.34e ±0.53	92.98e ±0.33	45.75bc ±1.71	52.00b ±2.83	27.45bc ±1.02	31.20b ±1.70
2 kg	0 mg ZnO_2 + 0 mg TiO_2	95.74c ±0.75	94.17cd ±0.37	43.00bc–e ±1.41	50.00b ±2.45	25.80b–e ±0.85	30.00b ±1.47
	200 mg ZnO_2 + 60 mg TiO_2	93.99e ±0.89	93.34e ±0.59	47.75b ±1.71	52.50b ±2.08	28.65b ±0.57	31.50b ±1.35
	300 mg ZnO_2 + 80 mg TiO_2	92.58f ±0.67	91.87f ±0.82	54.25a ±6.24	58.75a ±5.19	32.55a ±3.74	35.25a ±3.11
$LSD_{0.05}$		0.84	0.60	3.45	3.44	2.02	2.06

Means marked with the same letters do not differ significantly at 0.05.

3.3. Fruit Quality

The data in Table 4 showed that the fruit weight and fruit flesh weight were markedly increased by the addition of 2 kg HA to the soil with the combination of 300 mg ZnO_2 + 80 mg TiO_2 (22.81 and 27.94%) (25.12 and 33.04%) and with 200 mg ZnO_2 + 60 mg TiO_2 (23.29 and 27.94%) (26.19 and 31.53%) compared to the control, respectively. The differences between the effect of the soil application of 1 or 0.5 kg from HA in combination with the spraying of 200 mg ZnO_2 + 60 mgTiO_2 and 300 mg ZnO_2 + 80 mg TiO_2 and with the usage of 2 or 1 kg per tree on the fruit or the flesh weights were insignificant in the two seasons. All the applied treatments, even the soil application of HA at 2, 1 and 0.5 kg only or in combination with the foliar spraying of 300 mg ZnO_2 + 80 mg TiO_2 or 200 mg ZnO_2 + 60 mg TiO_2, did not have a notable impact on the seed weight compared to control.

Table 4. The combined application of HA soil application with the spraying of TiO_2 and ZnO_2 nanoparticles on the fruit, flesh and seed weights of olive during the 2022 and 2023 seasons.

Treatments		Fruit Weight (g)		Flesh Weight (g)		Seed Weight (g)	
HA	**Fertilizers**	**2022**	**2023**	**2022**	**2023**	**2022**	**2023**
0 (Control)	0 mg ZnO_2 + 0 mg TiO_2 (Control)	2.47b ±0.1	2.45d ±0.06	1.55cd ±0.13	1.52f ±0.09	0.92a ±0.1	0.92a ±0.1
	200 mg ZnO_2 + 60 mg TiO_2	2.47b ±0.05	2.60cd ±0.08	1.50d ±0.08	1.67ef ±0.12	0.97a ±0.1	0.92a ±0.12
	300 mg ZnO_2 + 80 mg TiO_2	2.65b ±0.13	2.95b 0.25	1.55cd ±0.13	1.77de ±0.17	1.10a ±0.16	1.17a ±0.21
0.5 kg	0 mg ZnO_2 + 0 mg TiO_2	2.62b ±0.1	2.85bc ±0.19	1.72b–d ±0.19	1.87c–e ±0.15	0.90a ±0.27	0.97a ±0.17
	200 mg ZnO_2+ 60 mg TiO_2	2.77ab ±0.26	3.10ab ±0.08	1.62cd ±0.12	1.92cd ±0.09	1.15a ±0.35	1.17a ±0.05
	300 mg ZnO_2 + 80 mg TiO_2	2.97ab ±0.24	3.00ab ±0.29	1.85a–c ±0.21	2.10a–c ±0.18	1.12a ±0.34	0.90a ±0.14
1 kg	0 mg ZnO_2 + 0 mg TiO_2	2.70ab ±0.24	3.12ab ±0.09	1.70b–d ±0.16	2.02a–d ±0.17	1.00a ±0.42	1.10a ±0.11
	200 mg ZnO_2 + 60 mg TiO_2	2.90ab ±0.11	3.07ab ±0.30	1.87a–c ±0.15	2.02a–d ±0.12	1.02a ±0.12	1.05a ±0.24
	300 mg ZnO_2 + 80 mg TiO_2	2.97ab ±0.39	3.17ab ±0.22	1.97ab ±0.09	1.97b–d ±0.22	1.00a ±0.39	1.20a ±0.22
2 kg	0 mg ZnO_2 + 0 mg TiO_2	2.82ab ±0.27	3.12ab ±0.19	1.70bcd ±0.16	2.05a–d ±0.06	1.12a ±0.30	1.07a ±0.12
	200 mg ZnO_2 + 60 mg TiO_2	3.22a ±0.17	3.40a ±0.27	2.10a ±0.08	2.22ab ±0.09	1.12a ±0.15	1.17a ±0.19
	300 mg ZnO_2 + 80 mg TiO_2	3.20a ±0.28	3.40a ±0.22	2.07a ±0.15	2.27a ±0.12	1.12a ±0.15	1.12a ±0.15
$LSD_{0.05}$		0.34	0.26	0.22	0.18	0.41	0.22

Means marked with the same letters do not differ significantly at 0.05.

The data in Table 5 demonstrated that the effect of the applied treatments on the ratio between flesh and fruit weight was insignificant during the two experimental seasons. The combination of the soil utilization of HA at 0.5, 1 and 2 kg/tree only or in combination with the spraying of 200 mg ZnO_2 + 60 mg TiO_2 or 300 mg ZnO_2 + 80 mg TiO_2 improved the fruit length and fruit diameter in both experimental seasons. Moreover, the highest increments were obtained with the usage of 2 kg HA combined with the spraying of 200 mg ZnO_2 + 60 mg TiO_2 or 300 mg ZnO_2 + 80 mg TiO_2 in the two seasons. The fruit firmness was greatly ameliorated by the soil utilization of 0.5, 1 and 2 kg from HA alone or after the combination with 300 mg ZnO_2 + 80 mg TiO_2 (29.22 and 25.40%) and 200 mg ZnO_2 + 60 mg TiO_2 (23.59 and 18.93%). The treatment that gave the highest value from the fruit firmness was the usage of 2 kg HA in combination with the spraying of 300 mg ZnO_2 + 80 mg TiO_2.

Table 5. The combined application of HA soil application with the spraying of TiO_2 and ZnO_2 nanoparticles on the flesh/fruit ratio, fruit length, diameter, and firmness of olive during the 2022 and 2023 seasons.

Treatments		Flesh/Fruit Weight (g)		Fruit Length (cm)		Fruit Diameter (cm)		Fruit Firmness (Ib/inch2)	
HA	**Fertilizers**	**2022**	**2023**	**2022**	**2023**	**2022**	**2023**	**2022**	**2023**
0 (Control)	0 mg ZnO_2 + 0 mg TiO_2 (Control)	0.62a ±0.04	0.62ab ±0.04	2.03d ±0.03	2.11e ±0.03	1.26b ±0.12	1.40e ±0.08	11.82f ±0.4	12.42f ±0.3
	200 mg ZnO_2 + 60 mg TiO_2	0.60a ±0.03	0.64ab ±0.05	2.06d ±0.01	2.11e 0.03	1.42ab ±0.09	1.44de ±0.05	11.95f ±0.5	12.65ef ±0.4
	300 mg ZnO_2 + 80 mg TiO_2	0.58a ±0.05	0.60b ±0.03	2.11cd ±0.06	2.22de ±0.06	1.50a ±0.08	1.58bcd ±0.10	13.30d ±0.2	13.80d ±0.4
0.5 kg	0 mg ZnO_2 + 0 mg TiO_2	0.66a ±0.09	0.66ab ±0.09	2.03d ±0.05	2.21de ±0.06	1.42ab ±0.09	1.52cde ±0.09	12.42e ±0.2	13.40de ±0.3
	200 mg ZnO_2+ 60 mg TiO_2	0.59a ±0.09	0.62ab ±0.08	2.17cd ±0.17	2.23de ±0.05	1.50a ±0.08	1.57bcd ±0.09	12.72e ±0.5	13.30de ±0.2
	300 mg ZnO_2 + 80 mg TiO_2	0.62a ±0.08	0.70a ±0.02	2.19bcd ±0.08	2.28cd ±0.03	1.62a ±0.09	1.57bcd ±0.05	13.72d ±0.3	14.00cd ±0.3
1 kg	0 mg ZnO_2 + 0 mg TiO_2	0.64a ±0.11	0.65ab ±0.03	2.20bcd ±0.18	2.31cd ±0.08	1.52a ±0.09	1.52cde ±0.09	12.75e ±0.3	13.50de ±0.4
	200 mg ZnO_2 + 60 mg TiO_2	0.65a ±0.04	0.66ab ±0.09	2.14cd ±0.05	2.37bc ±0.09	1.62a ±0.17	1.72ab ±0.05	14.42c ±0.1	14.67bc ±0.4
	300 mg ZnO_2 + 80 mg TiO_2	0.67a ±0.09	0.62ab ±0.01	2.24bc ±0.06	2.40bc ±0.08	1.52a ±0.12	1.70ab ±0.08	14.77c ±0.5	15.00b ±0.6
2 kg	0 mg ZnO_2 + 0 mg TiO_2	0.61a ±0.08	0.66ab ±0.03	2.25bc ±0.19	2.21de ±0.06	1.52a ±0.09	1.60bcd ±0.03	14.40c ±0.2	14.10cd ±0.4
	200 mg ZnO_2 + 60 mg TiO_2	0.65a ±0.03	0.66ab ±0.05	2.35b ±0.13	2.47b ±0.09	1.67a ±0.09	1.67abc ±0.09	15.47b ±0.1	15.32b ±0.5
	300 mg ZnO_2 + 80 mg TiO_2	0.65a ±0.02	0.67ab ±0.03	2.60a ±0.14	2.60a ±0.08	1.67a ±0.15	1.80a ±0.08	16.70a ±0.4	16.65a ±0.5
$LSD_{0.05}$		0.11	0.05	0.11	0.10	0.15	0.10	0.44	0.65

Means marked with the same letters do not differ significantly at 0.05.

The listed data in Table 6 cleared that TSS percentages were improved by the addition of HA at 0.5, 1 and 2 kg alone or with the combination of the spraying of 200 mg ZnO_2 + 60 mg TiO_2 and with 300 mg ZnO_2 + 80 mg TiO_2. The best increments in the fruit content from the TSS % resulted from the utilization of 2kg HA combined with 300 ZnO_2 + 80mg TiO_2 (18.43 and 17.36%), respectively, in the first and the second seasons. The oil percentage was greatly increased by the addition of 2 kg HA in combination with the spraying of 200 mg ZnO_2 + 60 mg TiO_2 (14.80 and 15.05%) or with 300 mg ZnO_2 + 80 mg TiO_2 (20.39 and 21.94%) and also by the application of 1 kg HA with 300 mg ZnO_2 + 80 mg TiO_2 (13.33 and 15.05%) or with 200 mg ZnO_2 + 60 mg TiO_2 (11.77 and 11.13%) in the first and second seasons, respectively. The results proved that there is a converse relation between the fruit oil content and the moisture content, where the highest percentage of the moisture content in the fruit was high with control treatment, while it was remarkably reduced by the addition of 0.5, 1 or 2 kg from HA alone or after combination with the spraying of 300 mg ZnO_2 + 80 mg TiO_2 and 200 mg ZnO_2 + 60 mg TiO_2. The lowest percentage for the moisture content was obtained with the addition of 2 kg HA combined with the spraying of 300 mg ZnO_2 + 80 mg TiO_2 (29.75 and 29.32%) in the first and second seasons.

Table 6. The combined application of HA soil application with the spraying of TiO_2 and ZnO_2 nanoparticles on fruit content from TSS %, oil % and moisture content of olive during the 2022 and 2023 seasons.

Treatments		TSS%		Oil %		Moisture Content %	
HA	**Fertilizers**	**2022**	**2023**	**2022**	**2023**	**2022**	**2023**
0 (Control)	0 mg ZnO_2 + 0 mg TiO_2 (Control)	14.07f ±0.22	14.42e ±0.22	16.12e ±0.38	16.37f ±0.33	74.05a ±1.45	71.27a ±2.33
	200 mg ZnO_2 + 60 mg TiO_2	14.55e ±0.17	14.35e ±0.26	16.40e ±0.41	16.47f ±0.36	70.95b ±2.07	66.32b ±1.48
	300 mg ZnO_2 + 80 mg TiO_2	14.97d ±0.29	15.17d ±0.40	16.90de ±0.26	17.42de ±0.29	70.32b ±1.44	66.90b ±1.15
0.5 kg	0 mg ZnO_2 + 0 mg TiO_2	15.20d ±0.32	15.15d ±0.51	17.00de ±0.29	16.90ef ±0.26	69.43bc ±2.04	63.55c ±1.30
	200 mg ZnO_2+ 60 mg TiO_2	15.40d ±0.32	15.80c ±0.45	17.10de ±0.42	17.22de ±0.45	69.02bc ±2.34	63.27c ±0.07
	300 mg ZnO_2 + 80 mg TiO_2	16.22bc ±0.31	16.47b ±0.17	17.40cd ±0.42	17.67cde ±0.17	65.50cd ±1.18	61.60cd ±1.22
1 kg	0 mg ZnO_2 + 0 mg TiO_2	15.47d ±0.22	15.75c ±0.33	17.60cd ±0.35	17.72cde ±0.24	68.11bc ±2.41	62.80cd 0.28
	200 mg ZnO_2 + 60 mg TiO_2	16.55b ±0.13	16.45b ±0.10	18.27bc ±0.39	18.42c ±0.42	62.97de ±2.03	60.68d ±0.16
	300 mg ZnO_2 + 80mg TiO_2	16.42b ±0.17	16.60b ±0.22	18.60b ±0.63	19.27b ±0.19	60.62ef ±2.94	58.44e ±2.21
2 kg	0 mg ZnO_2 + 0 mg TiO_2	15.92c ±0.30	16.17bc ±0.21	17.70cd ±0.39	17.85cd ±0.21	66.26cd ±3.11	63.02cd ±1.31
	200 mg ZnO_2 + 60mg TiO_2	16.72b ±0.1	16.70b ±0.24	18.92b ±0.50	19.27b ±0.19	59.02fg ±3.07	56.90ef ±0.56
	300 mg ZnO_2 + 80 mg TiO_2	17.25a ±0.31	17.45a ±0.13	20.25a ±0.51	20.97a ±0.89	57.07g ±3.60	55.11f ±2.33
$LSD_{0.05}$		0.38	0.44	0.69	0.59	2.78	1.81

Means marked with the same letters do not differ significantly at 0.05.

3.4. Leaf Mineral Content from Macronutrients

Table 7 showed that the leaf mineral content including N, P and K was markedly increased by the soil addition of HA at 0.5, 1 and 2 HA only or with the combination of 200 mg ZnO_2 + 60 mg TiO_2 and 300 mg ZnO_2 + 80 mg TiO_2 in both experimental seasons. The treatment that gave the highest values from these nutrients was obtained from the soil addition of HA at 2 kg combined with the spraying of 300 mg ZnO_2 + 80 mg TiO_2 where it gave increments in N % (17.61 and 17.88%), P (39.1 and 31.43%) and K (23.62 and 17.69%) in both experimental seasons.

Table 7. The combined application of HA soil application with the spraying of TiO_2 and ZnO_2 nanoparticles on leaf mineral content from N, P and K percentages of olive during 2022 and 2023 seasons.

Treatments		N%		P%		K%	
HA	**Fertilizers**	**2022**	**2023**	**2022**	**2023**	**2022**	**2023**
0 (Control)	0 mg ZnO_2 + 0 mg TiO_2 (Control)	1.45e ±0.02	1.47e ±0.02	0.39d ±0.03	0.48f ±0.03	0.97d ±0.05	1.07e ±0.02
	200 mg ZnO_2 + 60 mg TiO_2	1.46e ±0.02	1.48e ±0.02	0.40d ±0.02	0.48f ±0.02	1.00d ±0.04	1.10de ±0.03
	300 mg ZnO_2 + 80 mg TiO_2	1.49e ±0.03	1.50e ±0.01	0.42d ±0.02	0.49ef ±0.03	1.07c ±0.01	1.13d ±0.02
0.5 kg	0 mg ZnO_2 + 0 mg TiO_2	1.54d ±0.01	1.56d ±0.04	0.47c ±0.04	0.51def ±0.02	1.07c ±0.03	1.13d ±0.03
	200 mg ZnO_2 + 60 mg TiO_2	1.59d ±0.02	1.60cd ±0.03	0.47c ±0.03	0.54cde ±0.03	1.07c ±0.03	1.14d ±0.01
	300 mg ZnO_2 + 80 mg TiO_2	1.62c ±0.04	1.63c ±0.02	0.51c ±0.02	0.60b ±0.02	1.13c ±0.02	1.18c ±0.02
1 kg	0 mg ZnO_2 + 0 mg TiO_2	1.56d ±0.02	1.59cd ±0.05	0.47c ±0.04	0.55cd ±0.02	1.09c ±0.03	1.13d ±0.03
	200 mg ZnO_2 + 60 mg TiO_2	1.65c ±0.01	1.65c ±0.01	0.55b ±0.02	0.62b ±0.03	1.14c ±0.02	1.19c ±0.03
	300 mg ZnO_2 + 80 mg TiO_2	1.66c ±0.02	1.69b ±0.03	0.56b ±0.02	0.65b ±0.03	1.21b ±0.04	1.23b ±0.02
2 kg	0 mg ZnO_2 + 0 mg TiO_2	1.58d ±0.02	1.60cd ±0.04	0.48c ±0.01	0.56c ±0.01	1.10c 0.03	1.14d ±0.03
	200 mg ZnO_2 + 60 mg TiO_2	1.71b ±0.03	1.73b ±0.02	0.61a ±0.03	0.63b ±0.03	1.24ab 0.03	1.25b ±0.03
	300 mg ZnO_2 + 80 mg TiO_2	1.76a ±0.03	1.79a ±0.04	0.64a ±0.03	0.70a ±0.04	1.27a ±0.03	1.30a ±0.04
$LSD_{0.05}$		0.03	0.04	0.04	0.03	0.05	0.03

Means marked with the same letters do not differ significantly at 0.05.

4. Discussion

From the comparison between the composition of the soil before and after the addition of humic aid, it was observed that the electrical conductivity and the concentrations of Na^+, K^+, Ca^+ and Mg^+ as well as the concentrations of the anions Cl^-, HCO_3^-, CO_3^{2-} and SO_4^{2-} were decreased, which was probably because humic acid raises the soil's capacity to hold water. Additionally, from the same table, it was also observed that the pH was decreased, which is reflected in the increased availability of nutrients from macronutrients such as N, P and K or micronutrients like Fe, Zn, Mn, and Cu, which ultimately improved the vegetative growth and productivity. From this comparison, there is a clear influence of the application of humic acid in improving soil fertility. These results were formerly clarified by a lot of authors, where humic substances improve soil fertility by raising the water-holding ability [35], changing the soil's physical, chemical, and biological structure [36], improving the permeability of plant membranes, and encouraging the absorption of elements under salinity [37]. HA raises the availability of important elements for the plant's vegetative growth such as nitrogen, phosphorous and potassium [38] and raises the soil's water-holding capability through high water absorption [39]. Applying HA may lead to minimizing the chlorophyll decay and boosting the leaf chlorophyll content under salinity conditions by increasing the cell membrane stability and boosting the absorption of nutrients such as nitrogen which is related to the chlorophyll synthesis [40], and it can improve the leaf water content under osmotic stress [13]. Furthermore, HA is an organic fertilizer that can positively impact plant growth and enhance the uptake of nutrients such as calcium, magnesium, phosphorous, potassium, nitrogen, and potassium [41]. The soil addition of HA as potassium humate at 75, 100, 125 and 150 on cv. Red Delicious apple trees greatly raised the percentages of fruit set and retention as well as fruit yield and leaf mineral content from macro and micronutrients, and greatly minimized the fruit drop percentages [42]. The addition of HA at 0 and 75 g per tree to olive trees markedly enhanced the fruit productivity, soluble solids, total carbohydrates and oil percentage in the fruits [43]. Similarly, applying HA on lime trees at 10, 20 or 30 mL·tree^{-1} remarkably increased the available nutrients in the soil such as N, P, K, Ca, Mg, Fe, Mn, Zn, Cu, and b. Moreover, it also improved the soil microbial activity, vegetative growth, tree canopy, leaf chlorophyll, number and weight of fruits and, consequently, the final productivity, as well as the fruit content from juice and soluble solids [44].

Exo spraying of TiO_2 increased the uptake of macro- and micro-nutrients and improved the plant height, leaf photosynthesis rate and leaf number, while it reduced the undesirable impacts of salinity [45]. Additionally, the application of TiO_2 NPs treatments might raise the plant nitrogen content [46,47]. Furthermore, under salinity, it was noticed that some plant species treated with TiO_2 NPs ameliorated the photosynthetic rates, chlorophyll fluorescence, and soluble sugars [17,48] and promoted crop productivity and oil production [49]. Additionally, TiO_2 has been shown to facilitate the absorption of essential nutrients, including iron, potassium, calcium, magnesium, and nitrogen [50]. Additionally, the application of TiO_2 NPs stimulated the photosynthesis process in plants, and growth parameters are also positively related to the absorption of essential elements in the treated plants under salinity conditions [9,45]. Spraying mango cv. Keitt with TiO_2 at 40, 60 and 80 mg/L improved the number, length, and thickness of shoots, leaf area surface, and leaf chlorophyll compared to untreated trees. Moreover, the applied treatments also ameliorated the fruit set percentages, fruit yields, fruit weight, size, length, and diameter. Additionally, the sprayed trees gave fruit with a high content from soluble solids, VC, carotene content, total and reduced sugars, as well as high nutritional content from nitrogen, potassium and phosphorous [51].

Concerning the influence of the spraying of Zn, it was stated that Zn is also an essential micronutrient for all the plants that participate in the synthesis of chlorophyll and participates in many cellular processes and the synthesis of phytohormones like auxin, cytokinin, and gibberellin [52]. ZnO_2 NPs affect fruit quality and tryptophan synthesis to modulate the effects of auxin [53,54]. Since Zn is necessary for the production of protein, chlorophyll, and indole acetic acid as well as for maintaining the integrity of the cell membrane by preventing the plant from absorbing too much Na+ and Cl, the exo spraying of ZnO_2 NPs enhanced the growth and physiological parameters of the plants under NaCl stress [55]. ZnO_2 NPs also play a crucial role in enhancing chlorophyll formation and photosynthetic activity [56,57] and mitigating salt stress in plants [58]. Moreover, ZnO_2 NPs are involved in improving the growth attributes, photosynthesis, yield, biomass production, nutrient uptake efficacy, sugar, and total nitrogen in numerous crops [23,59]. Spraying of peach cv. Florida prince with Zn NPs at 2.5, 5 and 7.5 mg/L notably increased the shoot diameter, leaf area surface, leaf total chlorophyll, flower percentage, fruit productivity, fruit weight, length, diameter, size, and firmness. Additionally, the sprayed Zn NPs notably raised the fruit content from soluble solids, total, reduced and non-reduced sugar percentages, anthocyanin and vitamin C, while they minimized the fruit content from acidity compared to untreated trees [60]. Treating pomegranate cv. Wonderful by ZnO_2 NPs at 500 and 1000 ppm enhanced the shoot length, leaf chlorophyll, leaf area, leaf number per shoot, leaf content from N, P, K, Ca, Zn, and B, fruit set and fruit preservation percentages as well as the fruit yield compared to the control [61].

5. Conclusions

From the obtained results, it could be concluded that the use of HA has a functional influence on reducing the undesirable effect of salinity because it improves soil fertility and increases the nutrients in the soil. Additionally, the spraying of TiO_2 and ZnO_2 has a great influence on reducing the side effects of salinity. The merged effect of the soil addition of 2 kg per tree HA combined with the spraying of 300 mg ZnO_2 + 80 mg TiO_2 significantly improved the vegetative growth, productivity, and fruit quality attributes as well as leaf mineral content from macronutrients rather than the other applied treatments. Moreover, more genetic studies should be performed to define the genes that are responsible for increasing the tolerance of olive trees to salinity.

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