



Article

Effects of Salt Water on Growth and Quality of *Raphanus sativus* L. and Physiological Responses against Salt Stress

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Abstract: To determine the optimal salinity of irrigation water for fruit radish cultivated in peat, five levels of salinized water were used to evaluate their effect on the growth and quality of fruit radish (*Raphanus sativus* L.). Results showed that with an increase in salinity, the leaf growth was somehow inhibited, but the fleshy root growth increased, and quality improved with more soluble solids, sugar, protein, and Vitamin C substances in fleshy roots. With an increase in water salinity up to 4.2 dS/m, the weight of fleshy roots increased by 51.10% with a high increment in the root/shoot ratio. With the same salt concentration, the content of soluble solids in both root peel and pulp was the highest and improved by 11.06% and 6.70%, respectively. The soluble sugar content was the highest in root peels with the 4.2 dS/m treatment and in fleshy roots with the 7.4 dS/m treatment, with a 55.85% and 32.30% increase, respectively. The content of both soluble protein and vitamin C with the 4.2 dS/m treatment increased by 11.99% and 113.36%, respectively. Strong evidence derived from the study has indicated that 4.2 dS/m salinized irrigation water is optimal for growing ‘ice-cream’ fruit radishes and maintaining ultimate root weight and quality.

Keywords: salinized irrigation water; fruit radish; root/shoot ratio; fleshy root weight; fleshy root quality



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1. Introduction

Approximately 1.1×10^9 hm² of saline soil is distributed in more than 100 countries and regions around the world, and the acreage of soil salinization is still on the rise [1]. There is 3.6×10^7 hm² salinized land in China, accounting for 4.88% [2] of the arable farm soil. In the meantime, in the salinized regions where fresh water is usually in shortage and unavailable for agricultural production in colossal quantity. To make it possible to sustain agriculture in saline lands, mildly salty water has gradually been used for farming as an unconventional water source [3]. In recent years, many scholars have carried out research on saline water irrigation and crop salt tolerance mechanisms [4,5]. Excessive salinity in soil or irrigation water will lead to ion imbalance and osmotic interference, affecting crop growth, yield, and quality [6]. However, some studies have shown that moderate salt stress can promote the growth of plant roots, improve the root/shoot ratio of plants, cultivate strong seedlings, and significantly improve the quality of crop product organs [7–9]. Qi et al. [10] found that salt stress could significantly increase the root biomass and root/shoot ratio of *Robinia pseudoacacia* seedlings. Ouyang et al. [11] pointed out that a low salinity irrigation could promote the growth and yield of cucumber fruit, but it was not conducive to the accumulation of sugar in fruit, while a high salinity irrigation was more effective in improving fruit quality. Akrami et al. [12] found that melons could increase the content of proline and soluble sugar in plants with different salt concentrations. Gong

et al. [13] demonstrated that under saline–alkali stress, the proline content in the leaves of *Toona sinensis* seedlings increased along with various increased salt concentrations, while the content of soluble protein and soluble sugar increased first and then decreased. Yao et al. [14] indicated that the content of soluble sugar, soluble protein, and proline in cherry radish leaves increased significantly under salt stress, while Šmídová et al. [15] found that salt stress could have increased the nutritional value in tomato fruits. It seems that various research findings have demonstrated different results due to different salinity levels and test crops. Therefore, it is critically important to determine the threshold of the salinity level in irrigation water under which the crop yield is not affected but the quality increases [16].

Peat substrate as a widely used substrate for experiments in greenhouses or as soil amendment material for field trials in saline lands has attracted much attention due to its properties of high aeration, permeability, and cation exchange capacity (CEC) [17]. Radish (*Raphanus sativus* L. (Brassicaceae)) belongs to root vegetables and is theoretically inclined to improve its growth and quality of fleshy roots. Being popular for its crunchy feature and sweet taste, the ‘ice-cream’ fruit radish is one of the vegetables grown in saline soils from late autumn to early winter in coastal areas of China. Due to its tolerance against salt stress, it has become an ideal vegetable species for saline-water-irrigated cultivation. However, the mechanism pertaining to radish salt tolerance remains unknown, and the salinity threshold of brackish water set forth for radish quality improvement has not been undetermined at present. With this study, the ‘ice-cream’ fruit radish cultivated in peat substrate would be used and irrigated with irrigation water at different salinity levels in a greenhouse to evaluate their effect on the growth, quality, and physiological responses of radish against salt stress so as to reveal the salt tolerance mechanism and to promote the cultivation of ‘ice-cream’ fruit radish with salt water, providing a theoretical basis for understanding radish salt tolerance.

2. Materials and Methods

2.1. Temperature and Humidity in the Greenhouse

The experiment was carried out in a greenhouse with Weifang University of Science and Technology, located in a region under a temperate monsoon climate with a 12.3 °C annual average temperature, a 650 mm annual average precipitation, and the 2607.4 h of annual average sunlight hours (Figure 1).

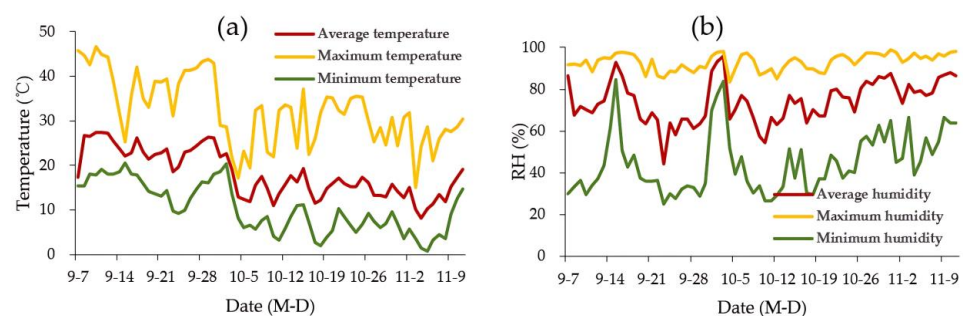


Figure 1. Daily changes in temperature (a) and relative humidity (RH) (b) in the greenhouse.

2.2. Experiment Design and Planting Plots

On 7 September 2022, Fruit radish (*R. sativus* L.) seedlings were cultivated in five containers (4 m long, 20 cm wide, and 16 cm deep), 10 in each container and 40 cm apart. Each container was filled with 10 cm thick peat substrate containing 1.5–3.5% ashes and 96.5–98.5 organic materials (H = 55–65%, pH = 5.5–6.5). A dripping irrigation line with an emitter for each plant was set up to supply water. A univariate and fully randomized trial design was used in this study. Five water salinity treatments were designated as 1.0 dS/m (CK), 2.6 dS/m, 4.2 dS/m, 7.4 dS/m and 10.6 dS/m.

Each of the irrigation water treatments was prepared by mixing calculated sea salt with local groundwater (CK) accordingly (Table 1). After transplanting, all seedlings were

first watered with fresh well water and then followed with salt water subsequently once every 4 days, 300 mL per plant with 3 g of compound fertilizer ($N + P_2O_5 + K_2O$ 20-20-20 + TE, TE containing $Fe + Zn + B \geq 0.21\%$). All radish plants were routinely maintained according to normal radish cultivation guidelines in the field. Mature radish roots were harvested on 11 November, and all growth indexes and quality indicators for leaves and fleshy roots were measured immediately after harvest.

Table 1. Ion concentration in different salinized irrigation water treatments.

Irrigation Water Salinity (dS/m)	Ion Concentration (mg/L)						
	Na^+	Ca^{2+}	Mg^{2+}	K^+	Cl^-	HCO_3^-	SO_4^{2-}
1.0	53	54.4	45.2	5.5	104.2	311.1	44.3
2.6	431.4	60.1	46.1	10	683.9	316	62.6
4.2	809.8	65.8	47	14.5	1263.6	320.9	80.9
7.4	1566.6	77.2	48.8	23.5	2423	330.7	117.5
10.6	2323.4	88.6	50.6	32.5	3582.4	340.5	154.1

2.3. Measurement of Growth and Quality Parameters

2.3.1. Growth Indexes

After harvesting, the growth indexes of all 10 radish plants of each treatment were evaluated, and 8 measurements of each growth index were calculated for mean after removing the highest and lowest readings as outliers. The growth indexes include the leaf area and leaf weight, the root length and width (diameter), root weight, and root volume.

The leaf area of radish was measured following the leaf area formula developed by Chen et al. [18], and the fresh leaf weight was measured using an analytical balance with an accuracy of 0.001 g.

The root pulp of each radish plant within each treatment was measured for its length and diameter in the middle. The root volume was measured by a water drainage method, through which the flesh radish root was submerged in a large container filled with water to measure the volume difference before and after removing the submerged root from the water in the container. The fresh weight of each flesh root was measured by an analytical balance with an accuracy of 0.001 g.

The root/shoot ratio was calculated by flesh root weight and all fresh leaf weight through the following formula:

$$\text{Root/shoot ratio (R/S)} = \text{Fresh weight of radish fleshy roots} / \text{Fresh weight of all radish leaves}$$

2.3.2. Quality Indicators

To determine the quality of fruit radish, four fleshy roots uniform in growth from each treatment were chosen; their 1/3 portion in the middle was cut and separated for their peel and pulp, respectively. Both peel and pulp samples were ground thoroughly to be further analyzed.

The content of soluble sugars and soluble solids in fruit peel and pulp samples was determined using the anthraquinone colorimetric method (630 nm) and digital sugar meter (LC-DR-32B, Shanghai Lichen Instrument Technology Co., Ltd., Shanghai, China) [19].

Measure vitamin C (Vc) using a reduced ascorbic acid (ASA) assay kit, and adjust the wavelength of the UV spectrophotometer to 420 nm to measure the absorbance values of each tube. The BCA protein content determination kit measures soluble proteins and adjusts the wavelength to 562 nm to measure the absorbance values of each tube. (UV spectrophotometer, Shanghai Prism Technology Co., Ltd., Shanghai, China; reagent kit purchased from Suzhou Keming Biotechnology Co., Ltd., Suzhou, China) [19].

2.3.3. Other Indicators

Electrical conductivity (EC) and pH in different plant parts were measured by chopping leaf and root samples into small pieces after removing peels, grinding each sample in a juicer, and collecting the juice in a test tube for further use. Each radish juice sample was mixed with deionized water at a ratio of 1:5, stirred thoroughly, and measured using a conductivity meter (Thunder Magnetic DDS-307A, Shanghai Yidian Scientific Instrument Co., Ltd., Shanghai, China) and pH meter (Thunder Magnetic PHS-2F, Shanghai Yidian Scientific Instrument Co., Ltd., Shanghai, China).

2.4. Data Collection and Analyses

All data collection and statistical analysis were performed using Excel and IBM SPSS Statistics 26.

3. Results

3.1. Effect of Different Salinity Levels in Irrigation Water on EC and pH Values of Substrate

The EC value in the substrate at harvest demonstrated a linear upward trend (Figure 2a), and there was a significant difference between treatments, indicating that the saltwater irrigation has effectively maintained various salinity in the substrate as expected through the experiment. However, the pH value remained at a constant level between 6.53 and 6.82 (Figure 2b), suggesting that different water salinity did not change soil pH over the growing season.

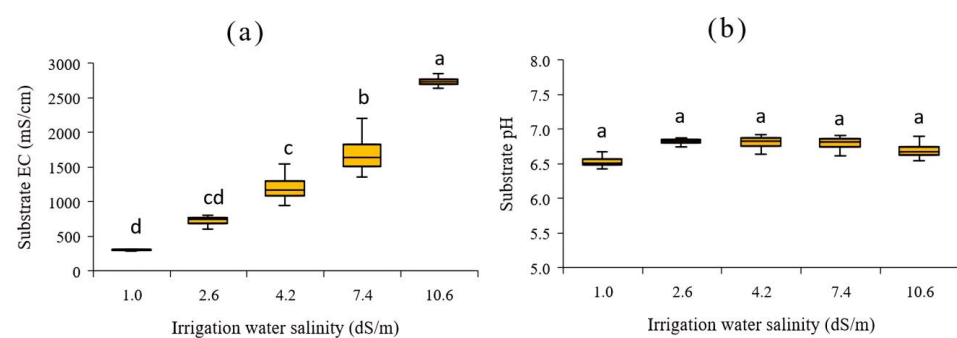


Figure 2. EC (a) and pH (b) value at harvest with each of five salinity levels in peat substrate irrigated with salt water ($n = 4$, $p < 5\%$). The box plot includes a minimum, median, IQR, maximum, and outlier number, which applies to all of the following charts. Different lowercase letters indicate significant differences between treatments at 0.05 level.

3.2. Effect of Different Salinity Levels in Irrigation Water on the Growth of Fruit Radish

3.2.1. Effect of Different Salinity Levels in Irrigation Water on the Growth of Stems and Leaves

Compared with the CK (1.0 dS/m) treatment, the fresh leaf weight and the area of a single plant were significantly reduced in all treatments at a significantly different level. At the same time, the differences in the leaf area and the fresh leaf weight with 2.6 dS/m, 4.2 dS/m, 7.4 dS/m, and 10.6 dS/m treatments were not significant (Figure 3), indicating that leaves of 'ice-cream' fruit radish were sensitive to salinity in irrigation water, and their growth was significantly inhibited when the salinity level in irrigation water exceeded 2.6 dS/m.

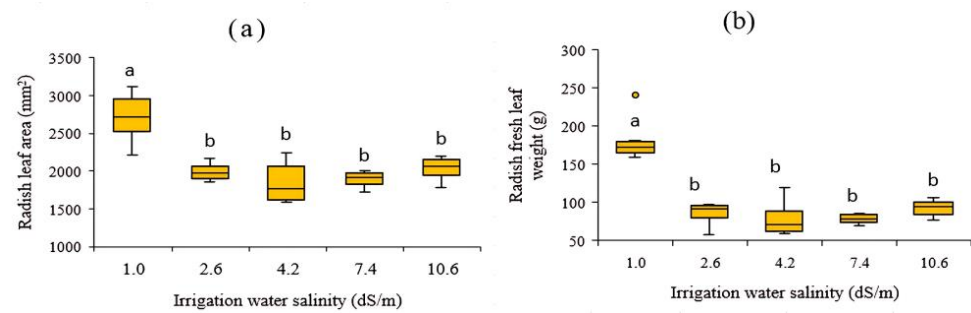


Figure 3. Comparison of radish leaf area (a) and weight (b) between different salinity levels in irrigation water ($n = 8$, $p < 5\%$). The dots in the box plot represent outlier numbers, which also applies to all of the charts below. Different lowercase letters indicate significant differences between treatments at 0.05 level.

3.2.2. Effect of Different Salinity Levels in Irrigation Water on the Growth of Fleshy Roots of Fruit Radish

The diameter, length, volume, and weight of radish fleshy root increased when the salinity level in irrigation water was low or medium, and the increase in these parameters was maximized with the 4.2 dS/m treatment (Figure 4). The average root volume with the 4.2 dS/m treatment was significantly higher than that with the CK treatment (Figure 4c), but there were no significant differences between other treatments and CK treatment. The average root weight with the 2.6 dS/m, 4.2 dS/m, and 7.4 dS/m treatment was significantly higher than that with CK treatment, and the increase was 35.11%, 51.10%, and 39.09%, respectively (Figure 4d). It has been shown that a salinity level of 4.2 dS/m in irrigation water had the best effect on the fleshy root growth of fruit radishes.

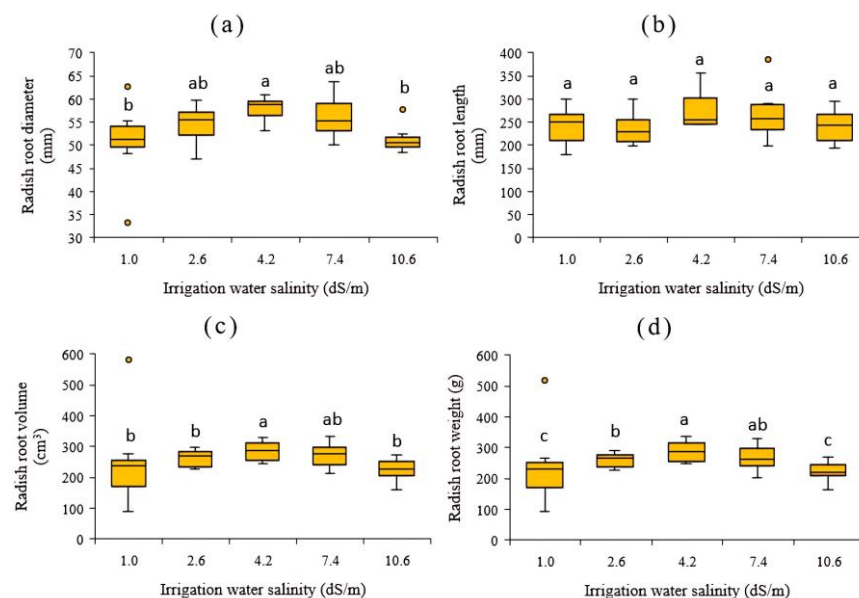


Figure 4. Diameter (a), length (b), volume (c), and weight (d) of fleshy radish roots under different salinity levels in irrigation water ($n = 8$, $p < 5\%$). Different lowercase letters indicate significant differences between treatments at 0.05 level.

3.2.3. Effect of Different Salinity Levels in Irrigation Water on Root/Shoot Ratio of Fruit Radish

The root/shoot ratio of fruit radish plants under saltwater irrigation indicated a trend of an increase first at a low salt concentration in irrigation water and then a decline with a further rise in the salinity level (Figure 5). Root/shoot ratios with each of all treatments were 1.8 to 2.9 times higher than those of CK treatment. The root/shoot ratio of radish plants in

the 4.2 dS/m treatment reached the highest value, indicating that saltwater irrigation at a lower concentration could have promoted the growth of succulent roots and significantly inhibited leaf growth, thereby increasing its root/shoot ratio.

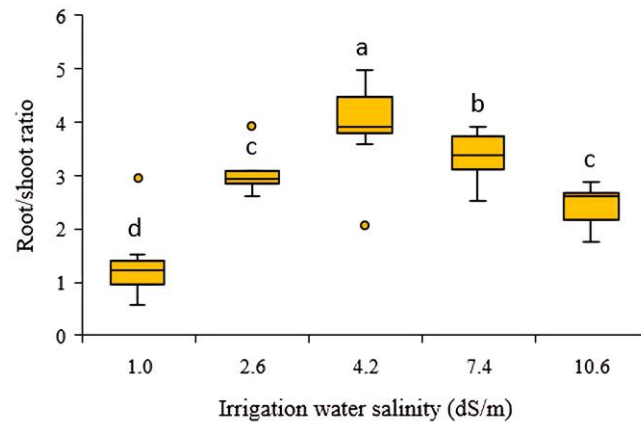


Figure 5. The Root/shoot ratio of fruit radish plants irrigated with salt water at various salinity levels ($n = 8$, $p < 5\%$). Different lowercase letters indicate significant differences between treatments at 0.05 level.

3.3. Effect of Different Salinity Levels in Irrigation Water on EC and pH Value of Fruit Radishes

EC values in leaves, root peel, and root pulp (Figure 6a) showed that the EC concentration was high in leaves, medium in root peels, but low in root pulps, indicating that water potential in leaf cells was lower than that in fleshy roots to maintain a continuous water flow from roots to leaves. In this test, the EC value in radish leaves did not fluctuate significantly with an increase in salinity in irrigation water, but it increased in both root peels and pulp rapidly with a higher salinity level, especially the EC value in root peels and pulp reached the highest level in the 10.6 dS/m treatment. The accumulation of Na^+ and Cl^- in fleshy roots irrigated with salt water was significantly higher than that in leaves, resulting in a decrease in the water transport from roots to leaves under saltwater irrigation and seriously affecting the leaf growth.

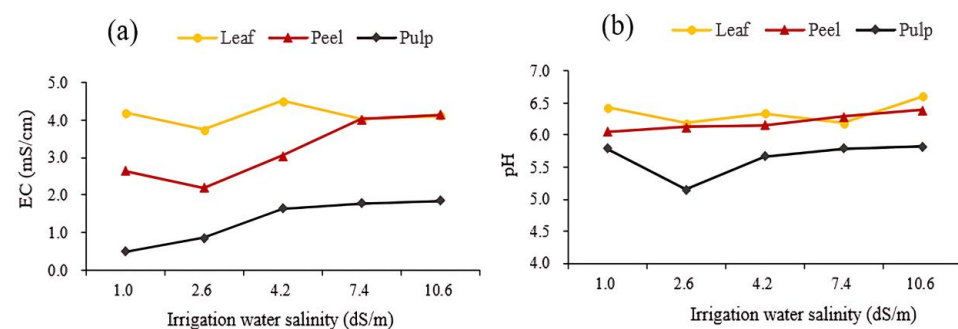


Figure 6. EC (a) and pH (b) values of fruit radishes treated with different concentrations of salt water.

The basic trend of pH values in various salinity treatments indicated that the pH value in leaves is greater than that in root peels and fleshy roots (Figure 6b). The pH value in leaves showed no significant changes with salinity levels in irrigation water, while its value in root peels showed a slight increase with the increase in water salinity in comparison with that with CK treatment. There were no significant changes in the pH value in fleshy roots with the 4.2 dS/m, 7.4 dS/m, and 10.6 dS/m treatment, but a certain decrease of pH in the 2.6 dS/m treatment, indicating that salinity levels in irrigation water had little effect on pH in radish leaves and fleshy roots.

3.4. Effect of Different Salinity Levels in Irrigation Water on Quality of Fruit Radish

3.4.1. Effect on the Soluble Solids and Soluble Sugar Content

The salinity level of irrigation water had a significant effect on the content of soluble solids and soluble sugars in fleshy roots. The content of soluble solids and sugars in both root peel and pulp showed a tendency to increase and then decrease with the increased salinity level in irrigation water. The maximum soluble solids content in both peel and pulp of fleshy roots appeared in the 4.2 dS/m treatment, accounting for an 11.06% and 6.70% significant increase, respectively, compared to CK treatment (Figure 7a). The soluble solid content in the root peels of each treatment was significantly higher than that in fleshy roots, and about 22.98%, 17.58%, 27.91%, 23.17%, and 53.28% higher than that in fleshy roots, respectively (Figure 7a). The maximum soluble sugar content in the fleshy root peels appeared in the 4.2 dS/m treatment, while the maximum value in root pulp appeared in the 7.4 dS/m treatment, compared with CK treatment, it increased by 55.85% and 32.30%, respectively (Figure 7b). The soluble sugar content in the root peels with CK treatment, 2.6 dS/m, and 4.2 dS/m treatment was significantly higher than that in the fleshy roots. However, there was no significant difference between root peels and pulp with the 7.4 dS/m and 10.6 dS/m treatment (Figure 7b). Therefore, to promote an accumulation of soluble solids and sugars in the fleshy roots of fruit radishes, an optimized salt concentration in irrigation water should be administrated around 4.2 dS/m.

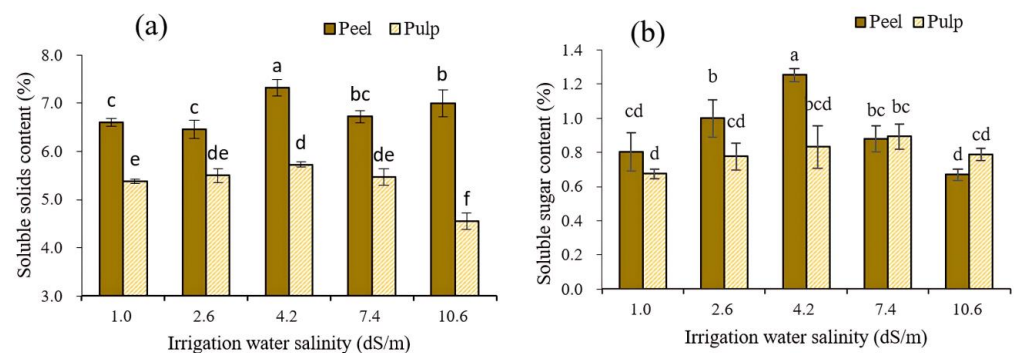


Figure 7. Soluble solids (a) and soluble sugar content (b) of radish fleshy roots treated with different concentrations of salt water ($n = 4$, $p < 5\%$). Different lowercase letters indicate significant differences between treatments at 0.05 level.

3.4.2. Effects on Soluble Protein and Vitamin C Content

With an increase in salinity level in irrigation water, the content of soluble protein in fleshy roots increased first and then decreased among treatments. The soluble protein content increased by 26.78%, 11.99%, 11.42%, and 21.76%, respectively, with 2.6–10.6 dS/m treatments compared to CK treatment. (Figure 8a). The Vc content in fleshy roots also increased first and then decreased along with the increase in salinity levels, which reached the highest in 7.4 dS/m treatment, and the content of Vc in 2.6 dS/m, 4.2 dS/m, 7.4 dS/m and 10.6 dS/m treatment increased by 39.32%, 112.36%, 414.20% and 315.07% compared with CK treatment (Figure 8b). That indicates an optimized concentration of salt in irrigation water could have facilitated the synthesis and accumulation of soluble protein and vitamin C in radish fleshy roots.

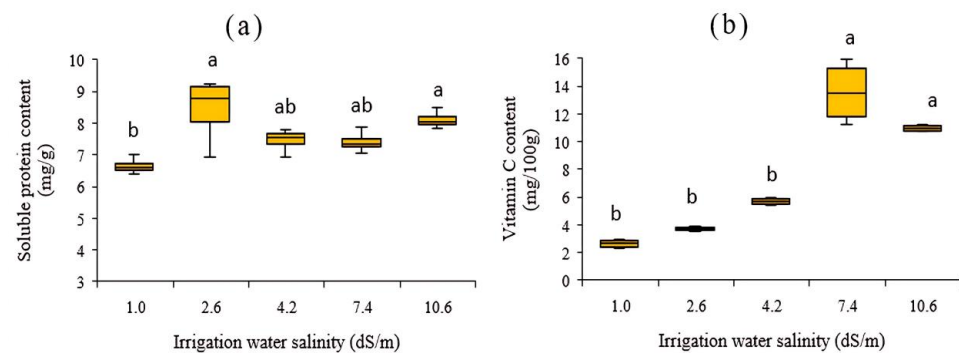


Figure 8. Content of soluble protein(a) and vitamin C (b) in fleshy roots of radish irrigated with salt water with different salinity levels ($n = 4$, $p < 5\%$). Different lowercase letters indicate significant differences between treatments at 0.05 level.

4. Discussion

The results derived from this experiment have demonstrated that the pH in the peat substrate was not significantly affected by salinity levels in irrigation water, while the EC value showed a significant upward trend with the increase in salinity, suggesting that the salinity level in salt water had more Na^+ and Cl^- for a higher EC value in the substrate. However, the effect of peat substrate on pH value is relatively insignificant, which is consistent with the research findings observed by Saqib et al. [20] and Watanabe et al. [21]. In addition, due to a high CEC, peat substrate has a strong buffering capacity to prevent Na^+ from entering plant roots. The unique buffering capacity of peat substrate may well be one of the reasons why the growth of fruit radish leaves was somehow inhibited at low concentrations of water salinity below 2.6 dS/m, but so when the salinity level water was over 2.6 dS/m and up to 10.6 dS/m.

It is obvious that the vegetative (leaf and shoot) growth of ‘ice cream’ fruit radish was significantly inhibited with irrigation with salt water, shown by a significantly reduced fresh leaf weight and area due to salt stress that may have interfered with photosynthetic, osmotic potential, water potential, water transpiration rate, leaf temperature, and relative water content of plant leaves under salt stress [22]. Qi et al. [23] and Chartzoulakis et al. [24] have also demonstrated that drought causes an increased salt content and leads to plant water deficiency, which promotes leaf stomatal closure to reduce transpiration and prevents the entry of CO_2 , resulting in a decrease in the ability of plant leaves to obtain water and produce nutrients, and inhibiting stem and leaf growth leaves. In our experiment, the effect of salinity levels in irrigation on radish root growth was insignificant. All root growth indicators such as root length, diameter, volume, weight, and root/shoot ratio were optimized when the salt concentration in irrigation water was maintained below 4.2 dS/m and then gradually decreased along with salinity level rises. The result has strongly demonstrated that mild salt water could and should be beneficial for fleshy root growth and quality of fruit radish, indicating that low concentrations of salt water can promote the growth of radish fleshy roots. Albacete et al. [25] have demonstrated that the growth of tomato roots remained unaffected but the growth of above-ground parts was inhibited, through their analysis of plant biomass allocation. Guo et al. [26] have also confirmed that the low concentration of NaCl has significantly promoted the growth and development of sugar beet. Our results have also indicated that the low salinity level in irrigation water has significantly promoted the fleshy root growth of fruit radishes. The underlying possibility behind the inconsistent growth of fruit radish leaves and fleshy roots under salt irrigation is that the outcome is likely due to high salt content and reduced water potential in the substrate, resulting in reduced water absorption by radish roots and causing a possible temporary physiological drought. However, fleshy roots reduce the water potential of fleshy root cells by increasing the accumulation of Na^+ and Cl^- , especially in the fleshy root peels. Therefore, roots could have taken in as much water as possible

to compensate for high salt levels but transported less water out for leaf growth, thereby causing reduced leaf growth [27]. Another evidence derived from our data of EC values in both leaves and roots has indicated that with an increase in salinity levels in irrigation water, the EC value in fleshy roots rapidly increased, but not in leaves, fully supporting the hypothesis that under salt stress, more salt ions are accumulated in fleshy roots than those in leaves, causing a decrease in root cell water potential, thereby reducing water movement toward leaves. The accumulation of salt ions in roots and prevention of water movement toward leaves may have also played a significant role in protecting leaves from stress and in maintaining their normal photosynthesis [28,29]. In this study, the regulatory mechanism of biomass allocation ratio in different parts of fruit radish under salt stress was revealed from fluctuations of the root/shoot ratio, while the physiological mechanism involved in salt tolerance in fruit radish was derived from the Na^+ accumulation and cellular osmotic regulation by radish fleshy roots and leaves. Currently, related studies have been conducted from combination stresses of Pi starvation and salt stress-responsive micro RNAs of soybean root [30], and *Funneliformis constrictum* modulates polyamine metabolism to enhance tolerance of *Zea mays* L. to salinity [31] have shed light on the salt tolerance mechanism of plants, providing a direction for the study of salt tolerance mechanism in radish.

Previous studies have shown that under low salt stress, plants tend to regulate the content of soluble sugar, the cell osmosis potential, the enhanced expression of stress-responsive proteins, the plant protein content, and the quality of productive organs to a certain extent [32,33]. However, high salt stress can have an irreversible effect on the growth of most plants. High concentrations of salt ions in plant cells can disrupt the structure and activity of intracellular enzymes, causing metabolic disorders and affecting plant photosynthesis [34], which can cause degradation of photosynthetic products and proteins, leading to a decrease in the content of soluble solids, soluble proteins, soluble sugars, and vitamin C. Eventually, crop quality will be seriously affected under high salt stress [35–40]. Our results are strongly consistent with the previous findings by showing a changing trend in the content of soluble solids, sugars, proteins, and vitamin C in harvested ‘ice cream’ radish fleshy roots irrigated by salt water. All these quality indicators have shown an increase while the salinity level is low and then a significant decrease while the salinity level in irrigation water increases further. With more soluble sugars and proteins in fleshy root cells, the crispy, watery, and tasty ‘ice cream’ fruit radish roots should have high quality and more market values for their favorable consumers.

Through this study, we have demonstrated that ‘ice cream’ fruit radish can be grown well in peat substrate with mildly salinized water with a salinity level of less than 4.2 dS/m. Even with a slight reduction in foliage growth, the yield and quality of radish roots are not affected. With an increase in salinity in irrigation water up to a tolerable level, the root quality actually increases in its flavor, Vc level, and soluble protein content. Although the greenhouse experiment is designed to simulate the salt soil scenario in the field with peat as substrate, the results may have shed some light on how fruit radish plants may grow in the real saline soil. Nevertheless, large-scale trials in a field setting in saline lands around coastal China are needed and are of paramount importance to further optimize conditions and practices for growing fruit radishes by dripping mildly salty water into areas where fresh water is absent.

5. Conclusions

In this study, with a saltwater irrigation experiment at different salinity levels, the leaf growth of ‘ice cream’ fruit radish is inhibited by even mild salt stress, while root growth and quality of the radish are not affected but enhanced by a low salinity level in irrigation water. The highest value of the weight of the fleshy root, the root/shoot ratio, the soluble solid content in both root peel and pulp, and the soluble sugar content in peels of the root was measured in the 4.2 dS/m treatment with an increase of 51.10%, 117.75%, 11.06%, 6.70%, and 55.85% compared to those in CK (1.0 dS/m), respectively. Moreover,

although the highest soluble protein content and vitamin C content were not measured in the 4.2 dS/m treatment, they increased 11.99% and 113.36%, respectively. In conclusion, we recommend that salt water containing 4.2 dS/m salt be used to irrigate fruit radishes for optimized yield and quality of fleshy roots. This experiment has helped understand the mechanism involved in regulating biomass allocations and physiological responses of fruit radish under salt stress in terms of its growth and quality, but its biochemical responses at the cellular level to salt stress need to be further explored for a better understanding of radish salt tolerance.

Author Contributions: The research presented here was carried out in collaboration between all authors. H.Z. and M.L. carried out the data analysis and prepared the first draft of the manuscript; M.L. and H.X. carried out the experiment; D.F. conceived the idea and designed this study; D.F. and X.S. edited, finalized, and revised the manuscript. All authors have read and agreed to the published version of the manuscript.

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Data Availability Statement: All data will be made available on request to the corresponding author's email with appropriate justification.

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

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