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Evaluating Seawater and Wood Distillate for Sustainable Hydroponic Cultivation: Implications for Crop Growth and Nutritional Quality

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Abstract: The adoption of innovative cultivation methods, such as hydroponics and aeroponics, is gaining attention due to the unprecedented demand for food that an increasing population is posing on agricultural systems, exacerbating the pressure on already limited arable land. Seeking sustainable and circular economy solutions is imperative, aiming to optimize water consumption and enhance crop yields and quality without resorting to synthetic chemical fertilizers. This study investigated the use of seawater at various concentrations as a base for nutrient solutions, with and without the addition of a natural biostimulant, wood distillate (*WD*). Four seawater (SW) concentrations (0, 3, 6, and 12%) and two wood distillate concentrations (0 and 0.2%) were applied to assess their impacts on lettuce growth. Findings reveal that seawater at low concentrations (<6%) serves as an effective water-saving strategy, despite the reduction in the plant ascorbic acid contents. The addition of *WD* did not inflate growth; in fact, the results obtained are comparable to that of the controls for each concentration of seawater, except at the highest concentration (12% SW), resulting in reduced fresh leaf weights and root areas. Significantly, there was a notable increase in the ascorbic acid contents in all plants grown with *WD*. Moreover, the *WD* increased the leaf concentrations in Ca, Mg, P, and K, indicating the higher nutritional value of the crop. This research highlights the potential of combining seawater and *WD* for sustainable and efficient plant cultivation, suggesting new strategies for exploration across diverse plant species and hydroponic applications.

Keywords: crop production; lettuce plants; pyroligneous acid; soilless cultivation; sustainable agriculture; wood vinegar

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1. Introduction

We are witnessing a climate crisis (CC) characterized by long-term alterations in temperature, precipitation, and other atmospheric conditions and patterns at the global level. A major contributor to CC is the rise in greenhouse gas concentrations in the atmosphere, largely resulting from human activities such as burning fossil fuels and deforestation [1]. These environmental changes impact agriculture in several ways: coastal erosion, the heightened frequency and severity of coastal flooding, and reductions in cultivated areas, where marine intrusion alters the average salinity of coastal areas, posing challenges to crop cultivation [2,3]. It is estimated that by 2030, in Europe alone, there will be a reduction of approximately 4.2 million ha of cultivated areas, undoubtedly impacting food production [4].

In this context, numerous strategies have been implemented to address the shortage of cultivable areas, such as hydroponic and aeroponic systems [5–7]. These cultivation

methods are suitable for urban agriculture and land-limited areas [8,9]. Vertical hydroponic systems optimize space utilization, enabling the growth of various crops in defined areas [10]. Hydroponic cultivation lies in the nutrient solution that facilitates rapid plant growth by providing the necessary nutrients [11]. The use of seawater (SW) in hydroponic cultivation can be a sustainable alternative to the use of freshwater [12]. Seawater is rich in nutrients such as magnesium, calcium, and potassium, which are crucial for the growth of certain plants and algae. While seawater does contain water, its high salinity generally limits its direct use for irrigation, making its mineral content the more valuable component in such agricultural practices [13–15].

The incorporation of materials possessing substances (i.e., algae extracts, charcoal, and biostimulants) that can enhance the growth of plants in hydroponics, accelerating and improving both crop yields and quality, has gained increasing popularity in recent years [16–18]. Biostimulants are regarded as a promising and eco-friendly strategy for crop production [19]. Biostimulants are considered eco-friendly since they enhance plant growth and resilience using natural or organic substances, reducing the need for synthetic fertilizers and pesticides [20]. They stimulate the plant's natural processes, improving its nutrient uptake, stress tolerance, and overall health without introducing harmful chemicals into the environment [21]. This minimizes soil and water contamination, promotes sustainable agriculture, and supports biodiversity by maintaining healthy ecosystems [22]. However, research on the direct introduction of biostimulants into hydroponic solutions is still limited, and further investigation is needed to validate their effects on crop production.

One of the most interesting biostimulants on the Italian market is undoubtedly wood distillate (*WD*), also known as pyroligneous acid or wood vinegar [23]. Over the last decade, this product has proven effective at improving crop yields and quality [24,25]. Since 2018, it has been approved for use in organic farming in Italy [26], offering an alternative to synthetic chemical fertilizers, which are to be reduced by 50% by 2030, as outlined in the Farm-to-Fork Directive by the European Commission [27]. Numerous scientific studies have demonstrated the positive effects of *WD* on field crops through both foliar applications and fertigation [24,25,28]. However, little is known about its actual applicability in hydroponic cultivation [29,30]. Wood distillate is a byproduct of the pyrolysis process of woody biomass, obtained through the condensation of gases produced during the combustion process [23]. The composition of *WD* is extensive, containing >300 active molecules that play a fundamental role in plant growth [31]. Given that *WD* has been shown to stimulate root growth in plants [32], its direct application to the root system can have an even more pronounced positive effect on their growth. A crucial characteristic of *WD* is its dose dependency [33]: above a certain concentration (ranging between 0.2% and 0.5%, depending on the application method), the product becomes toxic, leading to plant mortality [34].

This study advances our understanding of the largely unexplored effects of using *WD* in hydroponics and, to the best of our knowledge, represents the first investigation into the combined use of *WD* and SW in hydroponic cultivation. Specifically, this study aimed to assess the effects of different concentrations of seawater (3%, 6%, and 12% SW; percentages are related to the entire mineral content of the seawater), both alone and in combination with 0.2% (*v/v*) *WD*, in the nutrient solutions of a hydroponic system on lettuce (*Lactuca sativa* L., crop model plant) growth. The evaluation included biometric parameters (fresh leaf weight, fresh root weight, root length, root surface area), photosynthetic parameters (chlorophyll *a* and *b*, carotenoids, photosynthetic efficiency), and biochemical parameters (ascorbic acid, macro- and microelements), which were the main parameters for evaluating the effects of seawater and wood distillate used in this study.

2. Materials and Methods

2.1. Experimental Design and Growing Conditions

Eighty lettuce seedlings were acquired from a local nursery, Horta do Campo Grande, located in Lisbon, Portugal. In the laboratory, the roots of each seedling were thoroughly

washed with tap water to remove any adhering soil. Subsequently, each seedling was placed inside a plastic basket filled with sterilized clay, and the baskets were placed into 15 rectangular plastic containers (22 × 8 × 33 cm). These containers had a volumetric capacity of 8 L, and they were filled with a modified Hoagland's solution (Table S1, following the methodology reported in Cruz et al. [35]) dissolved in 3 different concentrations (3%, 6%, and 12%) of seawater (SW), in addition to a control without SW (features in Table S2). Seawater was collected at the Carcavelos beach (Cascais, Portugal) and stored in a climatic chamber until use. The different seawater solutions were prepared by dissolving the pure seawater and analyzing the mineral content by ICP-MS and the electrical conductivity with a conductivity meter. Wood distillate (*WD*) was added to the treatment solution to reach a concentration of 0.2%, following the approach documented by Fedeli et al. [24]; the complete chemical characterization of the *WD* is reported in Table S3 [36]. "*No WD*" refers to samples without the addition of wood distillate in the nutrient solution, and "*WD*" refers to samples with the addition of wood distillate in the nutrient solution. The entire experiment took place within a controlled chamber set at a temperature of 20 ± 1 °C, a relative humidity (RH) of 70 ± 2%, and a photosynthetic photon flux density (PPFD) of 300 µmol m^{-2} s^{-1} (Figure 1). The environmental conditions followed a day/night cycle of 16/8 h. Upon concluding the experiment, plants were removed from the hydroponic system and, once in the laboratory, were immediately analyzed or stored at −80 °C for subsequent analysis.

Figure 1. Picture of the experimental plantation.

2.2. Photosynthetic Parameters

Leaf pigments were extracted from three leaf discs (Ø 2 mm) in 2 mL of methanol and maintained at +4 °C for 24 h in the dark. Using a UV–Vis spectrophotometer (SHIMADZU UV-1800, Tokyo, Japan), the sample's absorbance was measured at 470 nm, 652.4 nm, 665.2 nm, and 700 nm after incubation. The content of assimilation pigments was determined using the equations provided by Lichtenthaler and Buschmann [37]:

$$C_a(\text{ug/cm}) = 16.72\ A_{665.2} - 9.16\ A_{652.4}$$

$$C_b(\text{ug/cm}) = 34.09\ A_{652.4} - 9.16\ A_{665.2}$$

$$C_{(x+c)}(\text{ug/cm}) = \frac{(1000\ A_{470}\ - 1.63\ C_a\ - 104.96\ C_b)}{221}$$

Following 15 min of dark adaptation, leaves were exposed to a 1-second pulse of saturating red light at 650 nm with an intensity of 2400 µmol m^{-2} s^{-1}. The fluorescence emitted by the leaf was then measured using a plant efficiency analyzer (Handy PEA, Hansatech Ltd., Norfolk, UK). Two key photosynthetic efficiency indicators, namely, F_V/F_M (the maximum quantum yield of PSII, as outlined by Vannini et al. [38]) and PI_{ABS} (performance index, serving as an indicator of plant vitality) were utilized as indicators of the plants' health statuses. The measurements were repeated every 3 days for 21 days to better understand the effects of the treatment during plant growth: T_1 (after 3 days); T_2 (after 6 days); T_3 (after 9 days); T_4 (after 12 days); T_5 (after 15 days); T_6 (after 18 days); T_7 (after 21 days, end of the experiment).

2.3. Biometric Parameters

At the end of the experiment, the leaves and roots were weighted and then immediately frozen at −80 °C. Root images for each sample were taken using a digital camera (iPhone 14, Apple Inc., Cupertino, CA, USA) against a black background. Afterwards, root length and surface were evaluated using Fiji/ImageJ software (v. 1.54 h). The image scale was calibrated to relative pixels, equivalent to 1 mm.

2.4. Macro- and Micronutrient Contents

The analysis of the leaf and root mineral macro- and micronutrient contents was run at the Ionomics Laboratory (CEBAS-CSIC) according to ISO 11.885 (2007) [39], using Inductively Coupled Plasma–Optical Emission Spectrometry (ICP-OES) on a Thermo ICAP 6500 Duo instrument (Thermo Fisher Scientific, Waltham, MA, USA). Dried leaves were ground to a fine powder, and 200 mg of powder per sample was added to a 25 mL test tube along with 4 mL of a solution containing 68%-purity HNO_3 and 1 mL of 33%-purity H_2O_2 for digestion, to which a Teflon reactor containing 300 mL of high-purity deionized water, 30 mL of 33%-purity H_2O_2, and 2 mL of 98%-purity H_2SO_4 was added. Sample digestion was performed using a microwave heating program of three steps: starting at 20 °C and 40 bar, increasing by 10 bar per minute for 30 min until 220 °C, and maintaining the temperature for 20 min. After cooling, the mineralized samples were transferred to 10 mL (for micronutrients) and 25 mL (for macronutrients) double-gauge tubes, and the volume was adjusted using high-purity deionized water. Calibration standards were prepared using a standard solution containing 31 minerals supplied by SCP Science (Quebec, QC, Canada) in high-purity deionized water. ICP-OES analyses included two control samples of high-purity deionized water and a multi-mineral standard. Specifically, aluminum (Al), arsenic (As), and barium (Ba) were included as micronutrients because, at trace levels, they can play specific roles in biological systems [40]. Although these elements can be toxic at higher concentrations, in minute amounts, they may contribute to certain physiological processes or act as signaling agents [40].

2.5. Leaf Ascorbic Acid

The determination of the leaf ascorbic acid content followed the method described by Fedeli et al. [41]. Approximately 200 mg of leaves was homogenized in 0.8 mL of 10% (*w*/*v*) trichloroacetic acid (TCA). The resulting mixture was filtered through gauze, placed at −20 °C for 5 min, and then centrifuged at 3000 rpm for 5 min. Subsequently, 0.4 mL of the supernatant was mixed with 1.6 mL of dH_2O and 0.2 mL of 0.2 M Folin–Ciocalteu reagent (Carlo Erba, Cornaredo, MI, Italy). After 10 min of dark incubation, the samples were measured at 760 nm using a UV–Vis spectrophotometer (Agilent 8453, Santa Clara, CA, USA). The concentrations of the samples were determined using a calibration curve prepared with pure L-ascorbic acid (BioXtra, ≥99.0%, crystalline).

2.6. Statistical Analysis

The data approached a normal distribution (Shapiro–Wilk test, $p < 0.05$) and are presented as mean ± standard error (N = 10). One-way ANOVA was applied to check for differences in SW treatments, followed by LSD post hoc tests ($p < 0.05$) to compare the means, utilizing CoStat 6.45 software (CoHort, Berkeley, CA, USA). To compare the differences between the "*No WD*" and "*WD*" samples, a *t*-test was performed.

3. Results

3.1. Effects of SW Addition

The leaf chlorophyll *a* and carotenoid contents showed increases in plants grown at 50% seawater (SW) and 12%SW, while chlorophyll *b* showed an increase in plants grown at 12%SW (Table 1). Leaf photosynthetic parameters (F_v/F_m and PI_{abs}) were not affected by SW, at any time (Figure 2).

Table 1. Contents of chlorophyll *a*, chlorophyll *b*, and carotenoids (mean ± standard error, N = 10) in leaves of lettuce plants treated with different seawater (SW) concentrations [from 0 (control) to 12%]. "*No WD*" means plants grown without the addition of wood distillate, while "*WD*" means plants grown with 0.2% wood distillate addition. Lowercase letters indicate significant differences ($p < 0.05$) among the different SW concentrations without the addition of 0.2% *WD*, whereas uppercase letters indicate significant differences ($p < 0.05$) among the different SW concentrations with the addition of 0.2% *WD*, evaluated by the LSD test. * = significant differences between *No WD* and *WD* treatments within the same SW concentration evaluated by *t*-test ($p < 0.05$).

Seawater [%]	**0**		**3**		**6**		**12**	
	No WD	***WD***	***No WD***	***WD***	***No WD***	***WD***	***No WD***	***WD***
Chlorophyll *a* (μg cm^{-1})	2.03 ± 0.18 ^{c*}	3.39 ± 0.39 ^{C}	3.59 ± 0.25 ^{b*}	4.97 ± 0.28 ^{A}	2.94 ± 0.39 ^{bc*}	4.42 ± 0.3 ^{AB}	5.19 ± 0.42 ^{a*}	3.71 ± 0.37 ^{BC}
Chlorophyll *b* (μg cm^{-1})	0.5 ± 0.03 ^{b*}	0.68 ± 0.08 ^{B}	0.62 ± 0.07 ^{b*}	0.94 ± 0.05 ^{A}	0.57 ± 0.06 ^{b*}	0.83 ± 0.05 ^{AB}	1.02 ± 0.08 ^{a*}	0.70 ± 0.07 ^{B}
Carotenoids (μg cm^{-1})	0.71 ± 0.05 ^{c*}	1.12 ± 0.11 ^{B}	1.08 ± 0.09 ^{ab*}	1.56 ± 0.09 ^{A}	1.05 ± 0.11 ^{bc*}	1.34 ± 0.09 ^{AB}	1.52 ± 0.13 ^{a*}	1.16 ± 0.14 ^{B}

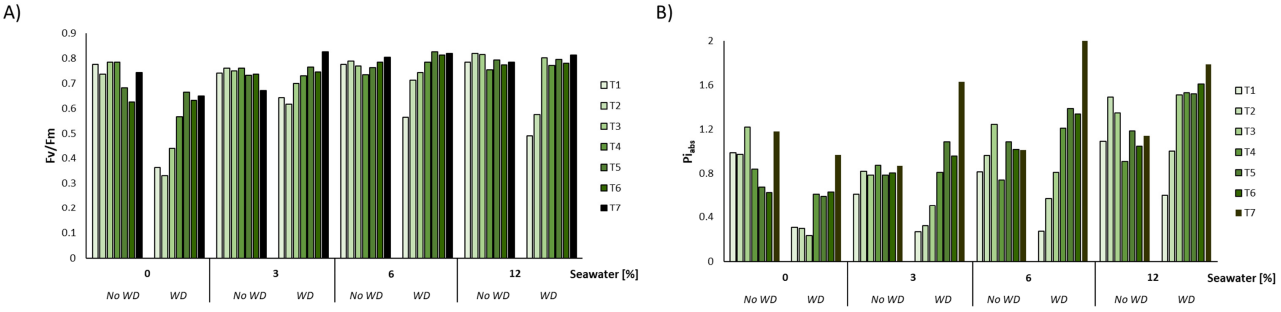

Figure 2. F_v/F_m (**A**) and PIabs (**B**) values (mean, N = 10) in lettuce plants treated with different seawater (SW) concentrations [from 0 (control) to 12%]. "*No WD*" means plants grown without the addition of wood distillate, while "*WD*" means plants grown with 0.2% wood distillate addition. Legend shows the different times when the measure was taken: T_1 (after 3 days); T_2 (after 6 days); T_3 (after 9 days); T_4 (after 12 days); T_5 (after 15 days); T_6 (after 18 days); T_7 (after 21 days, end of the experiment).

The addition of SW differently affected the biometric parameters (Figure 3). The leaf fresh weight and root length showed reductions in plants grown with 12%SW, while the root fresh weight and root area showed increases in plants grown both with 3%SW and 6%SW. Ascorbic acid decreased at all SW concentrations (Figure 4).

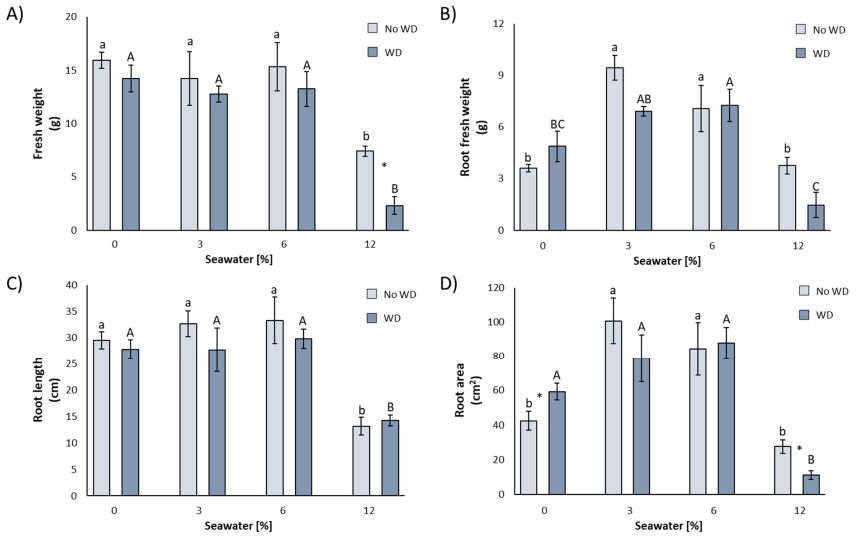

Figure 3. Leaf fresh weight (**A**), root fresh weight (**B**), root length (**C**), and root area (**D**) (mean ± standard error, N = 10) in lettuce plants treated with different seawater (SW) concentrations [from 0 (control) to 12%] are displayed on the horizontal axis. "*No WD*" means plants grown without the addition of wood distillate, while "*WD*" means plants grown with 0.2% wood distillate addition. Lowercase letters indicate significant differences ($p < 0.05$) among the different SW concentrations without the addition of 0.2% *WD*, whereas uppercase letters indicate significant differences ($p < 0.05$) among the different SW concentrations with the addition of 0.2% *WD*, evaluated by the LSD test. * = significant differences between *No WD* and *WD* treatments within the same SW concentration evaluated by *t*-test ($p < 0.05$).

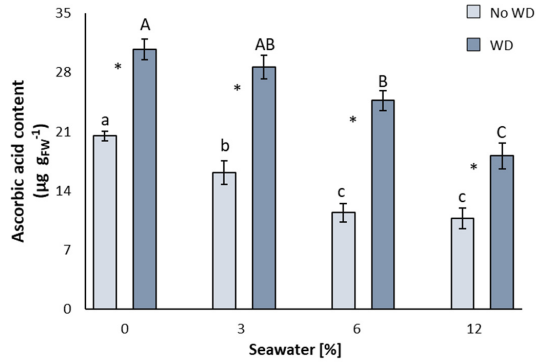

Figure 4. Contents of ascorbic acid (mean ± standard error, N = 10) in leaves of lettuce plants treated with different seawater (SW) concentrations [from 0 (control) to 12%] are displayed on the horizontal axis. "*No WD*" means plants grown without the addition of wood distillate, while "*WD*" means plants grown with 0.2% wood distillate addition. Lowercase letters indicate significant differences ($p < 0.05$) among the different SW concentrations without the addition of 0.2% *WD*, whereas uppercase letters indicate significant differences ($p < 0.05$) among the different SW concentrations with the addition of 0.2% *WD*, evaluated by the LSD test. * = significant differences between *No WD* and *WD* treatments within the same SW concentration evaluated by *t*-test ($p < 0.05$).

Leaf analyses showed increases in Al and Zn in plants grown with 6%SW and 12%SW, and in Na in plants grown with 3%SW (+100%), 6%SW (+200%), and 12%SW (+200%) (Table 2). On the contrary, decreases were evident for Cu, K, and Mg in plants grown with 3%SW, 6%SW, and 12%SW; for Mn in plants grown with 6%SW and 12%SW; and for P in plants grown with 3%SW (Table 2). Differences did not emerge for As, Ba, Ca, and Fe (Table 2).

Table 2. Contents of macro- and microelements (mean ± standard error, N = 10) in leaves of lettuce plants treated with different seawater (SW) concentrations [from 0 (control) to 12%]. "*No WD*" means plants grown without the addition of wood distillate, while "*WD*" means plants grown with 0.2% wood distillate addition. Lowercase letters indicate significant differences ($p < 0.05$) among the different SW concentrations without the addition of 0.2% *WD*, whereas uppercase letters indicate significant differences ($p < 0.05$) among the different SW concentrations with the addition of 0.2% *WD*, evaluated by the LSD test. * = significant differences between *No WD* and *WD* treatments within the same SW concentration evaluated by *t*-test ($p < 0.05$).

Micro- and Macroelements	Seawater [%]							
	0		3		6		12	
	No WD	*WD*	*No WD*	*WD*	*No WD*	*WD*	*No WD*	*WD*
Al (ug g_{DW}^{-1})	133 ± 10 ^{c}	140 ± 13	144 ± 6 ^{bc}	141 ± 8	175 ± 10 ^{a*}	138 ± 16	169 ± 10 ^{ab*}	134 ± 14
As (ug g_{DW}^{-1})	4.0 ± 0.4	4.6 ± 0.6	4.6 ± 0.8	4.6 ± 0.7	5.2 ± 0.4 *	4.2 ± 0.4	5.5 ± 0.4 *	3.3 ± 1.3
Ba (ug g_{DW}^{-1})	1.4 ± 0.2	1.3 ± 0.1	1.5 ± 0.1	1.5 ± 0.2	1.6 ± 0.1	1.4 ± 0.1	1.6 ± 0.1	1.3 ± 0.1
Ca (%)	2.2 ± 0.0 *	3.0 ± 0.1	2.2 ± 0.1 *	2.6 ± 0.2	2.3 ± 0.1	2.5 ± 0.1	2.2 ± 0.1	1.9 ± 0.4
Cu (ug g_{DW}^{-1})	97 ± 1 ^{a}	120 ± 23	57 ± 2 ^{b}	61 ± 19	42 ± 1 ^{c}	48 ± 3	45 ± 2 ^{c}	41 ± 4
Fe (ug g_{DW}^{-1})	134 ± 2 ^{ab*}	155 ± 6	151 ± 7 ^{a}	132 ± 10	129 ± 10 ^{b}	134 ± 6	130 ± 3 ^{b}	133 ± 7
K (ug g_{DW}^{-1})	0.8 ± 0.0 ^{a*}	0.9 ± 0.0 ^{A}	0.4 ± 0.1 ^{b*}	0.7 ± 0.1 ^{B}	0.3 ± 0.1 ^{b*}	0.6 ± 0.0 ^{B}	0.4 ± 0.0 ^{b*}	0.5 ± 0.0 ^{B}
Mg (%)	0.4 ± 0.0 ^{a*}	0.6 ± 0.0	0.3 ± 0.0 ^{b*}	0.5 ± 0.1	0.2 ± 0.0 ^{b*}	0.4 ± 0.0	0.2± 0.1 ^{b*}	0.4 ± 0.1
Mn (ug g_{DW}^{-1})	202 ± 3 ^{a*}	184 ± 9 ^{A}	167 ± 13 ^{b*}	190 ± 4 ^{A}	157 ± 11 ^{b}	158 ± 4 ^{B}	173 ± 8 ^{ab}	164 ± 13 ^{B}
Na (ug g_{DW}^{-1})	0.1 ± 0.0 ^{c}	0.1 ± 0.0 ^{B}	0.2 ± 0.0 ^{b}	0.3 ± 0.1 ^{A}	0.3 ± 0.1 ^{ab}	0.3 ± 0.1 ^{A}	0.3 ± 0.0 ^{a}	0.3 ± 0.1 ^{A}
P (%)	0.6 ± 0.0 ^{a}	0.7 ± 0.1	0.4 ± 0.0 ^{b*}	0.7 ± 0.1	0.5 ± 0.1 ^{ab}	0.6 ± 0.1	0.6 ± 0.0 ^{a}	0.6 ± 0.1
Zn (ug g_{DW}^{-1})	32 ± 1 ^{c*}	36 ± 2 ^{B}	50 ± 4 ^{ab*}	40 ± 3 ^{AB}	63 ± 8 ^{a*}	44 ± 2 ^{B}	42 ± 1 ^{bc*}	32 ± 5 ^{B}

Al: aluminum; As: arsenic; Ba: barium; Ca: calcium; Cu: copper; Fe: iron; K: potassium; Mg: magnesium; Mn: manganese; Na: sodium; P: phosphorus; Zn: zinc.

Root analyses showed an increase in Zn in plants grown with 3%SW and 6%SW and in Na in plants grown with 3%SW, 6%SW, and 12%SW (Table 3). A decrease emerged for Cu, K, Mg, and Mn in plants grown with 3%SW, 6%SW, and 12%SW (Table 3). Differences were not found for Al, As, Ba, Ca, Fe, and P (Table 3).

Table 3. Contents of macro- and microelements (mean ± standard error, N = 10) in roots of lettuce plants treated with different seawater (SW) concentrations [from 0 (control) to 12%]. "*No WD*" means plants grown without the addition of wood distillate, while "*WD*" means plants grown with 0.2% wood distillate addition. Lowercase letters indicate significant differences ($p < 0.05$) among the different SW concentrations without the addition of 0.2% *WD*, whereas uppercase letters indicate significant differences ($p < 0.05$) among the different SW concentrations with the addition of 0.2% *WD*, evaluated by the LSD test. * = significant differences between *No WD* and *WD* treatments within the same SW concentration evaluated by *t*-test ($p < 0.05$).

Micro- and Macroelements	Seawater [%]							
	0		3		6		12	
	No WD	*WD*	*No WD*	*WD*	*No WD*	*WD*	*No WD*	*WD*
Al (ug g_{DW}^{-1})	137 ± 3	129 ± 5	158 ± 14	142 ± 12	148 ± 20	162 ± 9	152 ± 13 *	129 ± 13
As (ug g_{DW}^{-1})	4.0 ± 0.6	5.0 ± 0.7 ^{AB}	4.8 ± 0.9	5.2 ± 0.2 ^{A}	5.2 ± 0.9 *	3.8 ± 0.1 ^{B}	5.9 ± 0.0 *	3.1 ± 0.1 ^{B}
Ba (ug g_{DW}^{-1})	1.5 ± 0.0	1.3 ± 0.2	1.5 ± 0.1	1.5 ± 0.1	1.4 ± 0.1	1.6 ± 0.0	1.4 ± 0.1	1.4 ± 0.1
Ca (%)	2.4 ± 0.1 ^{ab}	2.6 ± 0.1 ^{B}	2.3 ± 0.0 ^{ab*}	3.0 ± 0.1 ^{A}	2.5 ± 0.2 ^{a}	2.5 ± 0.0 ^{B}	2.1 ± 0.1 ^{b}	2.0 ± 0.1 ^{C}
Cu (ug g_{DW}^{-1})	95 ± 2 ^{a}	91 ± 3 ^{A}	67 ± 5 ^{b*}	44 ± 1 ^{B}	44 ± 3 ^{c}	47 ± 4 ^{B}	44 ± 2 ^{c}	44 ± 4 ^{B}
Fe(ug g_{DW}^{-1})	132 ± 1 *	169 ± 9	143 ± 16	127 ± 3	149 ± 7	161 ± 19	152 ± 19 ^{b*}	112 ± 24
K (ug g_{DW}^{-1})	0.6 ± 0.0 ^{a*}	0.8 ± 0.1	0.4 ± 0.0 ^{b*}	0.6 ± 0.0	0.4 ± 0.0 ^{b*}	0.6 ± 0.1	0.3 ± 0.0 ^{b*}	0.6 ± 0.1
Mg (%)	0.4 ± 0.0 ^{a*}	0.5 ± 0.0 ^{AB}	0.2 ± 0.0 ^{b*}	0.6 ± 0.0 ^{A}	0.3 ± 0.0 ^{b*}	0.4 ± 0.0 ^{B}	0.2 ± 0.0 ^{b*}	0.4 ± 0.0 ^{B}
Mn (ug g_{DW}^{-1})	200 ± 1 ^{a}	180 ± 9 ^{A}	144 ± 3 ^{c}	164 ± 7 ^{A}	165 ± 12 ^{bc}	177 ± 11 ^{A}	171 ± 9 ^{b*}	111 ± 17 ^{B}
Na (ug g_{DW}^{-1})	0.1 ± 0.0 ^{c}	0.1 ± 0.0 ^{B}	0.2 ± 0.0 ^{b}	0.3 ± 0.0 ^{A}	0.3 ± 0.0 ^{b}	0.3 ± 0.0 ^{A}	0.4 ± 0.0 ^{a}	0.3 ± 0.0 ^{A}
P (%)	0.5 ± 0.0 ^{ab*}	0.8 ± 0.1 ^{A}	0.4 ± 0.0 ^{b*}	0.7 ± 0.0 ^{A}	0.5 ± 0.0 ^{a}	0.5 ± 0.1 ^{B}	0.5 ± 0.0 ^{a*}	0.7 ± 0.1 ^{A}
Zn (ug g_{DW}^{-1})	34 ± 2 ^{c}	39 ± 1 ^{AB}	53 ± 3 ^{b*}	42 ± 2 ^{AB}	64 ± 4 ^{a*}	44 ± 3 ^{A}	42 ± 2 ^{c*}	33 ± 6 ^{B}

Al: aluminum; As: arsenic; Ba: barium; Ca: calcium; Cu: copper; Fe: iron; K: potassium; Mg: magnesium; Mn: manganese; Na: sodium; P: phosphorus; Zn: zinc.

3.2. Effects of WD Addition at Different SW Concentrations

Chlorophyll *b* and carotenoids showed increases in plants grown with 3%SW and 12%SW, while chlorophyll *a* showed an increase in plants grown with 3%SW and 6%SW (Table 1). All plants treated with *WD* at the different SW concentrations showed increasing trends of the photosynthetic parameters (F_v/F_m and PIabs), particularly from T4 (Figure 2).

The addition of *WD* at the different SW concentrations differently affected the biometric parameters (Figure 2). The leaf fresh weight, root length, and root area showed reductions in plants grown with 12%SW, while the root fresh weight showed an increase in plants grown with 6%SW.

The leaf concentration of ascorbic acid decreased along with the increase in SW, being, however, always higher in plants treated with *WD* (Figure 3).

Leaf analyses showed an increase in Na and decreases in K and Mn in plants treated with SW (Table 2). SW did not affect the leaf contents of Al, As, Ba, Ca, Cu, Fe, Mg, P, and Zn (Table 2).

Root analyses showed an increase in Na and decreases in Cu and P in plants grown with SW (Table 3). Differently, there was an increase in Ca in plants grown with 3%SW and a decrease in those grown with 12%SW (Table 3). No difference was found for Al, As, Ba, Fe, K, Mg, and Zn (Table 3).

3.3. Differences between WD-Untreated and WD-Treated Plants at Various SW Concentrations

The chlorophyll *a*, chlorophyll *b*, and carotenoids increased in plants treated with *WD* and grown with 0%SW, 3%SW, and 6%SW, while they decreased with 12%SW (Table 1).

The addition of *WD* also influenced the other parameters analyzed in plants grown at the same SW concentration (Figure 2). In the presence of *WD*, the leaf fresh weight showed a reduction in plants grown with 12%SW, while the root area showed an increase in plants grown with 0%SW and a decrease in those grown with 12%SW. Differences were not found for the root fresh weight and root length at any SW concentration (Figure 2).

Independently of SW treatment, the addition of *WD* to the root medium caused an increase in the ascorbic acid content (Figure 3).

Leaf analyses showed increases in K in plants grown with 0%SW, 3%SW, 6%SW, and 12%SW, in Mg in plants grown with 0%SW, 3%SW, and 6%SW, in Ca in plants grown with 0%SW and 3%SW, in P in plants grown with 3%SW, and in Fe in plants grown with 0%SW (Table 2). There were instead decreases in Al and As in plants grown with 6%SW and 12%SW (Table 2). Manganese showed a decrease in plants grown with 0%SW and an increase in those grown with 3%SW, similar to the Zn content, which showed an increase in plants grown with 0%SW and a decrease in those grown with 3%SW, 6%SW, and 12%SW (Table 3). Differences were not found for Ba, Cu, and Na (Table 2).

Root analyses showed increases in K and Mg in plants grown with 0%SW, 3%SW, 6%SW, and 12%SW, in P in plants grown with 0%SW, 6%SW, and 12%SW, and in Ca in plants grown with 3%SW (Table 3). There were instead decreases in Zn in plants grown with 3%SW, 6%SW, and 12%SW, in As in plants grown with 6%SW and 12%SW, and in Al and Mn in plants grown with 12%SW (Table 3). Differences were not found for Ba, Cu, and Na (Table 3).

4. Discussion

This work enhances our understanding of the scarcely known effects of the use of *WD* in hydroponics, and it allows us to investigate for the first time, to the best of our knowledge, the combined use of *WD* and SW in hydroponic cultivation. It is well documented in the scientific literature how low concentrations of salts and/or SW can have a stimulating effect on plant growth, both in traditional cultivation and hydroponics [42–45]. However, when a certain threshold of the saline concentration is exceeded, a metabolic shift occurs in plants, and treatments start to become toxic. Salt concentrations >12% typically have negative effects on plant growth [46], as is also evident from the results of this study. Even the combined treatment of *WD* and SW at concentrations >12% was not able to counteract

the impact of salinity on plant growth, resulting in decreases in all the biometric parameters analyzed. Moreover, the combined presence of a high SW concentration, already harmful to plants by itself, and *WD* in the nutrient solution amplified plant damage, significantly reducing the development of leaves and roots.

It is widely known that treatments with *WD* significantly increase the chlorophyll contents of crop leaves [47,48], and, even in our case, statistically significant increases were found in *WD*-treated plants compared to their controls, except for the highest concentration (12%SW). We speculate that a metabolic shift occurred due to the combined stress imposed by the high concentration of SW and the presence of *WD* in the nutrient solution. Seawater, with its high salinity, is well documented as being harmful to plants, primarily due to osmotic stress, ionic toxicity, and the disruption of nutrient uptake [49,50]. These factors alone are sufficient to impair photosynthesis, reduce the chlorophyll content, and ultimately limit plant growth and development. However, when combined with *WD*, which can introduce additional organic compounds and potentially phytotoxic elements into the solution, the stress on the plants is likely exacerbated. Wood distillate contains a complex mixture of organic acids, phenolic compounds, and other organic molecules that, although beneficial in low concentrations [36], can become harmful when present in higher quantities or when combined with other stressors like salinity. This combination likely disrupts the plants' metabolic processes even further, causing a compounded stress response. The stress response may include an alteration in the synthesis and degradation of photosynthetic pigments, leading to the observed reduction in assimilation pigments such as chlorophylls and carotenoids. Furthermore, this metabolic shift might have triggered a reallocation of resources within the plants, prioritizing stress defense mechanism overgrowth, thereby negatively affecting the biometric parameters analyzed.

The results obtained regarding the photosynthetic parameters at various plant growth stages are noteworthy. In a previous work [24], for the first time, the possible role of *WD* as a eustressor was hypothesized. Unlike distress, which is harmful stress, eustress is a type of stress that can have positive effects on plant performance [51]. The results of the analysis of the fluorescence parameters seem to support this hypothesis. Indeed, across all the tested SW concentrations, both investigated parameters, specifically F_v/F_m and Pi_{abs}, exhibited a similar pattern in plants treated with *WD* compared to the control plants, with an initial decrease and then a gradual increase until reaching values similar or higher than those measured in the control samples. The parameters F_v/F_m and Pi_{abs} are commonly used to assess the efficiency of photosystem II (PSII), which is a key component of the photosynthetic process [52]. These parameters provide insights into the health and performance of plants, particularly in terms of their ability to capture and use light energy for photosynthesis [53]. Interestingly, the addition of *WD* influenced the leaf content of ascorbic acid, which is an essential metabolite and a key element for the metabolism of all living organisms, especially plants [54]. The increase in the ascorbic acid content in lettuce leaves following exposure to *WD* can be attributed to a combination of enhanced antioxidant defense mechanisms, the activation of the stress responses, and the modulation of metabolic pathways and enzyme activities. The specific components of *WD*, such as phenolic compounds, organic acids, and terpenes, contribute to these effects by influencing the plant's internal stress responses and metabolic processes [24]. These compounds play an important role in the antioxidant system of plants by offering protection against oxidative stress [55]. At all the SW concentrations tested, we witnessed a statistically significant increase in the *WD*-treated plants compared to the relevant controls (min: +50.05%–max: +116.08%). The effect of *WD* on the increase in antioxidant compounds in traditionally cultivated plants is widely documented in the scientific literature [28,56], while only one recent study investigated the contents of polyphenols, flavonoids, and total antioxidant power in hydroponically grown lettuce [30]. The interaction between salinity stress and the compounds present in *WD* significantly influences the oxidative stress response in lettuce. Salinity induces oxidative stress through ROS accumulation, while *WD* compounds, particularly phenolic compounds and organic acids, can enhance antioxidant defenses

and mitigate oxidative damage. The combined presence of these factors can lead to a complex stress response, where *WD* components potentially alleviate some of the oxidative stress caused by salinity, thereby improving the plant's overall resilience. This interaction underscores the potential of using natural compounds like those in *WD* to support plant stress tolerance in challenging growing conditions. These results confirm that *WD*, in the early stages of application to plants, induces (eu)stress, activating defense mechanisms that subsequently allow plants to increase certain metabolic parameters, such as their antioxidant levels.

Plants grown with the addition of *WD* across the different SW concentrations showed increases in various elements (i.e., Ca, Mg, P, K) that play critical roles in plant growth, in particular in the presence of saline stress. Calcium is especially important during salt stress, as it helps to maintain the cell wall and membrane integrity while also regulating plant growth and development [57]. Magnesium (Mg) is essential for many biochemical and physiological processes in plants, including photosynthesis, protein synthesis, and nucleotide metabolism [58]. The interaction between salinity and P is extremely complex, with no clear mechanism explaining the changes in P uptake in response to salinity stress in various species [59]. However, the P concentration has been linked to the photosynthesis rate [60]. As a result, a lack of P in leaves can lead to slower plant growth. In general, negative effects of salinity on plant growth are attributed to a decrease in the osmotic potential of the growing medium, specific ion toxicity, and nutrient ion deficiency caused by K^+ nutrition disruption. The accumulation of inorganic ions (Na^+, Cl^-, and K^+) and compatible organic solutes (soluble carbohydrates, amino acids, and proline) can cause this disruption [61–63]. Osmotic adjustment in both roots and leaves helps to maintain water uptake and cell turgor, which are required for vital physiological processes such as stomatal opening, photosynthesis, and cell expansion [64,65]. Regarding As and Al, the study observed decreases in their levels in lettuce leaves cultivated with high concentrations of SW, specifically at 6% and 12%SW, in the presence of *WD*. This reduction can be attributed to several factors. High salinity induces osmotic stress and creates competition for ion uptake, which can decrease the absorption of metals such as Al and As, as the elevated ion concentration from seawater likely reduces their availability for plant uptake [48,66,67]. Additionally, *WD* contains organic compounds like phenolics and organic acids that may function as chelators, binding to Al and As and thereby reducing their bioavailability. Wood distillate may also alter soil properties and interact with the effects of salinity, influencing metal uptake [48,66,67]. The combined presence of *WD* and high salinity might result in a synergistic effect, where *WD* helps alleviate some of the oxidative stress and metal accumulation typically caused by high salinity, with its chelating properties contributing to lower levels of Al and As. Overall, these findings suggest that *WD* could serve as an effective additive for managing metal contamination in crops under saline conditions, potentially improving both plant health and food safety.

While our work suggests a role for *WD* in hydroponic culture with saltwater irrigation, further studies are needed to understand whether the effects are solely related to osmotic regulation or whether other mechanisms are put in place and how they depend on the *WD* composition.

5. Conclusions

The findings of this study offer valuable insights into the potential of integrating low concentrations of seawater (SW) and wood distillate (*WD*) into hydroponic systems for sustainable agriculture. The use of SW and *WD* not only provides a viable alternative to freshwater irrigation but also opens up new possibilities for resource management in agriculture. By reducing the dependency on freshwater, these practices contribute to more sustainable water use and offer a practical solution for regions facing water scarcity. The synergistic effects observed between SW and *WD* highlight the opportunity for developing integrated water and nutrient management strategies. This approach could optimize resource use and improve plant health and productivity. Exploring the specific interactions

between SW, *WD*, and various plant species could yield tailored solutions that maximize benefits and mitigate any potential drawbacks.

In addition, this study underscores the importance of ongoing research to refine these practices and assess their scalability and long-term impacts. Future investigations should focus on evaluating the economic feasibility, environmental benefits, and potential trade-offs associated with the use of SW and *WD* in diverse agricultural settings, while also studying the response to other crop types.

Supplementary Materials: The following supporting information can be downloaded at https://www.mdpi.com/article/10.3390/su16167186/s1, Table S1. Chemical composition of the base nutrient solution; Table S2. Chemical composition of seawater; Table S3. Main physicochemical characteristics of wood distillate.

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