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Deficit Irrigation with *Ascophyllum nodosum* Extract Application as a Strategy to Increase Tomato Yield and Quality

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Abstract: Deficit irrigation is applied to several agricultural crops as a water-saving irrigation strategy. The tomato plant is sensitive to water stress; however, integration with biostimulant applications, based on seaweed extracts, could be a strategy for plants adapting to this abiotic condition. The objective of this study was to evaluate agronomic and quality aspects of tomato cultivated under deficit irrigation combined with *Ascophyllum nodosum* extract (ANE) application. The experiment was conducted using a completely randomized design with two water replacement levels, 70 and 100% of crop evapotranspiration (ETc), and five doses of ANE (0, 0.1, 0.2, 0.3 and 0.4%) applied via soil drench. The interaction between ANE and ETc was significant ($p < 0.05$) in terms of plant growth, physiological parameters, fruit yield, yield components and fruit quality. Results indicated that when the tomato plant is under deficit irrigation, a higher ANE dose is required to achieve better development when compared to the 100% ETc condition, where the dose is lower. Under deficit irrigation, the largest fruit yield was obtained with 0.3 and 0.4% ANE, and with 100% ETc, the largest fruit yield was obtained with 0.2% ANE. ANE applications were also effective in increasing plant height, stem diameter, plant biomass, leaf area, chlorophyll and relative water content. In addition, tomato quality was also favored under deficit irrigation and seaweed extract application. We conclude that ANE applications attenuate water deficit effects in tomato plants and provide a strategy to ameliorate tomato yield, tomato quality and water use in agriculture.

Keywords: biostimulant; *Solanum lycopersicon*; sustainability; water saving; water stress

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1. Introduction

Tomato (*Solanum lycopersicon*) is one of the most produced and consumed fruit worldwide [1], and is a crop which is sensitive to abiotic stresses, especially water deficit [2]. The reproductive stage is the most critical, as the reduction in the amount of water available to the plants limits crop evapotranspiration, causing changes in plant physiological parameters [3–5]. The water deficit effect can vary during the crop cycle according to the cultivar, bunch position on the plant, and soil type [6,7].

Deficit irrigation is a strategy where the water supply is reduced to levels below the crop water requirements, where plants deal with stipulated stress. This technique has a high potential for improving the sustainability of tomato production [7]. It can be applied during the entire crop cycle or at specific phenological stages, usually those less sensitive to water deficit [8,9]. Deficit irrigation combined with other management techniques benefits tomato quality and yield [10,11] and contributes to increase water use efficiency and water productivity [12–15].

One promising study is that of biostimulant use based on seaweed extracts to reduce water stress impacts on crop yields and contribute to sustainable agriculture [16]. *Ascophyllum nodosum* is a seaweed related to increased tolerance to drought stress in tomato plants [2]. It can be used to promote plant growth, plant defense against biotic and abiotic stress and increase product quality [17]. The action mechanisms of *Ascophyllum nodosum* extract (ANE) in plants related to water stress are described by De Saeger et al. [18]. Many mechanisms are influenced by seaweed components, such as polyphenols, carbohydrates, amino acids, vitamins, macro- and micronutrients, cytokinins, auxins, and abscisic acid [2,19,20]. Generally, these components affect positively the endogenous balance of plant hormones, regulate the transcription of some transporters to alter and increase nutrient absorption and improve the performance of photosynthesis; therefore, plants under stress can mitigate water stress and have better development [18].

Deficit irrigation effects on agronomical and physiochemical aspects in tomato production have been reported [7,8], but few works have investigated ANE effects on tomato agronomical response, especially in protected environments under subtropical conditions. Moreover, these works have investigated the foliar application of the extract, so the application to the soil through irrigation water is little explored. Thus, studying different doses of the extract applied via fertigation has novel contributions for the tomato production sector, which may be another option for applying the biostimulant.

The objective was to analyze deficit irrigation effects on tomato production (Tucaneiro hybrid), exploring changes in plant growth (plant height, stem diameter, total leaf area, leaf, stem and root dry mass), physiological aspects (relative water content and chlorophyll content), fruit yield and yield components (bunch number plant^{-1}, bunch length, fruit number plant^{-1}, fruit mass, and fruit transversal diameter) and fruit quality (total acidity, pH, total soluble solids, ratio, and total polyphenols) and analyze the impact of different doses of *Ascophyllum nodosum* extract as a biostimulant to attenuate water stress. Therefore, it is expected that deficit irrigation will result in negative effects on tomato production, while the use of *Ascophyllum nodosum* extract may be able to minimize these negative effects.

2. Materials and Methods

2.1. Local and Environmental Conditions

The experiment was conducted at the Technical Irrigation Center of the State University of Maringá (23°23″ S, 51°57″ W and 542 m of altitude), Brazil. The city's climate according to Köppen is classified as humid mesothermal subtropical with 22.1–23.0 °C of average temperature, 70.1–75% of annual relative humidity and 1400–1600 mm of annual precipitation [21].

The tomato plants were cultivated in a protected environment of 25 m of length and 14 m of width, with a plastic cover of 150 µm thick and 3.5 m ceiling height surrounded by an anti-aphid nylon mesh. During the tomato crop cycle, temperature and relative humidity were measured by a meteorological station with a CR-23X datalogger and sensors Campbell HMP45C^{TM} placed at 2 m, present inside the experimental area (Figure 1).

The soil of the experimental area is classified as Ultisol by Soil Taxonomy [22], with 71% clay, 17% silt and 12% sand, and chemical characteristics of pH $CaCl_2$: 6.4; carbon organic: 10.6 g dm^{-3}; calcium: 7.4 cmol$_c$ dm^{-3}; magnesium: 2.1 cmol$_c$ dm^{-3}; potassium: 0.6 cmol$_c$ dm^{-3}; phosphorus (Mehlich-I): 90 mg dm^{-3}; sulfur: 104 mg dm^{-3}; aluminum: 0 cmol$_c$ dm^{-3}; hydrogen: 2.3 cmol$_c$ dm^{-3}; boron: 0.5 mg dm^{-3}; copper: 14.1 mg dm^{-3}; iron: 80.7 mg dm^{-3}; manganese: 141.4 mg dm^{-3}; and zinc: 7.5 mg dm^{-3}. The analyses were carried out according to Silva [23].

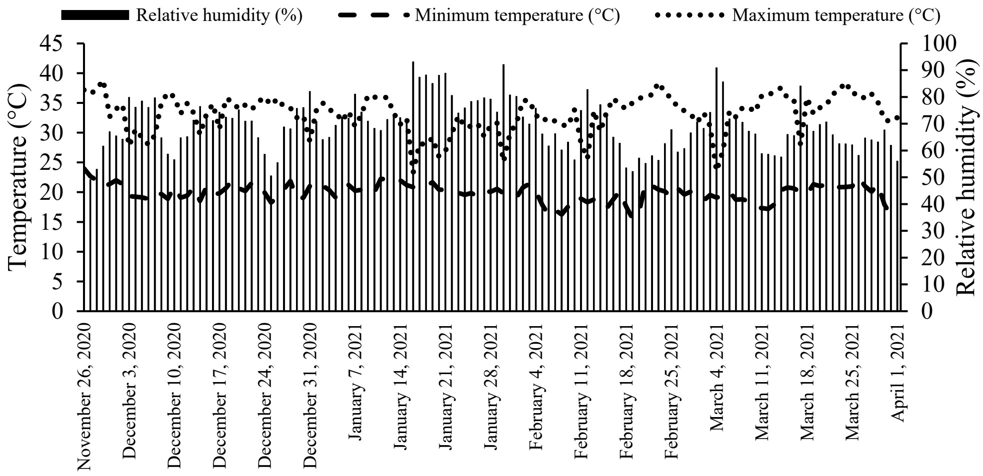

Figure 1. Maximum and minimum temperature (°C) and relative humidity (%) inside the protected environment during tomato crop cycle.

2.2. Treatment Applications and Experimental Design

The tomato cultivation (Tucaneiro hybrid^{TM}) occurred from 27 November 2020 to 1 April 2021; the experiment was conducted in a completely randomized design, with a factorial scheme (2 × 5), with two water replacement levels (70% and 100% of crop evapotranspiration (ETc)) and five ANE doses (0, 0.1, 0.2, 0.3 and 0.4%). Four beds were built of 3 × 0.5 m per treatment, totaling forty beds with six plants each, and the useful area consisted of three central plants, totaling twelve replicates per treatment.

The method used to determine crop evapotranspiration was lysimetry. It used three lysimeters of constant water table with compensation installed inside the protected environment, with two tomato plants each, according to Andrean et al. [24]. The crop evapotranspiration was determined daily at 8 h, by direct measurement from the lysimeters, through the difference in the volume of water used by the plants during 24 h. The average evapotranspirate volume of the three lysimeters corresponded to 100% ETc, and the 70% ETc was obtained by reducing 30% of this volume. During the crop cycle, the treatments that received 100% ETc had a water consumption of 470.08 mm and the treatments with 70% ETc 329.06 mm.

The product Acadian^{TM}, derived exclusively from the seaweed *Ascophyllum nodosum*, was used. In addition to fresh seaweed, the product contains potassium hydroxide, which is used in the extraction process, water, and 0.5% citric acid complexing agent. It is a liquid product; thus, the doses correspond to 0, 1, 2, 3 and 4 mL of the extract for each 1000 mL of water. ANE was applied five times during the tomato crop cycle, comprising all phenological phases at 15, 30, 45, 60 and 75 days after transplanting (DAT), applying a volume of 2 L per bed and application, by soil drench, applied over the soil surface of each bed.

2.3. Crop Management

The tomato seedlings were planted 30 days after sowing, with a spacing of 0.5 m between plants and 1.5 m between beds. The tomato plants were conducted on a single stem with the tutors' help, removing all secondary stems during plant development. The apical meristem was cut to stop plant growth at 90 DAT.

2.4. Irrigation and Fertigation

Irrigations were realized daily, using a drip irrigation system with self-compensating drippers spaced at 0.25 m, with a flow rate of 5 L h^{-1}, and a service pressure of 2 bar was installed. The reduction of water volume applied to the treatments with 70% ETc was started at 7 DAT for tomato seedling acclimatization.

The irrigation system presented a uniform flow, presenting 96% of Christiansen's uniformity coefficient (CUC) and a distribution uniformity coefficient (CUD) of 93%. The quality parameters of water used were analyzed according to Parron et al. [25] and presented apparent color < 0.2 (Hazen unit), conductivity of 158.6 µS cm^{-1}, pH of 7.8, turbidity of 0.4 (turbidity unit), total $CaCO_3$ of 49 mg L^{-1}, calcium hardness of 36.3 mg L^{-1} and magnesium hardness of 12.6 mg L^{-1}.

Fertigation was carried out every fifteen days, with calcium nitrate and potassium nitrate applied through the drip irrigation system, with dosages of 40 kg ha^{-1} N and 60 kg ha^{-1} K_2O, respectively. Boric acid (1 g L^{-1}) was applied to inflorescences, and calcium chloride (2 g L^{-1}) was applied to fruit in the formation stage by spray application [26].

2.5. Plant Growth Parameters and Physiological Aspects

Regarding plant growth parameters, plant height, stem diameter, total leaf area, leaf, stem and root dry mass were analyzed. After removing the apical meristem, plant height was measured with a graduated tape and stem diameter measured using a digital caliper (0.01 mm) at 5 cm from the soil. At the end of the tomato harvest, all leaves were collected to estimate the total leaf area $plant^{-1}$, measured with a LI 3100 area meter (LI-COR™). The leaves, stem and root were dried at 65 °C until reaching a constant weight, in order to obtain plant dry matter using semi-analytical balance (±0.001 g).

The physiological aspects analyzed were chlorophyll content and relative water content. Chlorophyll content determinations were performed in leaves from the plants' upper third at 90 DAT, starting at 8 h using SPAD-502 Plus (Konica Minolta^{TM}), expressed in SPAD index. The same leaves used for chlorophyll measurements were used to estimate the relative water content (RWC), foliar segments were cut and weighed to obtain fresh mass, and were then submerged in distilled water for 24 h, recording the turgor mass; the dry mass was obtained after 72 h at 65 °C. RWC was calculated using the method adopted by Ihuoma and Madramootoo [27]:

$$\text{RWC}\ (\%) = \left(\frac{(\text{Mf} - \text{Md})}{(\text{Mt} - \text{Md})}\right) \times 100 \quad (1)$$

where RWC = relative water content (%); Mf = fresh mass (g); Mt = turgor mass (g); and Md = dry mass (g).

2.6. Fruit Yield and Yield Components

All fruits with a red color from each plant were harvested, weighed (g) and the transversal diameter (cm) measured with a digital caliper. Fruit yield (kg) was obtained by the fruit mass sum of each plant, bunch number $plant^{-1}$ was evaluated, and bunch length (cm) was measured with a graduated tape from bunch insertion until the last fruit.

2.7. Fruit Quality

Regarding some fruit quality parameters, total acidity (TA), pH, total soluble solids (TSS), ratio and total phenolic compounds (TPC) were analyzed. Three tomatoes from each plant were crushed to obtain a pulp, 5 g was added to 50 mL of distilled water, stirred for 10 min, titrated with NaOH (0.1 N) up to 8.2 for TA measurement, which was calculated using Equation (2) and expressed in (%) of citric acid [28]. The pH measurement was carried out with a pH-meter (Tecnopon, mPA-210). The TSS were determined using a digital refractometer (ATAGO-P32), expressed in (%), and the ratio was determined by dividing the TSS by the TA values.

$$\text{Citric acid}(\%) = \left(\frac{(\text{V})(\text{N})(\text{Weq})}{\text{m}}\right) \times 100 \quad (2)$$

where V = NaOH used in titration (mL); N = NaOH solution normality (meq mL^{-1}); Weq. = equivalent weight of citric acid (0.064 g meq^{-1}); and m = sample fresh mass (g).

The TPCs were determined according to Singleton and Rossi [29], with modifications. A phenolic extract was prepared using 1 g of pulp with 4 mL of Metanol (100%). A phenolic extract aliquot (125 μL) was mixed with 125 μL of Folin–Ciocalteu 50% reagent and 2250 μL of sodium carbonate (28 g L^{-1}). Absorbance at 725 nm was measured in a spectrophotometer (Evolution™ 300, Thermo Fisher Scientific, Waltham, MA, USA) and compared with a standard curve of gallic acid solution. Results are expressed in grams of gallic acid equivalents per kilogram of fresh sample (g kg^{-1}).

2.8. Statistical Analysis

Data were submitted to two-factor analysis of variance (ANOVA) by F test at 5% of significancy using Sisvar software [30]. After detecting that the interaction between the two factors (ANE × ETc) was significant, data were compared using Tukey's test to separate results into homogeneous groups and verify which dose was better for the water conditions analyzed. Regression analysis was performed for ANE doses, and Pearson linear correlation was carried out between some tomato parameters.

3. Results

3.1. Plant Growth Parameters

The interaction between ANE values and ETc was significant in all plant growth parameters evaluated (Table 1). Deficit irrigation imposition during the tomato crop caused a reduction in plant height, stem diameter, and leaf area, as well as in the dry matter of leaves and roots, compared to unstressed plants (100% ETc). However, through association with ANE application in a water stress condition the tomato plants were able to achieve better growth and development. With deficit irrigation, higher doses of ANE were more efficient in attenuating the effects of water stress. In general, ANE applications (0.3 and 0.4%) provided better conditions for plant development under stress. A linear increment in plant height, stem diameter, leaf area, leaf dry matter, stem dry matter and root dry matter was obtained as a function of ANE doses (Figure 2). Analyzing the water replacement of 100% ETc, lower doses (0.1 and 0.2%) were more responsive to tomato development, the quadratic models of which represent variable behavior as a function of ANE doses (Figure 2).

Table 1. Plant growth parameters and physiological aspects of tomato (Tucaneiro hybrid) in function of two water repositionings and different doses of *Ascophyllum nodosum* extract.

ANE (%)	ETc (%)	Plant Height (m)	Stem Diameter (mm)	Total Leaf Area (cm^2)	Plant Dry Matter (g)			RWC	SPAD
					Leaf	Stem	Root	(%)	Index
Control	70	1.73 ^{c}	12.92 ^{e}	6040.7 ^{c}	55.65 ^{d}	37.52 ^{c}	6.69 ^{c}	74.87 ^{c}	44.50 ^{d}
	100	1.79 ^{bc}	13.42 ^{d}	7222.8 ^{bc}	69.76 ^{c}	41.73 ^{bc}	9.61 ^{ab}	82.36 ^{b}	46.90 ^{c}
0.1	70	1.81 ^{b}	13.60 ^{c}	6328.7 ^{c}	63.17 ^{c}	38.40 ^{c}	6.99 ^{bc}	74.26 ^{c}	46.45 ^{c}
	100	1.91 ^{ab}	14.79 ^{c}	8369.3 ^{ab}	83.99 ^{ab}	51.53 ^{a}	10.33 ^{a}	86.75 ^{ab}	48.48 ^{bc}
0.2	70	1.86 ^{ab}	14.45 ^{c}	8445.8 ^{ab}	76.31 ^{b}	44.19 ^{b}	10.11 ^{a}	86.50 ^{ab}	48.45 ^{bc}
	100	1.95 ^{a}	15.90 ^{a}	9369.1 ^{a}	95.15 ^{a}	56.18 ^{a}	11.19 ^{a}	87.50 ^{ab}	50.80 ^{a}
0.3	70	1.90 ^{ab}	15.32 ^{b}	8994.0 ^{a}	78.89 ^{b}	47.45 ^{b}	10.12 ^{a}	88.02 ^{a}	48.60 ^{bc}
	100	2.00 ^{a}	16.12 ^{a}	9433.2 ^{a}	91.14 ^{a}	54.91 ^{a}	9.57 ^{ab}	89.02 ^{a}	48.73 ^{bc}
0.4	70	1.95 ^{a}	15.20 ^{b}	8794.9 ^{a}	84.60 ^{ab}	52.34 ^{a}	10.69 ^{a}	87.16 ^{ab}	48.28 ^{bc}
	100	1.91 ^{ab}	15.47 ^{b}	8897.7 ^{a}	87.23 ^{ab}	51.73 ^{a}	9.73 ^{ab}	89.74 ^{a}	48.90 ^{b}
CV (%)		4.5	3.5	7.7	7.9	10.1	12.0	5.6	4.6
ANE		***	***	***	***	***	***	***	***
ETc		***	***	***	***	**	*	***	***
ANE × ETc		***	*	*	*	*	*	***	*

ANE (*Ascophyllum nodosum* extract); ETc (crop evapotranspiration); RWC (relative water content); CV (variation coefficient). Mean values followed by different letters in the same column are significantly different according to Tukey's test ($p < 0.05$). *** $p < 0.001$; ** $p < 0.01$; * $p < 0.05$.

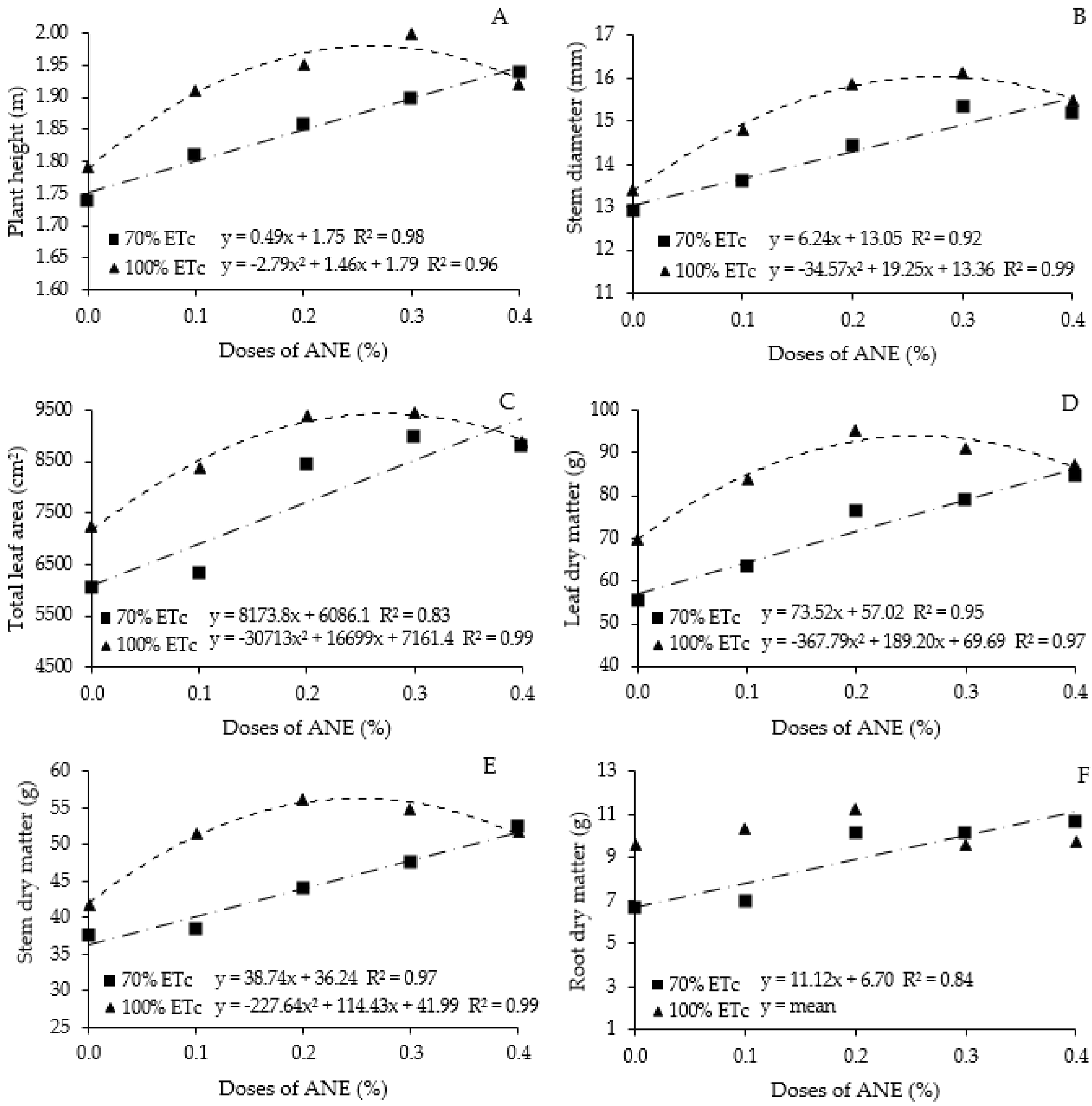

Figure 2. Regression models for the plant growth parameters analyzed ((**A**): plant height; (**B**): stem diameter; (**C**): total leaf area; (**D**): leaf dry matter; (**E**): stem dry matter; (**F**): root dry matter) in function of *Ascophyllum nodosum* extract doses for each condition of water replacement during tomato production. ANE: *Ascophyllum nodosum*; ETc: crop evapotranspiration; R^2: determination coefficient; y: tomato variables; x: *Ascophyllum nodosum* extract doses.

In a deficit irrigation condition, a dose of 0.4% ANE is required to achieve better plant height and leaf dry matter values compared to 100% ETc, which requires only 0.2% ANE. The condition of 100% ETc with 0.2% ANE was the treatment that showed the best increase in stem diameter, whereas 0.3% ANE with 70% ETc was the second best. For stem dry matter, 0.1% ANE with 100% ETc was similar to 0.4% ANE with 70% ETc. In a condition of 100% ETc, there was no significant difference regarding ANE application in the development of root biomass; in this condition without ANE application it was statistically similar to 70% ETc with 0.2% ANE.

3.2. Fruit Yield and Yield Components

The interaction between ANE values with ETc was significant for fruit yield and all yield components evaluated (Table 2). Some yield components were negatively affected by deficit irrigation; without ANE applications there was an 11.4% reduction in bunch length, 9.4% in fruit mass and 9.4% in fruit transversal diameter, causing an 18.3% fruit yield reduction in relation to control plants with 100% ETc.

Plants that received ANE (0.3% and 0.4%) presented the same yield compares to plants with 100% ETc and 70% ETc. However, in unstressed plants, ANE low doses (0.1 and 0.2%) were superior to treatments with deficit irrigation in almost all yield components. A quadratic behavior was observed in plants with 100% ETc (Figure 3); in this condition, ANE (0.2%) provided the highest yield, bunch length and fruit number plant^{-1}, with increments of 54.6%, 23.5%, and 45%, respectively, regarding control treatment.

Plants under deficit irrigation had linear behavior in all yield parameters as a function of ANE doses (Figure 3). The two highest doses (0.3 and 0.4% ANE) provided the best performance, increasing by 43.4% and 50.5%, respectively, in plant yield. It is observed that plants without ANE application and 100% ETc, presented the same yield as plants with 70% ETc and 0.2% ANE.

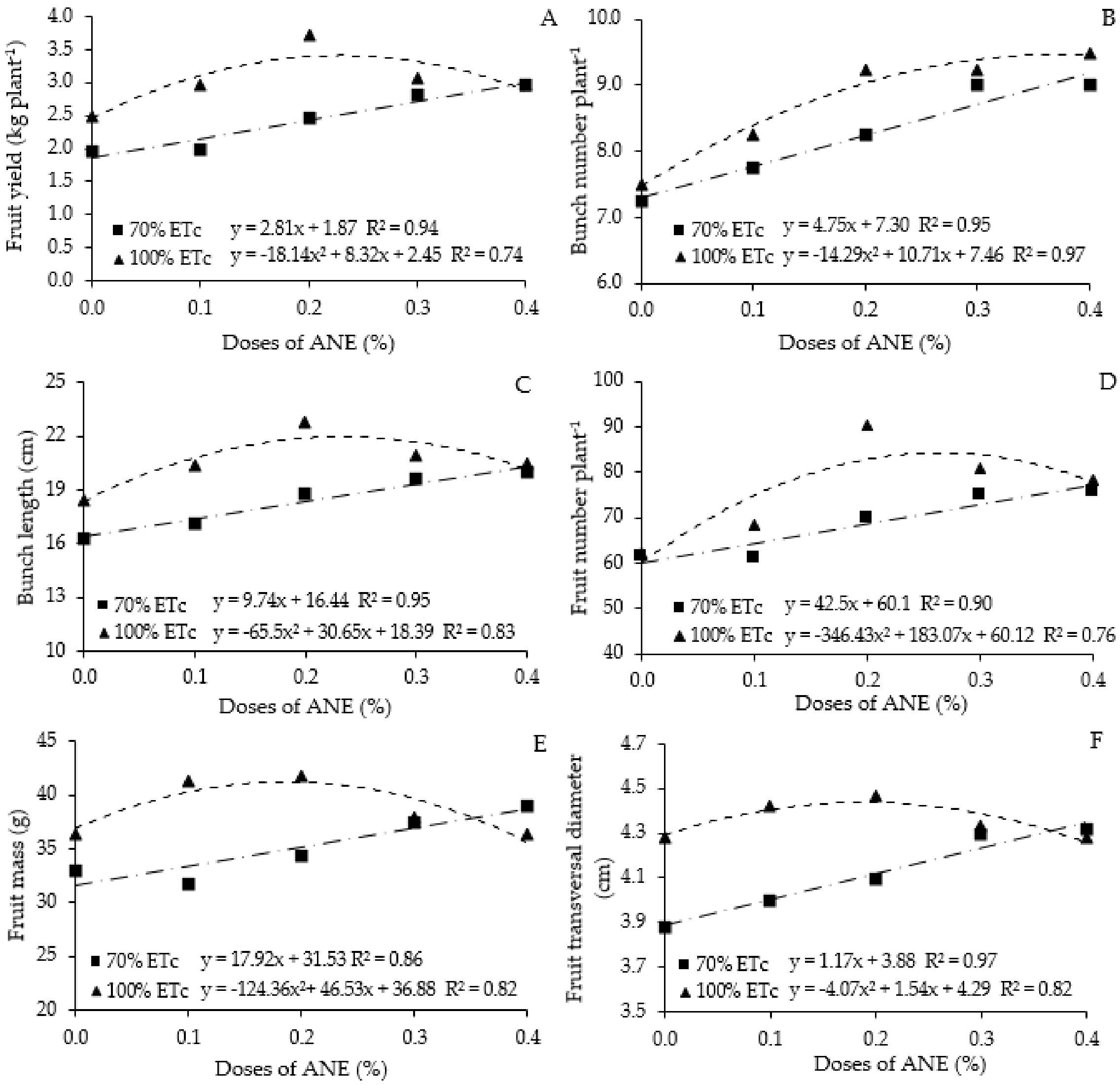

Figure 3. Regression models for the fruit yield and yield components analyzed ((**A**): fruit yield; (**B**): bunch number plant^{-1}; (**C**): bunch length; (**D**): fruit number plant^{-1}; (**E**): fruit mass; (**F**): fruit transversal diameter) in function of *Ascophyllum nodosum* extract doses for each condition of water replacement during tomato production. ANE: *Ascophyllum nodosum*; ETc: crop evapotranspiration; R^2: determination coefficient; y: tomato variables; x: *Ascophyllum nodosum* extract deses.

Table 2. Fruit yield and yield components of tomato (Tucaneiro hybrid) in function of two water repositionings and different doses of *Ascophyllum nodosum* extract.

ANE (%)	ETc (%)	Fruit Yield (kg Plant^{-1})	Bunch Number Plant^{-1}	Bunch Length (cm)	Fruit Number Plant^{-1}	Fruit Mass (g)	Fruit Transversal Diameter (cm)
Control	70	1.96 ^{d}	7.25 ^{c}	16.36 ^{e}	61.50 ^{e}	32.90 ^{d}	3.88 ^{d}
	100	2.40 ^{c}	7.75 ^{bc}	18.46 ^{d}	62.25 ^{e}	36.31 ^{bc}	4.27 ^{b}
0.1	70	1.98 ^{d}	7.75 ^{bc}	17.12 ^{e}	61.00 ^{e}	31.74 ^{d}	4.00 ^{cd}
	100	2.95 ^{b}	8.25 ^{b}	20.37 ^{bc}	68.25 ^{d}	41.26 ^{a}	4.42 ^{a}
0.2	70	2.45 ^{c}	8.25 ^{b}	18.81 ^{d}	69.75 ^{d}	34.43 ^{c}	4.10 ^{c}
	100	3.77 ^{a}	9.25 ^{a}	22.79 ^{a}	90.25 ^{a}	41.75 ^{a}	4.47 ^{a}
0.3	70	2.81 ^{b}	9.00 ^{ab}	19.68 ^{c}	75.00 ^{c}	37.50 ^{bc}	4.25 ^{b}
	100	2.99 ^{b}	9.25 ^{a}	20.94 ^{b}	80.75 ^{b}	37.97 ^{bc}	4.33 ^{ab}
0.4	70	2.95 ^{b}	9.00 ^{ab}	19.95 ^{c}	75.75 ^{bc}	38.98 ^{b}	4.33 ^{ab}
	100	3.00 ^{b}	9.50 ^{a}	20.40 ^{bc}	78.25 ^{b}	36.35 ^{bc}	4.28 ^{b}
CV (%)		5.5	7.5	5.5	6.6	4.9	4.3
ANE		***	***	***	***	**	**
ETc		***	*	***	***	***	***
ANE × ETc		***	*	***	**	***	***

ANE (*Ascophyllum nodosum* extract); ETc (crop evapotranspiration); CV (variation's coefficient). Means values followed by different letters in the same column are significantly different by Tukey test ($p < 0.05$). *** $p < 0.001$; ** $p < 0.01$; * $p < 0.05$.

3.3. Physiological Aspects

The interaction between ANE values with ETc were significant for both physiological aspects, relative water content and chlorophyll content (Table 1). Deficit irrigation imposition during tomato crop caused reduction in RWC and chlorophyll content, compared to unstressed plants. But, associating with ANE application tomato plants were able to increase RWC and chlorophyll content even under water stress. RWC and chlorophyll content were 9.1% and 5.1%, respectively lower in plants under deficit irrigation and without ANE application, related to plants with 100% ETc. However, 0.2% ANE with 70% ETc presented the same RWC compared to 100% ETc without ANE applications.

ANE applications at low doses were able to reduce water stress effect, showing no difference with stressed and unstressed plants for chlorophyll content. RWC tended to increase with an increasing in ANE doses, while chlorophyll content presented a quadratic curve appearance (Figure 4).

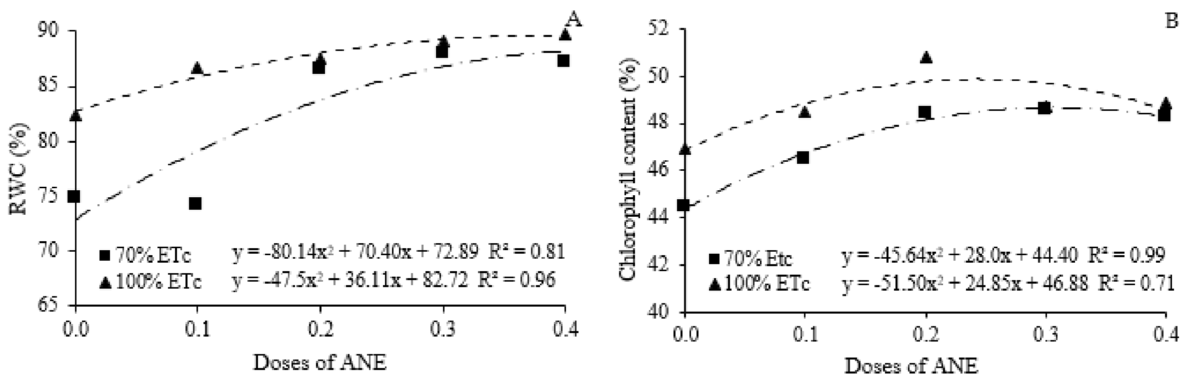

Figure 4. Regression models for the physiological parameters analyzed ((**A**): RWC; (**B**): chlorophyll content) in function of *Ascophyllum nodosum* extract doses for each condition of water replacement during tomato production. ANE: *Ascophyllum nodosum*; ETc: Crop evapotranspiration; R^2: determination coefficient; y: tomato variables; x: *Ascophyllum nodosum* extract doses.

3.4. Fruit Quality

The interaction between ANE values with ETc were significant for fruit quality parameters (Table 3). Fruit produced without ANE application under water stress in relation to 100% ETc presented 5.3% less acidity, with 3.2% pH increment and 16.5% in SST, reflecting an enhancement in fruit ratio. Total polyphenols increased 17.4%. Comparing the parameters, it is observed that most of the fruit quality were favored with deficit irrigation. Under 70% ETc low doses of ANE (0.1 and 0.2%) presented the best values in all evaluated parameters. As function of increasing ANE doses, fruit total acidity decreased, while pH values increased (Figure 5), ANE (0.1%) provided the greatest polyphenols accumulation for both irrigation condition, corresponding to 34.5% and 1.2% for 100% and 70% of ETc compared to control. With deficit irrigation ANE (0.1%) increased 13.4% TSS compared to control.

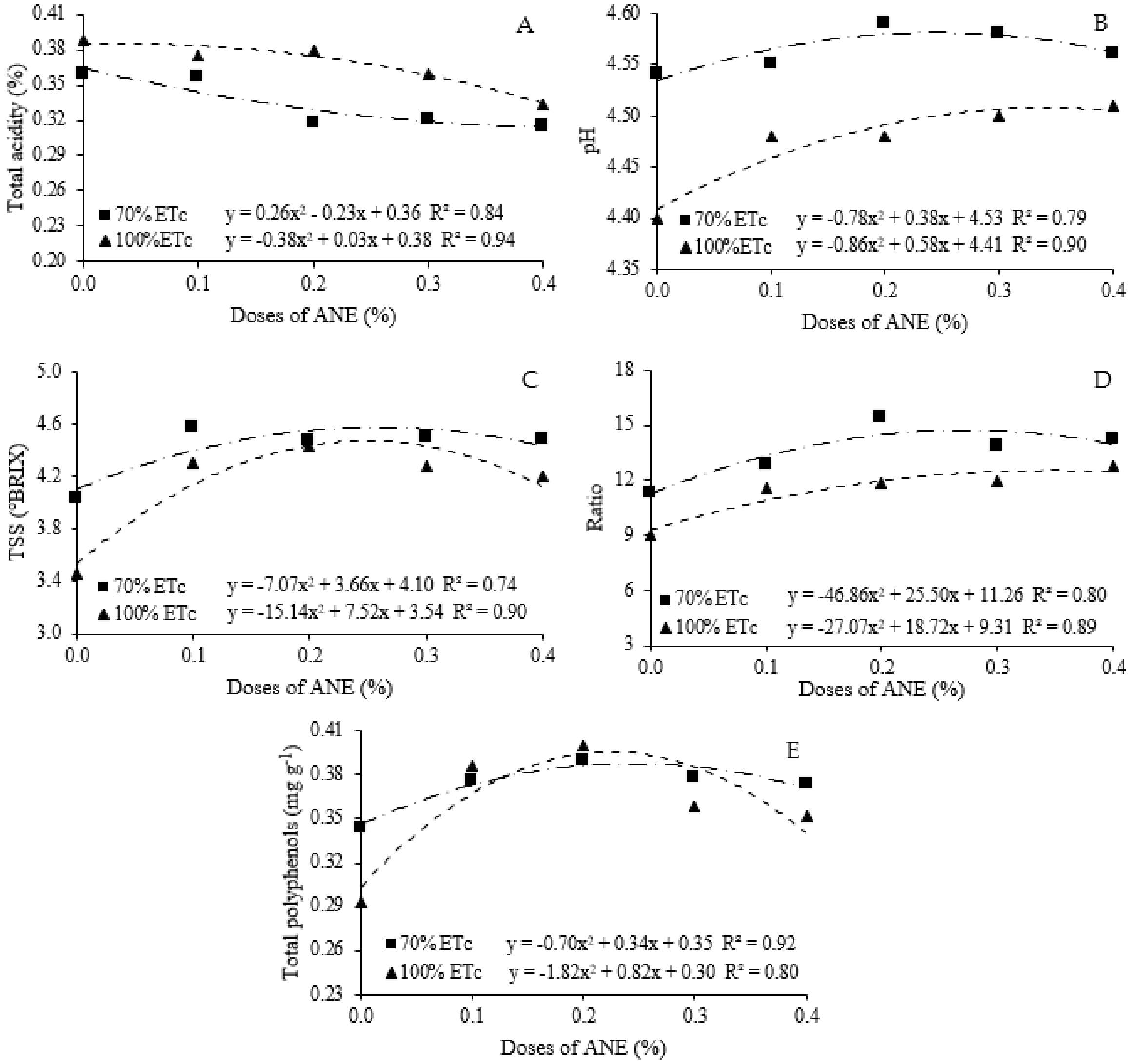

Figure 5. Regression models for the fruit quality parameters analyzed ((**A**): total acidity; (**B**): pH; (**C**): TSS; (**D**): ratio; (**E**): total polyphenols) in function of *Ascophyllum nodosum* extract doses for each condition of water replacement during tomato production. ANE: *Ascophyllum nodosum*; ETc: Crop evapotranspiration; R^2: determination coefficient; y: tomato variables; x: *Ascophyllum nodosum* extract doses.

Table 3. Fruit quality parameters of tomato (Tucaneiro hybrid) in function of two water repositionings and different doses of *Ascophyllum nodosum* extract.

ANE (%)	ETc (%)	Total Acidity (%)	pH	TSS (°BRIX)	Ratio	Total Polyphenols (mg g^{-1})
Control	70	0.36 ^{b}	4.54 ^{ab}	4.03 ^{c}	11.33 ^{bc}	0.34 ^{c}
	100	0.39 ^{a}	4.40 ^{c}	3.46 ^{d}	8.99 ^{d}	0.29 ^{d}
0.1	70	0.36 ^{b}	4.55 ^{ab}	4.57 ^{a}	12.87 ^{b}	0.38 ^{ab}
	100	0.38 ^{a}	4.48 ^{b}	4.30 ^{b}	11.59 ^{bc}	0.39 ^{a}
0.2	70	0.32 ^{c}	4.59 ^{a}	4.47 ^{a}	15.45 ^{a}	0.39 ^{a}
	100	0.38 ^{a}	4.48 ^{b}	4.43 ^{ab}	11.84 ^{bc}	0.40 ^{a}
0.3	70	0.32 ^{c}	4.58 ^{a}	4.50 ^{a}	13.87 ^{ab}	0.38 ^{ab}
	100	0.36 ^{b}	4.50 ^{b}	4.28 ^{b}	11.98 ^{bc}	0.36 ^{b}
0.4	70	0.32 ^{c}	4.56 ^{ab}	4.48 ^{a}	14.21 ^{ab}	0.37 ^{b}
	100	0.33 ^{c}	4.51	4.20	12.74 ^{b}	0.35 ^{c}
CV (%)		7.1	1.0	3.2	6.9	4.2
ANE		**	*	***	***	***
ETc		***	***	***	***	**
ANE × ETc		*	*	*	*	**

ANE (*Ascophyllum nodosum* extract); ETc (crop evapotranspiration); CV (variation coefficient); TSS (total soluble solids). Mean values followed by different letters in the same column are significantly different according to Tukey's test ($p < 0.05$). *** $p < 0.001$; ** $p < 0.01$; * $p < 0.05$.

3.5. Pearson Linear Correlation

Table 4 shows the correlation between some variables analyzed. It is observed that fruit yield positively correlates with bunches number plant^{-1} and with all morphologic and physiological parameters analyzed, showing a correlation of > 0.82, of which the largest correlations were with shoot biomass (0.96) and chlorophyll content (0.92). However, it presented a low correlation with fruit quality parameters.

Table 4. Pearson correlation between tomato parameters.

	Foliar Area	Shoot Biomass	Yield	Bunch Number	Total Soluble Solids	Total Polyphenols	Relative Water Content	Chlorophyll Content
Foliar area	1.00	-	-	-	-	-	-	-
Shoot biomass	0.96	1.00	-	-	-	-	-	-
Yield	0.89	0.96	1.00	-	-	-	-	-
Bunch number	0.92	0.91	0.84	1.00	-	-	-	-
Total soluble solids	0.35	0.31	0.24	0.39	1.00	-	-	-
Total polyphenols	0.43	0.43	0.42	0.37	0.92	1.00	-	-
Relative water content	0.96	0.90	0.82	0.87	0.18	0.27	1.00	-
Chlorophyll content	0.92	0.94	0.92	0.85	0.39	0.53	0.85	1.00

4. Discussion

Several studies have reported water reduction impacts in agricultural crops [4,14,31–34]. In this study, the effect of *Ascophyllum nodosum* extract applications in different doses by fertigation during the tomato crop cycle (Tucaneiro hybrid) was evaluated under two different irrigation conditions (100% of ETc and 70% of ETc), in order to analyze the impacts on plant growth, fruit yield, physiological aspects and fruit quality. The results showed an improvement in these parameters through the significant interaction between ANE values and ETc, and corroborated to create strategies for water use managing in tomato crop under subtropical conditions.

For plants under deficit irrigation, their development is negatively compromised; in this condition leaf growth is compromised by affecting the physio-biochemical processes which are important for maintaining plant metabolism [8]. The reduction in vegetative parameter development can be attributed to negative impacts on the photosynthesis process and transpiration rate [14]; possibly, fewer metabolites would be synthetized to promote plant growth.

Chlorophyll content tends to decrease under deficit irrigation conditions, being an important parameter for plant's water status [11], and whose reduction favors reactive oxygen species (ROS) production related to lipid peroxidation and chlorophyll degradation [35]; moreover, under water stress conditions, stomatal conductance decreases, causing plants to close stomata frequently [36]. It can be inferred that ANE provided better conditions for plant development under water stress, resulting in more chlorophyll content and water plant status, and more favorable conditions to promote plant growth and obtain high yields. ANE increases physiological performances under water stress [37], which can be related to higher leaf chlorophyll content. Several molecules present in seaweed extract could act by tolerating deficit irrigation by stomatal regulation, photosynthetic efficiency and activating antioxidative defense systems [19].

Regardless of ANE application, the best fruit yields were obtained with 100% ETc. The reduction in tomato yield under deficit irrigation can be the result of an increment in soil osmotic pressure due to low soil water content, attributed to water restriction during the tomato crop cycle, leading to low water uptake by the roots [6,38]. In addition, the root biomass reduced under this situation, resulting in less exploring area for nutrient acquisition, compromising plant development.

This study showed that plants under full irrigation presented better growth and fruit yield compared to plants submitted to deficit irrigation. These results changed with the seaweed extract application, which was able to attenuate the water stress effect, increasing root biomass and providing a greater area for soil exploration and nutrient absorption [39]. It positively influenced fruit yield increment, and suggests that ANE applied through fertigation would enhance soil fertility, increasing absorption and nutrient mobilization by plants [20], and consequently having better vegetative development, mainly attributed to the benefit which compounds supply which are present in seaweed extract, such as betaines and phytostimulatory substances [17]. The interaction with the ANE application demonstrates the ability of the biostimulant action of the seaweed extract to mitigate the negative effects caused by water stress, providing better plant development.

Plants that received ANE applications presented more favorable conditions for increasing morphological aspects, such as the leaf biomass responsible for intercepting light required for photoassimilate production, which are used by plants to improve yield components. The significant chlorophyll content and RWC increase under water stress can also assist plants to restore leave photosynthetic activity [2].

Abscisic acid (ABA) biosynthesis is related to plant stress; ABA accumulation reduces water loss by closing stomata, an important strategy under stress conditions [8]. Analyzing RWC in plants which received ANE applications suggests that plants keep their leaves more turgid and exhibit greater RWC; possibly, the transpiration rate was little compromised, even under water stress.

In plant metabolism, seaweed extract acts by regulating salicylic and jasmonic acid-signaling pathways, providing better health conditions that favor plant development and yield [40]. According to Goñi et al. [2], ANE reduces lipid peroxidation damage in leaf cell membranes which resulted from the photo oxidation process by ROS, favoring plant development by reducing plant stress and increasing their productive potential, as observed for yield component increments.

Some effects related to deficit irrigation in tomato postharvest characteristics can vary according to duration and water deficit severity, as well as soil physical characteristics and tomato variety [41]. Flowering and fruit formation stages are most sensitive to a water deficit [7]. In this study, plants were managed with deficit irrigation during the entire cycle, and this could have been one of the causes that provided low fruit yield under 70% ETc. These phenological phases present high transpiration rate and consequently the greatest water demand, which can influence the reduction in fruit yield [4,8].

Regulated deficit irrigation enhances some secondary metabolites such as carotenoids and phenolic components in plant tissues [6], affecting tomato nutritional value, mainly due to an antioxidant activity increase in tomatoes [9]. High total polyphenols from tomato under moderate water stress could resulted from a defense strategy against this abiotic stress [32]. Less water availability for tomato crop absorption could have resulted in a higher soluble solid concentration and consequently presented fruit with a better flavor ratio [14,41].

ANE promotes biosynthesis and phenolic compound accumulation in leaves, improves leaf gas exchanges and protects the photosystem II, resulting in an environment with less stress conditions [42]. ANE applications induce plant defense enzyme activities, such as polyphenol oxidase and phenylalanine ammonia lyase, correlated with phenolic content elevation, acting in the plant defense system and helping to promote plant growth [17,20]. Besides improving tomato plant parameters, ANE application was able to enhance the chemical and bioactive compounds in the tomato, mainly due to the seaweed composition, such as polysaccharides and phenolic compounds [18].

Under deficit irrigation, the higher the seaweed extract dose, the better the tomato vegetative, productive and fruit quality performance, in contrast to the optimal condition of irrigation (100% ETc), in which intermediate doses provided the best results. It can be inferred that those plants under deficit irrigation need a larger amount of beneficial compounds from ANE than unstressed plants for use in metabolic pathways to attenuate water stress conditions.

The results show that there is the possibility of producing tomatoes with an amount of water less than the optimal amount, using deficit irrigation of 70% ETc, in this case associated with the application of seaweed extract. A great advantage of this irrigation strategy is that it produces more with less water, it collaborates with the sustainability of world agricultural production, and may be a strategy in regions with limited water supply [43,44].

Therefore, the development of new tomato crop management techniques such as ANE application in the tomato crop (Tucaneiro hybrid) under deficit irrigation, has great potential to reduce the amount of water used during the crop cycle and increase agronomic and fruit quality aspects, contributing to improving fruit yield with less water use.

5. Conclusions

In this research, morphological and physiological tomato plants responses are negatively affected under deficit irrigation. However, the research presented a significant interaction between ANE applications, increasing plant height, stem diameter, plant biomass, leaf area, chlorophyll and relative water content. Tomato quality parameters were also favored under deficit irrigation and seaweed extract applications, and it was observed that most of the quality parameters were favored with 70% ETc. Under deficit irrigation, the maximum fruit yield was obtained with ANE (0.3 and 0.4%), which enhanced by 43.4 and 50%, in relation to untreated plants. However, with 100% ETc, ANE (0.2%) increased by 54.6% the fruit yield in relation to the control treatment. This demonstrates that under deficit irrigation, a higher dose of ANE is needed to increase yield parameters, but with no deficit, the intermediate dose was already sufficient to achieve similar results. Both with 70% ETc and applications of ANE (0.2%), tomato plants presented similar fruit yields compared to plants with 100% ETc without ANE applications, evidencing the effectiveness in alleviating water stress effects in the tomato crop, improving tomato yield and quality.

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