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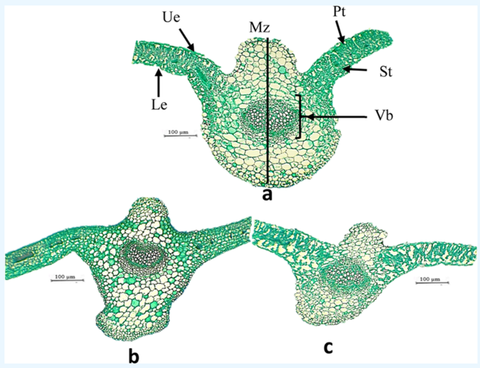
Article

Effect of Salt Stress on Botanical Characteristics of Some Table Beet (*Beta vulgaris* L.) Cultivars

Ayah T. Zaidalkilani, Aman H. Al-Kaby, Amira M. El-Emshaty,* Sadeq K. Alhag, Laila A. Al-Shuraym, Zakaria A. Salih, Amro Ahmed Taha, Ammar M. Al-Farga, Ashmawi E. Ashmawi, Saleh A. Hamad, Hany S. Abd El-Raouf, Shahinaz E. Ahmed, Ahmed M. El-Taher, Moses V. M. Chamba,* and Taghreed A. Badawi

Cite This: *ACS Omega* 2024, 9, 47788−47801



ABSTRACT: Salinity inhibits the uptake of nitrogen, which slows down the growth and prevents plant reproduction. Certain ions, especially chloride, are poisonous to plants; when their concentration increases, the plant becomes poisoned and eventually perishes. The adaptability of several table beet cultivars (*Beta vulgaris* L.) to saline water irrigation creates new opportunities for extending beet production, increases the added economic value, and has a positive environmental impact. A pot experiment is carried out for two successive seasons, 2019/2020 and 2020/2021, to investigate the effect of irrigation with agriculture saline drainage water on the growth and biochemical traits of three selected cultivars (Detroit Dark Red, Red Ball, and Red Ace). Four levels of salinity are applied (1000, 2000, 3000, and 4000 ppm) along with tap water of 260 ppm salinity, which serves as the control. Detroit Dark Red beets show the best results among the other cultivars under consideration. Irrigation with the first level of saline water (1000 ppm) at both seasons of cultivation results in a significant increase rate in growth parameters (13−23%). The second level of salinity (2000 ppm) shows the maximum increase rate of some chemical constituents, such as ascorbic acid (16.26%), nitrogen (58.21%), phosphorus (11.94%), potassium (34.66%), and sodium (85.14%). The levels of total soluble solids (TSS), anthocyanins, proline, total sugars, water saturation deficit, and sodium increase significantly in proportion to saline water concentrations. The selected table beet mature leaves show slight variations in anatomical structure, especially in the *B. vulgaris* L. cv. Detroit Dark Red under the highest salinity concentration (4000 ppm) was less than that of the control and the other two cultivars. Other cultivars may be the subject in the near future to study the effect of their salinity tolerance with the aim of increasing productivity, enhancing their characteristics, and preserving the environment.

■ INTRODUCTION

Table beet (*Beta vulgaris* L.) is a modern domesticated crop that belongs to the Caryophyllales order.[1] Following sugar cane, sugar beet is the most important global source of sugar for human consumption and beet pulp for animal feed each year.[2] It is a significant root crop, as its taproots are employed in the sugar production process. In many countries, the table beet is known as the garden beet, red beet, or beetroot. The table beet is a cold-loving plant, and its roots are the only portion of the plant that may be eaten raw in salads. In Egypt, the volume of drainage irrigated water may exceed 13.5 billion m^3/year, which flows unused to the Mediterranean Sea and the coastal lakes.[3] To alleviate water scarcity in the agricultural sector, some of this water should be utilized for irrigation; however, when water is alternated or coupled with good quality water supplies, saline water can be used for irrigation.[4] Cultivating sugar beet has recently proven to be promising not just in high fertility soils but also in low soils or soils with biological or abiotic difficulties, such as salty, alkaline, and calcareous soils.[5] Furthermore, it may be grown affordably in newly recovered soils.[5] Sugar beet crop is often the most important cash crop in the rotation and is able to prepare the soil for the next crop.[6] Due to osmotic and ionic stress, salt stress has a negative impact on plant development and output.[7] By disrupting cytoplasmic ionic concentration and osmotic gradients, salinity can drastically affect many plant morph physiological functions and inhibit several vital metabolic and physiological functions such as photosynthetic activity, protein synthesis, enzymatic activity, and nutritional balance by

Received: September 4, 2024
Revised: October 30, 2024
Accepted: November 6, 2024
Published: November 19, 2024

https://doi.org/10.1021/acsomega.4c08161
ACS Omega 2024, 9, 47788−47801

Table 1. Chemical Properties of Different Saline Water Irrigation Samples

concentrations (ppm)	EC	CO_3^-	HCO_3^-	Cl^-	SO_4^-	Ca^{++}	Mg^{++}	Na^+	K^+
260	0.41		3.1	1.4	0.1	1.8	1.4	1.1	0.3
1000	1.52		3.1	9.8	0.2	2.2	3.4	7.1	0.4
2000	3.12		3.1	22.5	0.4	2.5	6.2	16.8	0.5
3000	4.75		3.1	32.5	0.7	3.3	9.1	24.2	0.7
4000	6.38		3.1	45.7	1.2	3.9	14.1	31.1	0.9

Table 2. Plant Growth Conditions

	air temperature (°C)		light intensity (lux)		average relative humidity (%)	
month	2019/2020 season	2020−2021 season	2019/2020 season	2020−2021 season	2019/2020 season	2020−2021 season
October	28.90	27.72	70,375	70,271	39.54	40.84
November	25.20	25.12	60,273	60,122	35.16	34.53
December	22.30	19.53	54,932	52,867	34.11	33.61
January	18.55	15.82	52,897	51,958	33.17	32.18
February	22.40	23.34	60,891	59,786	31.21	32.11
March	25.00	24.92	62,176	63,462	34.06	33.39

reducing the absorption of essential elements such as N, P, K, and Ca.[8] Higher plant growth rates and net photosynthesis are slowed by salinity.[9] Previous research showed that irrigating table beet with saline water increased the number of leaves, leaf length, fresh weight of leaves, fresh weight of root, root diameter, leaf dry matter, root dry matter, total chlorophyll, carotenoids, and potassium (up to 1000 ppm), after which higher levels decreased these attributes.[10] TSS, sugars, anthocyanins, salt, and proline all increased dramatically with each increase in saline levels up to 4000 ppm.[10] Growing sugar beets on salinized farmland may be an effective technique for increasing sugar production while maximizing the use of salinized farmland.[11]

The impact of salinity on *Beta vulgaris* seed germination, early seedling growth, and anatomical structure appeared in their capability of mitigating the negative effects of salt stress.[12] Aali et al. discussed the effect of salinity on the anatomical structure.[13] Anatomically, increasing salinity levels decreased the thickness of the midrib region of the leaf blade, mesophyll tissue, the upper and lower epidermis, and the large motor cell besides decreasing the tangential dimension of the midrib vascular bundle, and the tangential dimension of the big xylem vessel as well.[14] The thickness of the leaf epidermal layers was thinner in salt-stressed plants than in control plants. The salinity effect was a concentration-dependent factor.[15] This decrease in the epidermal thickness could be attributed to the limited cell division and growth that occurred at higher salinity. Increased salt concentration causes a decrease in mesophyll thickness.[16−18] The considerable reduction in palisade tissue at high salinity (750 mM NaCl) could be an adaptation of this halophyte to reduce photosynthetic energy usage in higher saline environments.[19] On the other hand, anthocyanins are a group of natural pigments that belong to the flavonoid family and are thought to be responsible for the color and flavor of many fruits and vegetables.[9] Their potential and efficiency as antioxidants are almost twice the antioxidant capacity of other known antioxidants like (±) catechin and vitamin E, and synthetic antioxidants like BHA (butylated hydroxyl anisole) and BHT (butylated hydroxyl toluene), which is commonly utilized, in food technology.[20−23] Overall, salinity is considered one of the factors that has the greatest impact on crop production, limiting physiological activities and the productive potential of cultivated plants, where salinity stress has a number of negative consequences on plant growth and development. Many countries, particularly Egypt, face significant issues in terms of salinity tolerance.[24] As a result, innovative solutions are required to boost plant tolerance in salinity stress tactics and their ability to flourish in salinized environments. The aim of this study is to evaluate the influence of saline water irrigation at various concentrations on the growth metrics, plant chemical content, and chemical constituents measurements of three selected table beet cultivars (*Beta vulgaris* L.).

■ MATERIALS AND METHODS

Experiment Design. This research project was carried out at Zawiat Riziyn village (Private Farm), Menouf City, El-Menoufiya Governorate, Egypt, during the two successive winter seasons of 2019/2020 and 2020/2021. Saline water (salinity = 26,000 ppm) was obtained from Karoun Lake, El-Fayoum City, Egypt. Dilution with tap water (260 ppm) to 1000, 2000, 3000, and 4000 ppm was performed to prepare four levels of saline water concentrations along with tap water as an experimental control. The chemical analyses of the diluted saline drainage water are presented in Table 1.

Three table-beet cultivars (Detroit Dark Red, Red Ball, and Red Ace) were used in this study. The pots used in the experiments were 25 cm in diameter and 25 cm in length. Each pot contained 15 kg of clay loam soil. Seeds of table beets were sown on October 24th and 27th for the first and second seasons; 10 seeds/pot at equal distance and depth for 3 weeks, and then placed in clay soil. The pots were arranged in four replicates per treatment. Irrigation with saline water was started 10 days after planting twice a week at field capacity (FC). The field capacity was evaluated by applying −1/3 atm suction to a saturated soil sample using a pressure plate. When the water stopped leaving the soil sample, the soil moisture was measured gravimetrically and compared to the field capacity. Each pot was fertilized with ammonium sulfate (3.75 g), calcium superphosphate (3.5 g), and potassium sulfate (0.80 g). The fertilizers were divided into two equal parts. The first was added immediately after thinning, whereas the second was added 3 weeks later. Table 2 shows the average monthly air temperature, light intensity, and relative humidity at the experimental location during the two seasons of 2019−2020 and 2020−2021.

Soil Analysis. Soil acidity (pH) was measured in the soil paste by using a Gallen Kamp pH meter (A. Gallen Kamp Co. Ltd., UK), and the electric conductivity (EC) in a 1:2.5 soil/

https://doi.org/10.1021/acsomega.4c08161
ACS Omega 2024, 9, 47788−47801

Table 3. Physical and Chemical Properties of Experimental Soil[a]

physical properties							
soil type	fine sand (%)	coarse sand (%)	silt (%)	clay (%)	wilting point (% v/v)	SP (%)	hydraulic conductivity (cm h^{-1})
sandy clay loam	44.37	20.23	11.13	24.27	30.20	34.17	2.33
available water (% v/v)		HW (%)		bulk density (mg m^{-3})			total porosity (%)
7.99		6.77		1.65			55.56

chemical properties								
pH in suspension 1:2.5	organic matter (OM, %)	available nutrients (ppm)						
7.75	0.59	N	P	K	Fe	Mn	Zn	Cu
		25	17	76	13	12	1.3	0.76
soluble cations (mequiv/L)						soluble anions (mequiv/L)		
Ca^{++}	Mg^{++}	Na^{+}	K^{+}	CO_3^{-}	HCO_3^{-}	Cl^{-}		SO_4^{-}
0.64	0.46	0.97	0.27		0.58	1.25		0.51
SAR		ESP			$CaCO_3$ (%)			EC (ds/m)
1.87		3.44			3.26			0.86

[a]pH in suspension 1:2.5. EC (ds/m), soluble cations and anions (meq/L): in the previous saturated extract. EC: electric conductivity; HW: hygroscopic water; HC: hydraulic conductivity. Thus, the purpose of this research was to examine the effects of different levels of saline-irrigated water on the vegetative growth and chemical composition of some table beet varieties.

Table 4. Effect of Irrigation at Various Levels of Saline Water on the Leaf Number and Fresh and Dry Weight of Leaves of Some Table Beet Varieties in the Two Seasons[a]

		leaf number		fresh weight of leaves (g)		leaf dry weight (g/100 g FW)	
		seasons					
treatments		1st	2nd	1st	2nd	1st	2nd
A: Salinity Levels (ppm)							
control (260)		10.26C	10.14C	85.21C	87.81B	9.95C	9.55B
1000		12.97A	12.85A	95.9A	12.97A	13.20A	11.09A
2000		11.22B	11.15B	90.22B	11.22B	10.68B	9.87B
3000		9.24D	9.22D	81.38D	84.75C	8.91D	8.94C
4000		8.12E	7.80E	74.27E	76.26D	8.12E	7.92D
LSD at 5%		0.34	0.38	1.22	1.37	0.38	0.34
B: Varieties							
Detroit Dark Red		9.88C	10.46A	93.92A	95.98A	11.09A	10.11A
Red Ball		10.90A	10.61A	85.32B	89.2B	9.88B	9.36B
Red Ace		10.31B	9.44B	76.96C	75.08C	9.54C	8.94C
LSD at 5%		0.26	0.29	0.95	1.06	0.29	0.27
C: Interaction							
control (260)	Detroit Dark Red	9.93f	10.17fg	91.97d	98.05b	10.92d	10.15c
	Red Ball	10.07ef	10.03fg	86.30f	89.47d	9.69f	9.37def
	Red Ace	10.77d	10.23ef	77.35h	75.91h	9.25fg	9.13ef
1000	Detroit Dark Red	12.27bc	12.93b	107.56a	113.26a	13.75a	12.02a
	Red Ball	13.83a	13.60a	95.20c	99.34b	13.02b	11.06b
	Red Ace	12.83b	12.03bc	85.01f	82.29f	12.83b	10.18c
2000	Detroit Dark Red	10.63de	11.33dc	100.58b	95.57c	11.63c	10.37c
	Red Ball	11.83c	11.70 cd	89.45e	91.64d	10.56d	9.78 cd
	Red Ace	11.20d	10.43ef	80.63g	78.96g	9.87ef	9.45def
3000	Detroit Dark Red	8.60g	9.63fg	89.72e	96.94c	10.43de	9.54de
	Red Ball	10.10ef	9.70fg	81.12g	84.95e	8.24hi	8.91fg
	Red Ace	9.03g	8.33hi	73.31h	72.35i	8.07i	8.37g
4000	Detroit Dark Red	8.60g	9.63fg	89.72e	96.94c	8.74gh	8.48g
	Red Ball	10.10ef	9.70fg	81.12g	84.95e	7.90i	7.69h
	Red Ace	9.03g	8.33hi	73.31h	72.35i	7.71i	7.58h
LSD at 5%		0.59	0.65	2.12	2.38	0.65	0.59

[a]Mean values with different letters in a column are statistically different according to DMRT ($p < 0.05$). LSD: Least significant difference. The 260, 1000, 2000, 3000 and 4000 are four levels of salinity that were applied. Detroit Dark Red, Red Ball and Red Ace are some table beet (*Beta vulgaris L.*) cultivars.

water extract was determined using the described procedures.[25] The mechanical analysis of the soil was performed using the international pipette method using NaOH as a depressing agent.[26] To extract accessible nitrogen, the Devarda alloy method of steam distillation was used with a 1% potassium sulfate solution.[27,28] The amount of available phosphorus

https://doi.org/10.1021/acsomega.4c08161
ACS Omega 2024, 9, 47788−47801

Table 5. Effect of Irrigation at Various Levels of Saline Water on Fresh and Dry Weight of Root, and Root Diameter of Some Table Beet Varieties in Both Seasons[a]

treatments		fresh root weight (g)		root diameter (cm)		dry root weight (g/100 g FW)	
		seasons					
		1st	2nd	1st	2nd	1st	2nd
A: Salinity Levels (ppm)							
control (260)		100.79	107.24B	5.35B	5.20B	10.01C	11.01C
1000		119.84A	126.27A	6.07A	5.71A	12.21A	13.48A
2000		87.90C	99.17C	5.16B	5.33AB	10.87B	12.11B
3000		78.13D	84.35D	4.27C	4.40C	8.89D	9.94D
4000		66.11E	69.95E	3.55D	3.68D	7.54E	8.39E
LSD at 5%		3.31	2.03	0.24	0.44	0.55	0.39
B: Varieties							
Detroit Dark Red		106.32A	108.14A	5.35A	5.17A	10.87A	11.92A
Red Ball		86.35B	97.87B	5.09B	4.71B	9.77B	10.98B
Red Ace		79.09C	86.18C	4.20C	4.72B	9.07C	10.06C
LSD at 5%		1.79	1.57	0.19	0.34	0.43	0.30
C: Interaction							
control (260)	Detroit Dark Red	117.74b	126.44b	5.80b	5.53ab	11.04cd	11.99c
	Red Ball	95.68d	102.17d	5.63bc	5.10bcd	9.93ef	11.04d
	Red Ace	88.96e	93.10e	4.63fg	4.97bcd	9.06fg	9.99e
1000	Detroit Dark Red	139.10a	140.66a	6.63a	6.20a	13.43a	14.74a
	Red Ball	119.25b	127.50b	6.40a	5.57ab	12.19b	13.30b
	Red Ace	101.17c	110.66c	5.17de	5.37bc	11.01cd	12.41c
2000	Detroit Dark Red	101.11c	104.34d	5.57bcd	5.37bc	11.91bc	13.15b
	Red Ball	81.06f	100.83d	5.33 cd	4.93bcd	10.86de	12.05c
	Red Ace	81.53f	92.34e	6.40a	5.70ab	9.84f	11.13d
3000	Detroit Dark Red	93.18d	93.01e	4.83ef	4.70cde	9.90ef	10.58de
	Red Ball	71.62g	86.14f	4.37g	4.40de	8.46gh	10.07e
	Red Ace	69.59g	73.90gh	3.6h	4.10ef	8.31ghi	9.18f
4000	Detroit Dark Red	80.45f	76.26g	3.93h	4.03ef	8.08hij	9.14f
	Red Ball	64.12h	72.69h	3.73h	3.53f	7.42ij	8.45g
	Red Ace	53.77i	60.91i	3.00i	3.47f	7.11j	7.60h
LSD at 5%		4.02	3.52	0.42	0.78	0.69	0.67

[a]Mean values with different letters in the column are statistically different according to DMRT ($p < 0.05$). LSD: Least significant difference. The 260, 1000, 2000, 3000 and 4000 are four levels of salinity that were applied. Detroit Dark Red, Red Ball and Red Ace are some table beet (*Beta vulgaris L.*) cultivars.

(extracted using a 500 mM $NaHCO_3$ solution with a pH of 8.5) was measured using a Beckman Du 7400 spectrophotometer (GMI Co., Ramsey, MN) at a wavelength of 650 nm.[29] The available potassium was measured with a Corning flame photometer.[30] Ammonium acetate solution (1, pH = 7.0) was used as a starting point. The Walkley and Black chromic acid wet oxidation method was used to determine the amount of organic materials.[31] To extract accessible micronutrients from the soil samples, diethylene thiamine penta acetic acid (DTPA) solution was used.[32] The measurements were performed by using an atomic absorption spectrophotometer. According to the procedure outlined, the saturation percentage (SP%) was calculated according to the reported method,[13] and the hydraulic conductivity (K) values of the soil sample columns were determined using Smith's standard method.[33]

Plant Analysis. Determination of Total Chlorophyll and Carotenoids. Pigments were extracted from 0.5 g of fresh and young leaves in dimethyl formamide (DMF) overnight at 4 °C to estimate the mass of chlorophyll a, chlorophyll b, total chlorophyll, and carotenoids per leaf. The amount of pigments was determined by a Beckman Du 7400 spectrophotometer at wavelengths of 663, 470, and 647 nm, using Moran's equation.[34]

Determination of Plant Growth Parameters and Chemical Contents. The number of leaves was counted per plant. A kilogram balance was used to determine the weight of the fresh leaves and roots per plant. 100 g of fresh weight was dried at 70 °C in a laboratory oven until a constant weight was reached, and the dried weight of the leaves and roots was calculated as g/100 g fresh weight. The plant root diameter was measured in cm (using a Vernier caliper). Sulfuric and perchloric acid mixtures were used to wet digest plant samples.[35] The Kjeldahl method was used to determine the plant nutrients in an aliquot of nitrogen.[36] For phosphorus determination, a stannous chloride-reduced molybdo-phosphoric blue color technique was used,[36] while flame photometer was used for the determination of potassium and sodium.[36] Abbe refractometers were used to calculate the percentages of total soluble solids (TSS%).[37] Ascorbic acid was measured as mg/100 g of fresh weight using the 2,6-dichlorophenol indophenol dye technique.[38] Total sugars were measured in grams per 100 g of dry weight, according to DuBois.[39] The conventional approach of Cherifi et al. was used to determine the proline content of salt-stressed and control plants,[40] where − with minimal adjustments − proline was isolated from leaf samples (20 mg FW) according to the previously reported method.[41] The proline concentration was determined as moles g DW^{-1}. The water saturation deficit (WSD) was estimated as a percentage value.[42] Anthocyanins were measured using the reported method.[43,44]

https://doi.org/10.1021/acsomega.4c08161
ACS Omega 2024, 9, 47788−47801

Anatomical Studies. The tested material, including leaf lamina, was collected throughout the second growing season of 2020/2021 at the age of days from the sowing date. Anatomical characteristics of table beet leaves: upper epidermal layer thickness (μm), lower epidermal layer thickness (μm), palisade tissue thickness (μm), spongy tissue thickness (μm), midrib zone thickness (μm), length of the vascular bundle (μm), and width of the vascular bundle (μm). (each value represents 5 sections with 5 readings per section). The execution of the microtechnique was carried out according to the previously described method.[14]

Statistical Analysis. Appropriate analysis of variance was performed using COSTATE V 6.4(2005) for Windows and is depicted in Table 3. The least significant difference test at the 0.05 level of probability was used to compare the differences among the means of the various treatment combinations, as illustrated by the computer software program based on significant differences among the means of various treatments as determined by the least significant difference test.[45,46]

RESULTS

Plant Growth Parameters (Leaf Number and Fresh and Dry Weight of Leaves). The effect of different salinity levels of saline water irrigation (1000, 2000, 3000, and 4000 ppm) on vegetative growth criteria (fresh and dry weight of leaves and roots), leaf number, and root diameter are presented in Tables 4 and 5 of different table beet cultivars (Detroit Dark Red, Red Ball, and Red Ace). The results in Table 4 indicate that the fresh weight and leaf number of table beet cultivars irrigated with 1000 ppm saline increased gradually to 4000 ppm in both seasons. The Detroit Dark Red cultivar provided the highest significant increase ($p > 0.05$) of fresh weight, whereas the Red Ball cultivar provided the highest values of leaf number compared to the other cultivars. The interaction between salinity levels and plant varieties revealed that the Red Ball cultivar showed the highest increase in the fresh leaf weight (g) at a salinity level of 1000 ppm. In both seasons, the lowest values were obtained with a saline water level of 4000 ppm and Red Ace. The data indicated that there was a significant effect ($p > 0.05$) for the two seasons on leaf number by the individual addition or interaction between irrigation with saline water and different studied varieties. At the first level of salinity water irrigation (1000 ppm), the Red Ball cultivar parameters were significantly increased in both seasons by 31.08 and 35.59%, respectively, relative to the control.

Fresh and Dry Weight of Roots and Root Diameter. The results in Table 4 indicate that the highest values of leaf dry weight (g) were observed in plants treated with saline water irrigation at 1000 ppm (i.e., Detroit Dark Red cultivar) with a significant increase (25.92−18.42%) compared to the control in both seasons, followed by Red Ball and Red Ace cultivars at the same level of saline water irrigation. However, the lowest decrease (16.64−16.97%) compared to the control was recorded with 4000 ppm saline water irrigation level with the Red Ace cultivar in both seasons. The findings in Table 4 show that the highest values of fresh and dry weight of roots (g) were recorded in both seasons when plants were treated with 1000 ppm saline water irrigation with the cultivar (Detroit Dark Red), which increased (18.14−21.64%) compared to control in the first and in the second seasons (11.24−22.93%), followed by the same level of irrigation saline water with the Red Ball cultivar. The root diameter (cm) exhibited the highest increase with saline water irrigation at 1000 ppm added to Detroit Dark Red (14.31%) and (12.12%) in the first and second seasons, respectively. Also, the same increase was recorded at 2000 ppm in Red Ace in the first and second seasons, followed by 1000 ppm saline water with the Red Ball cultivar.

Chemical Constituents and Quality Parameters. *Total Soluble Solids, Ascorbic Acid, and Total Sugars.* Regarding the interaction effect, table beet plants treated with a combination of irrigation with saline water had the highest TSS and ascorbic acid values (Figures 1a−c and 2a−c). As depicted in Figure 1a−c, TSS had the highest increase in the first season for the Detroit Dark Red cultivar irrigated with 4000 ppm saline water (21.30%) compared to the control in the first season, followed by irrigation with 3000 ppm saline water. During the second season, the highest values were recorded with 3000 ppm of saline water irrigation to the Detroit Dark Red cultivar, as well as 4000 ppm of saline water was added to each of the two varieties (Detroit Dark Red and Red Ball).

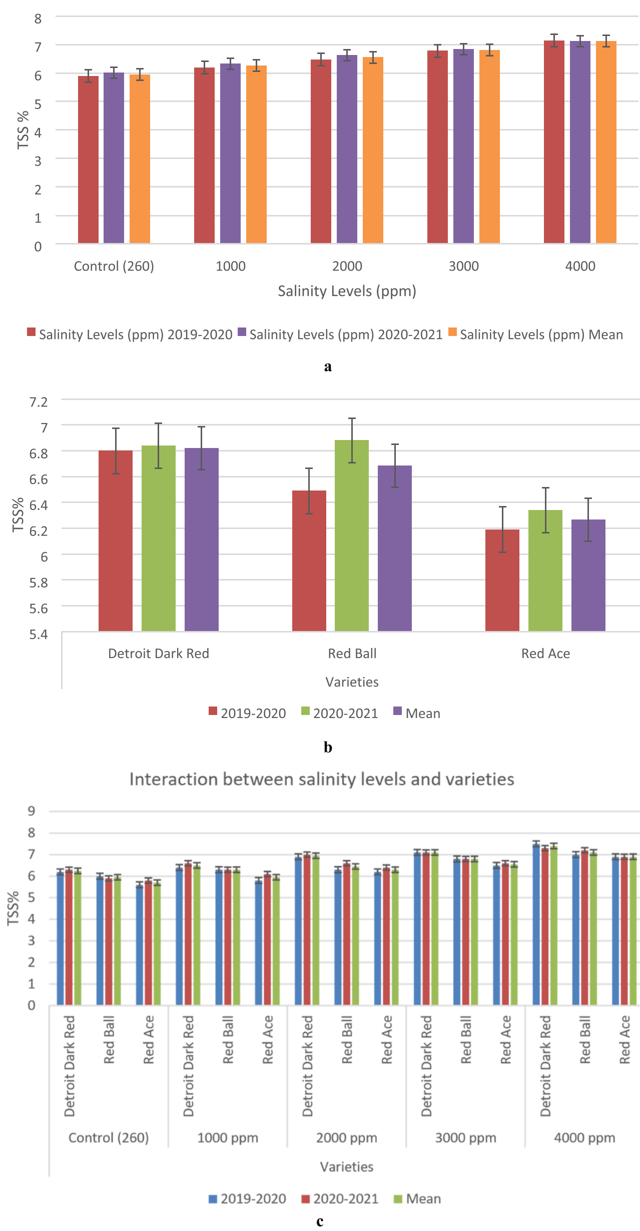


Figure 1. (a−c) Relationship between salinity levels and table beet varieties on total soluble solids (TSS %) in table beet plants.

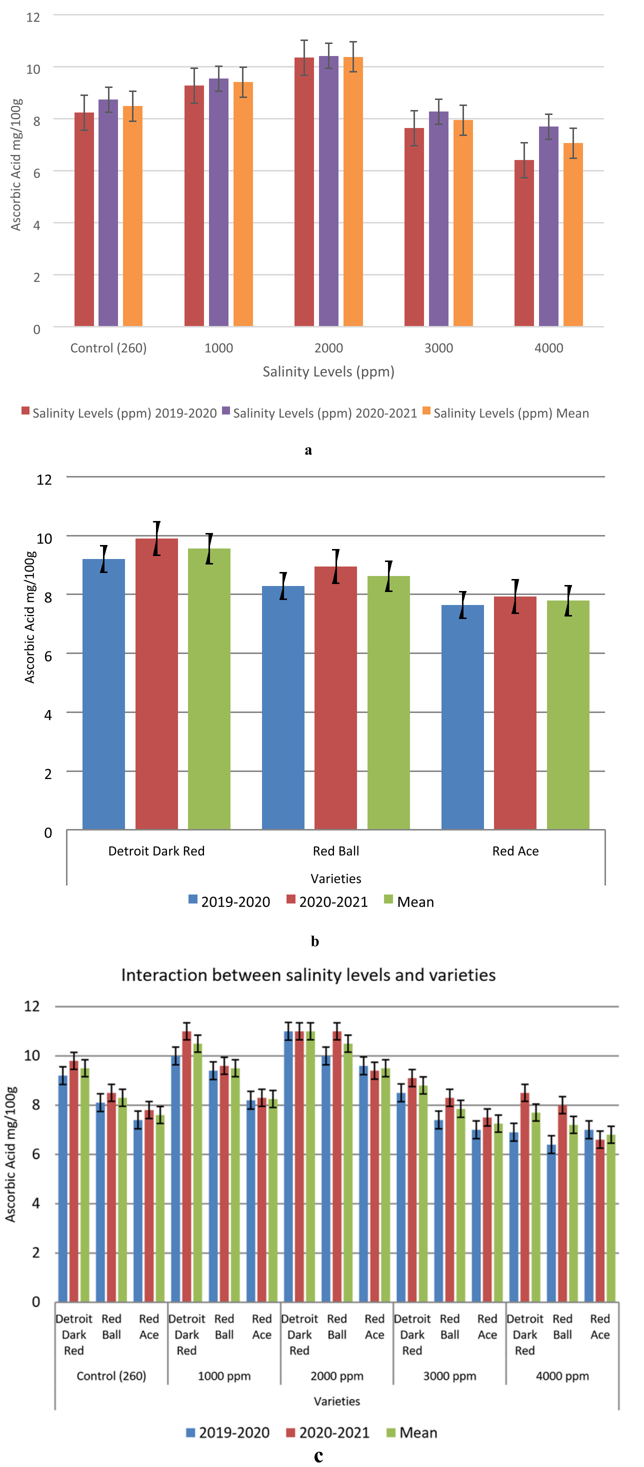


Figure 2. (a−c) Relationship between salinity levels and table beet varieties on ascorbic acid mg/100 g in table beet plants.

Figure 2a−c reflects the effect of saline-irrigated water and table beet varieties on ascorbic acid content at harvest time. In the first season, the combined treatment of 2000 ppm added to Detroit Dark Red yielded the highest significant increase, followed by the same water irrigation water concentration with Red Ball, whereas, in the second season, the highest increase (15.89%) compared to the control was presented with 2000 ppm saline water added to Detroit Dark Red in the first season and the same rate with 2000 ppm with Red Ball and 1000 ppm applied to Detroit Dark Red.

The data presented in Figure 3a−c demonstrate the effect of four different levels of saline-irrigated water (1000, 2000, 3000, and 4000 ppm) added to the three varieties of table beets and evaluated their effect on total sugars (g/100 g DW). In both seasons, total sugars increased significantly when 4000 ppm saline water was added to Detroit Dark Red, followed by 3000 ppm for the same cultivar.

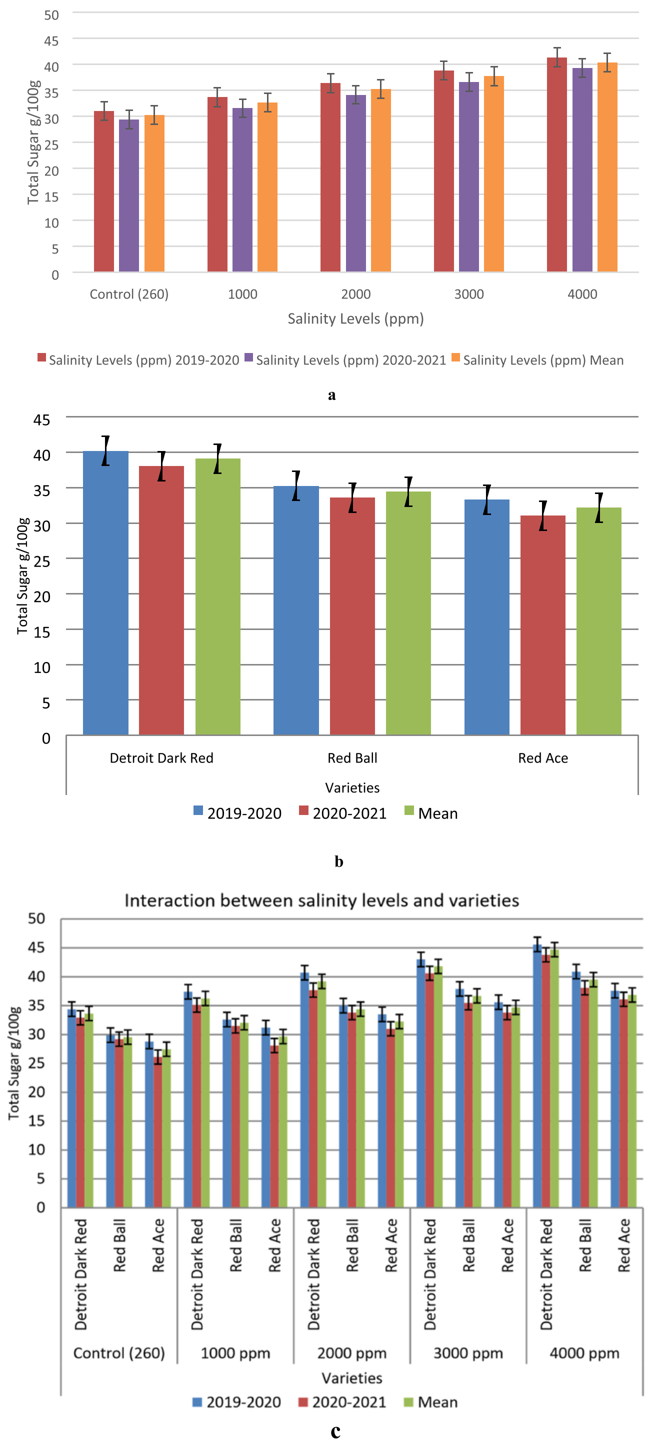


Figure 3. (a−c) Relationship between salinity levels and table beet varieties on total sugar g/100 g in table beet plants.

Total Chlorophyll, Total Carotenoids, and Anthocyanin. Chlorophyll is the most important pigment in photosynthesis, as

https://doi.org/10.1021/acsomega.4c08161
ACS Omega 2024, 9, 47788−47801

it absorbs and transforms light energy. As a result, chlorophyll concentration is an important physiological indicator of salt stress damage in plants. In terms of plant quality, the table beet plants irrigated with saline water exhibited significant increases in total chlorophyll and carotenoids ($p > 0.05$). Figures 4a−c and 5a−c show the effect of saline-irrigated water alone or in combination with different varieties of table beet (Detroit Dark Red, Red Ball, and Red Ace) on some quality parameters of table beet photosynthetic pigments (total chlorophyll mg 100 g FW, and carotenoids mg/100 g FW). In terms of quality parameters, the data showed a high positive increase relative to the control

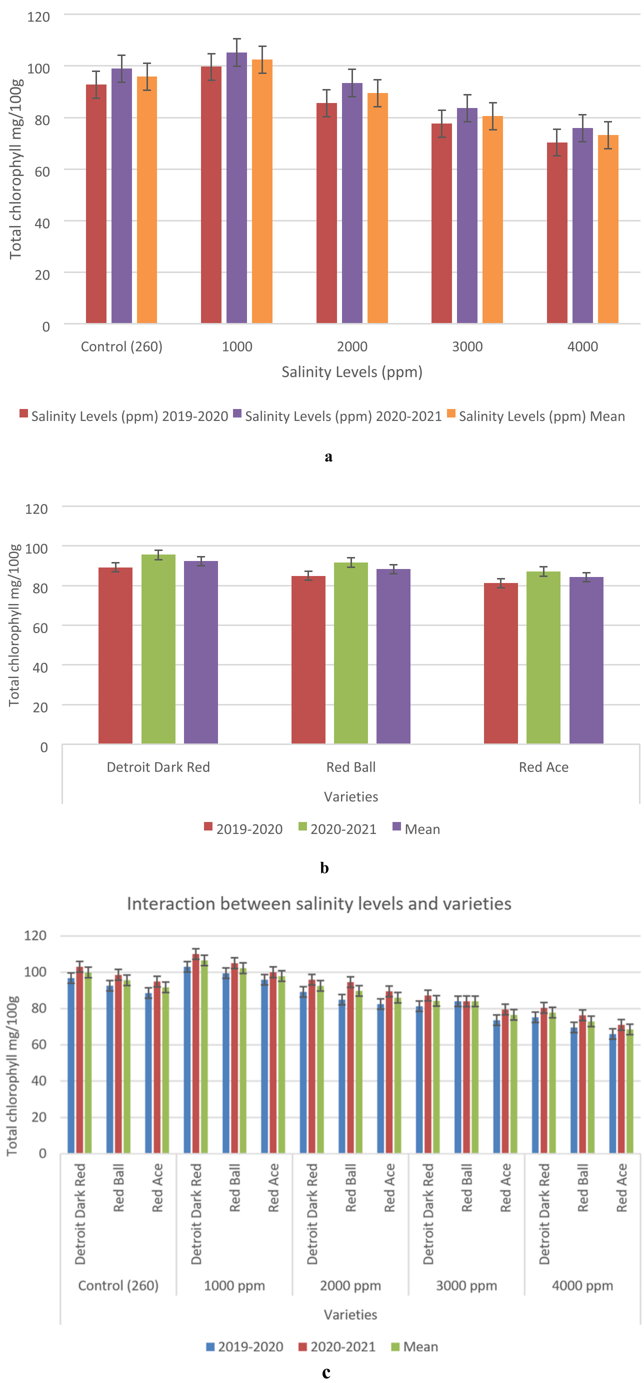


Figure 4. (a−c) Relationship between salinity levels and table beet varieties on total chlorophyll mg/100 g in table beet plants.

Figure 5. (a−c) Relationship between salinity levels and table beet varieties on total carotenoids mg/100 g in table beet plants.

(6.65−6.99%) of total chlorophyll irrigated by saline water at 1000 ppm level by adding to the Detroit Dark Red cultivar in both seasons, followed by 1000 ppm mixed with Red Balls in both seasons.

The highest increase in both seasons was observed at the 1000 ppm level of irrigation with Detroit Dark Red, as shown in Figure 6a−c for the effect of saline water with table beet verities on carotenoids. Figure 6a−c shows the highest increase of anthocyanine (mg/100 g. FW) values recorded with the addition of 4000 ppm saline water to the Detroit Dark Red cultivar, followed by the same concentration of irrigated water with Red Ball compared to the control, and the lowest value was recorded with 1000 ppm added to Red Ace.

Proline Content and Water Saturation Deficient. It is well known that during salt stress, compatible solutes such as proline accumulate in many plants, acting as compatible solutes, osmoprotectants, and positive agents for enzymes and cellular organelles. Under saline water conditions, all species showed an increase in proline content in the current study. Significant

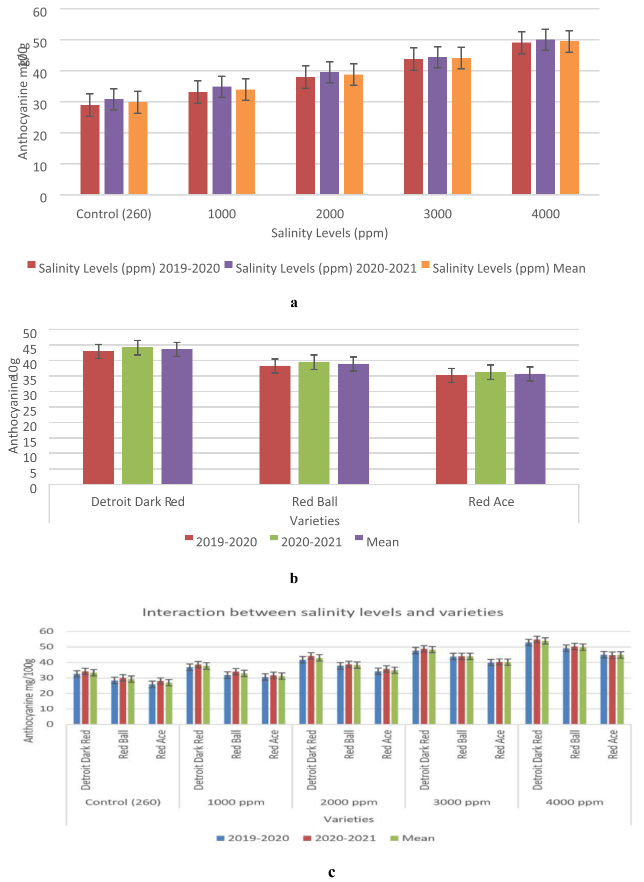


Figure 6. (a−c) Relationship between salinity levels and table beet varieties on anthocyanine mg/100 g in table beet plants.

differences in the proline content were observed. As shown in Table 6, the highest increase in proline content was measured at 4000 ppm with Detroit Dark Red (50.00−66.41%) as compared to control in both seasons, followed by the same concentration of saline water with Red Ball cultivar. Water saturation deficiency was also recorded, which showed the greatest increase at 4000 ppm salinity level applied to Red Ball, while the highest drop was at 1000 ppm added to Red Ace. The decrement ranged between 21.50% in the first season and 6.40% in the second season compared to the control.

Plant Chemical Contents. *Nitrogen, Phosphorus, Potassium, and Sodium Content in Plant.* The effect of saline water irrigation levels on nutritional element concentrations in several varieties of table beats, such as nitrogen (N), phosphorus (P), potassium (K), and sodium (Na), is shown in Table 7. The highest values of N and P contents were detected in the Detroit Dark Red variety irrigated with 2000 ppm saline water. However, the highest decrease of the aforementioned elements was observed for the irrigation of Detroit Dark Red with 4000 ppm saline water. The decrease ranged between 23.68% in the first season and 29.56% in the second season compared to the control. Potassium content decreased by increasing the water salinity concentration up to 2000 ppm and then decreased at salinity levels of 3000 and 4000 ppm in different varieties. The highest increase in K concentration ranging from 30.03 to 39.27%, compared to the control in table beet plants, was recorded at a concentration of 2000 ppm with the Detroit Dark Red variety, followed by a concentration of 1000 ppm with the same variety. However, increasing the concentration of salt-

Table 6. Effect of Irrigation at Various Levels of Saline Water on Proline and Water Saturation Deficit of Some Table Beet Varieties in 2019−2020 and 2020−2021 Seasons[a]

treatments		proline (mg/1 g DW) 1st season	proline (mg/1 g DW) 2nd season	water saturation deficit 1st season	water saturation deficit 2nd season
A: Salinity Levels (ppm)					
control (260)		17.12E	15.13E	31.40E	32.69E
1000		22.66D	20.88D	35.73D	35.12D
2000		29.36C	26.49C	38.57C	37.94C
3000		36.09B	34.15B	42.22B	41.61B
4000		43.51A	41.39A	46.08A	44.84A
LSD at 5%		0.57	0.66	1.50	0.73
B: Varieties					
Detroit Dark Red		31.83A	29.97A	37.97B	37.98B
Red Ball		29.83B	27.49B	41.06A	40.00A
Red Ace		27.58C	25.37C	37.36B	37.34B
LSD at 5%		0.45	0.51	1.16	0.57
C: Interaction					
control (260)	Detroit Dark Red	18.33m	16.52L	30.59h	31.54i
	Red Ball	17.15n	14.80m	35.52fg	34.96g
	Red Ace	15.88o	14.06m	28.09h	31.56i
1000	Detroit Dark Red	24.74j	23.05i	34.97g	34.89g
	Red Ball	23.01k	20.43j	38.09ef	36.90f
	Red Ace	20.23l	19.16k	34.13g	33.58h
2000	Detroit Dark Red	32.29g	29.33g	37.86ef	37.77f
	Red Ball	29.10h	26.32h	40.13de	39.31e
	Red Ace	26.69i	23.84i	37.71ef	36.75f
3000	Detroit Dark Red	37.95d	36.38d	41.14d	40.88d
	Red Ball	36.59e	34.72e	43.99bc	42.85c
	Red Ace	33.75f	31.36f	41.53 cd	41.10d
4000	Detroit Dark Red	45.82a	44.55a	45.31ab	44.81ab
	Red Ball	43.32b	41.20b	47.58a	45.98a
	Red Ace	41.38c	38.42c	45.34ab	43.72bc
LSD at 5%		1.00	1.14	2.59	1.27

[a]Mean values with different letters in the column are statistically different according to DMRT ($p < 0.05$). LSD: Least significant difference. The 260, 1000, 2000, 3000 and 4000 are four levels of salinity that were applied. Detroit Dark Red, Red Ball and Red Ace are some table beet (*Beta vulgaris L.*) cultivars.

irrigated water increased the sodium concentration, as shown in Table 7. In both seasons, the highest significant increase was observed for the 4000 ppm salinity level applied to Detroit Dark Red, followed by the same concentration applied to Red Ball beet.

Effect of Salt Stress on the Anatomical Structure in Leaves of the Studied Table Beet Cultivars. Microscopic measurements of certain anatomical characteristics in vertical sections of table beet leaves indicated the effect of salt stress at a salinity level of 4000 ppm, as a comparison between the studied table beet cultivars (Figures 7a−c and 8).

The thicknesses of the upper and lower epidermal layers, palisade and spongy tissues, midrib zone, and width of vascular bundles were slightly decreased by −14.2, −15.3, −8.5, −7.1, −10.8, and −7.2%, respectively, when treated with 4000 ppm in *B. vulgaris* L. cv. Detroit Dark Red cultivar compared to control or the other two cultivars (Figure 7a). In parallel, the thicknesses

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Table 7. Effect of Irrigation at Various Levels of Saline Water on Nitrogen, Phosphorus, Potassium, and Sodium Contents of Some Table Beet Varieties in 2019−2020 and 2020−2021 Seasons[a]

treatments		nitrogen (g/100 g DW)		phosphorus (g/100 g DW)		potassium (g/100 g DW)		sodium (g/100 g DW)	
		seasons							
		1st	2nd	1st	2nd	1st	2nd	1st	2nd
A: Salinity Levels (ppm)									
control (260)		1.27C	1.19C	0.31C	0.30C	2.09C	2.05C	0.27E	0.28E
1000		1.49B	1.47B	0.34B	0.33B	2.46B	2.36B	0.32D	0.32D
2000		2.02A	1.74A	0.35A	0.35A	2.77A	2.84A	0.38C	0.37C
3000		1.20D	1.07D	0.29D	0.28D	1.76D	1.75D	0.43B	042B
4000		0.98E	0.86E	0.26E	0.24E	1.58E	1.43E	0.50A	0.50A
LSD at 5%		0.06	0.04	0.007	0.006	0.06	0.15	0.14	0.008
B: Varieties									
Detroit Dark Red		1.57A	1.49A	0.34A	0.32A	2.74A	2.64A	0.40A	0.41A
Red Ball		1.34B	1.20B	0.31B	0.29B	1.98B	1.91B	0.36B	0.38B
Red Ace		1.26C	1.12C	0.29C	0.28C	1.67C	1.70C	0.37B	0.35C
LSD at 5%		0.05	0.03	0.005	0.005	0.05	0.11	0.11	0.006
C: Interaction									
control (260)	Detroit Dark Red	1.46d	1.31f	0.34 cd	0.33 cd	2.63c	2.47d	0.28i	0.31h
	Red Ball	1.22fgh	1.12h	0.32f	0.30g	1.93f	1.88fg	0.25j	0.28i
	Red Ace	1.14h	1.15gh	0.28gh	0.29gh	1.71g	1.80fg	0.29i	0.26j
1000	Detroit Dark Red	1.79c	1.90b	0.36bc	0.35bc	3.16b	3.16b	0.33gh	0.34f
	Red Ball	1.41de	1.30f	0.34de	0.32de	2.27d	1.87fg	0.29i	0.32g
	Red Ace	1.27fg	1.22fg	0.32ef	0.31f	1.95f	2.06ef	0.33h	0.30h
2000	Detroit Dark Red	2.19a	2.18a	0.38a	0.37a	3.42a	3.44a	0.41e	0.39de
	Red Ball	1.99b	1.65c	0.35bc	0.35bc	2.66c	2.77c	0.35fg	0.37e
	Red Ace	1.89bc	1.40d	0.33 cd	0.34b	2.24de	2.30de	0.37f	0.35f
3000	Detroit Dark Red	1.30ef	1.13h	0.33de	0.29gh	2.35d	2.24de	0.45 cd	0.45c
	Red Ball	1.16gh	1.08gh	0.28gf	0.27ij	1.63g	1.76g	0.43de	0.41d
	Red Ace	1.14h	1.01h	0.26i	0.27ij	1.31i	1.26h	0.42e	0.39de
4000	Detroit Dark Red	1.12h	0.91i	0.29g	0.26jk	2.14e	1.88fg	0.54a	0.55a
	Red Ball	0.94i	0.86i	0.26i	0.23L	1.44h	1.31h	0.49b	0.50b
	Red Ace	0.87i	0.81i	0.24j	0.23L	1.16j	1.09h	0.47c	0.45c
LSD at 5%		0.11	0.08	0.013	0.011	0.11	0.26	0.02	0.015

[a]Mean values with different letters in the column are statistically different according to DMRT ($p < 0.05$). LSD: Least significant difference. The 260,1000,2000,3000 and 4000 are four levels of salinity that were applied. Detroit Dark Red, Red Ball and Red Ace are some table beet (*Beta vulgaris L.*) cultivars.

of the upper and lower epidermal layers, palisade and spongy tissues, midrib zone, and width of vascular bundles were greatly decreased by −42.1, −27.2, −13.3, −27.0, −25.4, and −18.6%, respectively, at a saline water level of 4000 ppm compared to the control in *B. vulgaris* L. cv. Red Ball cultivar (Figure 7b). Also, the thicknesses of the upper and lower epidermal layers, palisade and spongy tissues, midrib zone, and width of vascular bundles were significantly decreased by −42.8, −33.3, −39.1, −34.6, −30.7, and −25.0%, respectively (Figure 7c), as a response to salt stress at a 4000 ppm level compared to the control in *B. vulgaris* L. cv. Red Ace cultivar.

From an anatomical view, the varied measurements in the studied layers correspond with clear dedifferentiation of the main vascular bundle's elements, especially xylem vessel elements as in both Red Ball and Red Ace cultivars (Figure 7b,c) compared to Detroit Dark Red cultivar, which is distinguished by the area occupied by collenchyma cells behind the main vascular bundle being occupied by larger size and more layers of collenchyma cells (Figure 7a).

Principal Component Analysis (PCA). The most significant amount of data variability was determined using principal component analysis (PCA), which was also used to investigate how different variables and the salinity of irrigation water applications and treatments interacted. In general, the four principal components, PC1 (93.165%), PC2 (4.408%), PC3 (1.399%), and PC4 (0.208%), explained 99.18% of the variability in the total data set (Figure 9). The growth factors (i.e., root diameter, leaf number, TSS, ascorbic acid, chlorophyll, total carotenoids, anthocyanin, proline content, and water saturation) and plant chemical contents (N, P, K, and Na) were all positively correlated with one another.

■ DISCUSSION

Based on research findings, most of the growth and productivity parameters decreased due to the inhibition of photosynthetic activity of sugar synthesis, translocation, and cell elongation above 2000 ppm of saline-irrigated water. Osmotic stress and ionic stress are two significant risks to plant growth caused by salt stress, where soil salinity has been reported to inhibit plant growth due to osmotic stress, followed by ion toxicity.[47] Salinity has an inhibitory effect on seed germination and seedling growth in a variety of crops,[48] as water absorption by the seed was decreased with increasing salinity levels. It had a smaller impact on the leaf number than on the leaf area, and the limitation of leaf expansion was thought to play a major role in the decline in the plant leaf area. This is in line with prior research findings, which revealed that high levels of salinity reduced the leaf area due to a combination of cell number and size reductions.[49] Salt stress in

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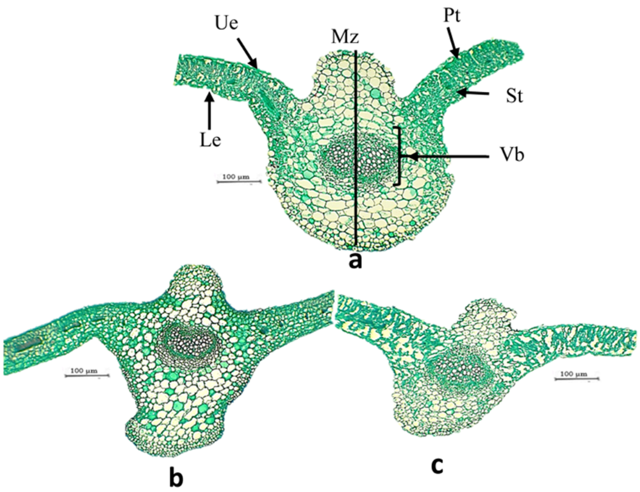

Figure 7. (a−c) A comparative botanical microphotograph of transverse sections through the leaf blade in *Beta vulgaris* L. cultivars under salt stress as follows: (a) *Beta vulgaris* L. Detroit Dark Red cultivar, (b): *Beta vulgaris* L. Red Ball cultivar, and (c) *Beta vulgaris* L. Red Ace cultivar. All from plants grown at 4000 ppm saline water. Abbreviations: Le = lower epidermis; Mz = midrib zone; Pt = palisade tissue; St = spongy tissue; Ue = upper epidermis; and Vb = vascular bundle.

plants has a negative impact on a number of metabolic and physiological processes as it causes oxidative damage in plants.[50]

Some specific proteins and lipids, such as fodder beets, were induced in salt-tolerant crops and contribute to the maintenance of cellular membrane function and structure.[51] Proline, glycine betaine, and polyamines are also known as cellular membrane-protecting agents as they help to stabilize and protect the cellular membrane.[52] It has been established that salt stress correlates with plasma membrane permeability and this property is useful for selecting salt-tolerant crop genotypes.[53] Proline functions as a signaling/regulatory molecule and, in the case of salt stress, could improve the plant's resistance to salinity.[54] Under salinity stress, one important adaptive mechanism of fodder beet plants and other halophytes is the expression of stress proteins.[55] This maintained the integrity, topology, and native configuration of the cell membrane.[56] To reduce cell water potential, fodder beets and other halophytes accumulated inorganic ions in their vacuoles because the energy consumed by synthesizing organic compounds is less than that consumed by absorbing inorganic ions.[57] These crops could accumulate Ca^{2+} and Mg^{2+} ions, thereby aiding in osmotic adjustment. However, both Na^+ and K^+ ion accumulation increased in fodder beets. It is critical to maintain low Na^+ and high K^+ ions in vacuoles in order to maintain various enzymatic processes.[58] This phenomenon demonstrates that fodder beet plants may have a distinct Na^+ absorption pathway that is distinct from the K^+ pathway. Many halophytes and fodder beet plants were found to accumulate proline in order to aid in osmotic adjustment and salinity stress mitigation.[59]

Proline concentrations were found to be higher in shoots of fodder beets and other salt-tolerant plants than in the shoots of salt-sensitive plants.[56] The physiological necessity for membrane stability and osmotic adjustment resulted in high proline content in salt-tolerant fodder genotypes.[60] However, compared to glycine betaine, proline's contribution to osmotic adjustment and salinity stress reduction is negligible.[61] In fact, increased proline under salt stress is thought to be an evolutionary trait in plants since it is an excellent combination of controlling and correcting osmotic pressure as well as a nonenzymatic antioxidant property.[62] The presence of inorganic salt ions in fodder beet and other halophytes was also important for osmotic correction during salinity stress.[63] High quantities of ions such as Cl^-, K^+, and N^+ in fodder beet seedling shoots were suggested to have a role in salinity stress reduction and successful osmotic adjustment during salinity stress in a previous study.[64] One of the explanations for table beet's high resistance to salt stress during the germination stage appeared to be the use of priming treatments to mitigate the detrimental effects of stress.[65] Seed priming increases the number of photosynthetic pigments in plants. One of the most serious consequences of salinity or drought stress in plants is the loss of photosynthetic pigments. This reduction could be due to stomatal or nonstomatal restrictions.[66]

During environmental stress like salinity or drought, stomatal closure is commonly considered the determining factor in the reduction of CO_2 absorption and photosynthesis.[67] Limited stomatal conductance, reduced activities of carbon fixation enzymes, lower quantities of photosynthetic pigments, and

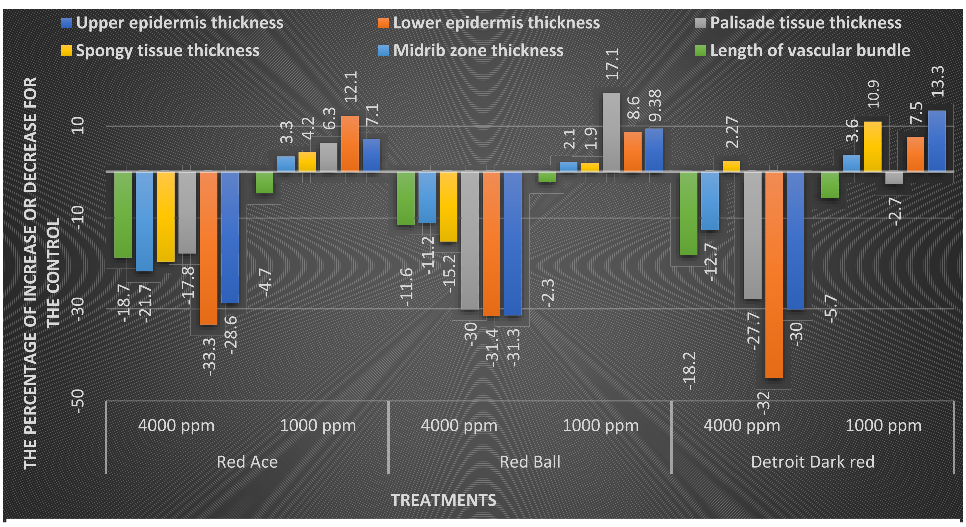

Figure 8. Effect of salt stress on the anatomical structure of Detroit Dark Red, Red Ball, and Red Ace cultivar leaves of table beets (*Beta vulgaris* L.).

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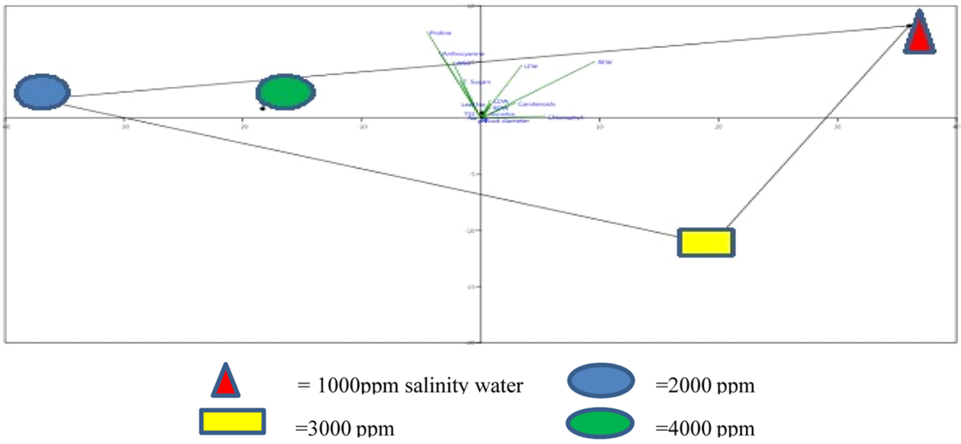

Figure 9. Relationships between parameters and treatment variability in table beet plants using principal component analysis (PCA). PCA clustering was used to analyze the entire data set. The growth parameters, including root diameter, leaf number, TSS, ascorbic acid, chlorophyll, total carotenoids, anthocyanin, proline content, and water saturation, were all positively correlated with each other. Chemical contents of the table beet plants (N, P, K, and Na). Abbreviations: LFW (leaf fresh weight), LDW (leaf dry weight), RFW (root fresh weight), and RDW (root dry weight).

degradation of the photosynthetic apparatus were among the primary mechanisms limiting photosynthesis under salinity stress conditions, according to the researchers.[68] Increased soluble sugar concentrations due to salinity could be attributed to increased enzyme activity, particularly amylase activity.[69] However, the more likely explanation is that cells consume energy in order to resist ion imbalance under stress conditions.[70] Our results clearly showed that the comparative anatomical study of three cultivars of table beets via the measurements of both the upper and lower epidermal layers, palisade and spongy tissues, midrib zone, and width of vascular bundles were slightly decreased, especially in *B. vulgaris* L.[71] Detroit Dark Red cultivar when treated with the highest salinity concentration (4000 ppm) was less than that of the control or the other two cultivars under the same level of saline water.[72] In this regard,[19,73,74] it was found that increasing the salinity caused a decrease in the epidermal thickness in *Salvadora persica* L., and this limited salt ion diffusion to the plants. These findings were found to agree with previous hypotheses and research studies.[63,75] Our future plan includes additional studies to examine plant resilience following the alleviation of salt stress using different varieties of the plant (sugar beet)[76−79] and others.[80] Long-term experiments will be considered to observe growth and physiological changes in sugar beets under prolonged salt stress, allowing for a more accurate assessment of tolerance and adaptability. This would provide a more comprehensive evaluation of the salt tolerance and recovery potential of plants and more accurate results.

■ CONCLUSIONS

The effect of saline water irrigation on the growth and quality parameters of the three beet cultivars (i.e., Detroit Dark Red, Red Ball, and Red Ace) was studied in two successive seasons. Four different salinity levels, 1000, 2000, 3000, and 4000 ppm, were applied, and the underlying parameters were analyzed. As a consequence, salinity levels had different impacts related to the various growth and quality parameters and cultivar types, where it seemed that Detroit Dark Red Beet had the best results among the cultivars under consideration. Irrigation with the first level of saline water (1000 ppm) in both seasons of cultivation showed a maximum increase rate of Detroit Dark Red Beet. Leaf number (mean 36.5%), fresh weight of leaves (mean 16.18%), dry weight of leaves (mean 22.17%), fresh root weight (mean 14.96%), dry root weight (mean 22.29%), and root diameter (mean 13.22%). The second level of salinity (2000 ppm) showed a maximum increase rate of ascorbic acid (mean 16.26%), nitrogen (mean 58.21%), phosphorus (mean 11.94%), potassium (mean 34.66%), and sodium (mean 85.14%). The fourth level of salinity (4000 ppm) exhibited the maximum increase of total sugar (mean 32.72%) and proline (mean 159.82%). On the other hand, the water saturation deficit had its maximum increase rate (mean 50.66%) at the fourth level of salinity for the Red Ball Beet cultivar compared with the other members of the study. The study findings showed that salt stress decreased the *Beta vulgaris* L. cultivars and induced changes in anatomical characteristics (i.e., decrease of epidermal cells, palisade, and spongy tissues), vascular system, and midrib zone parenchyma in leaves.

■ ASSOCIATED CONTENT

Data Availability Statement

The authors declare that all data concerning the research is available within the text in the form of tables and figures. For further information, please refer to the corresponding author.

■ AUTHOR INFORMATION

Corresponding Authors

Amira M. El-Emshaty − *Soils, Water and Environment Research Institute, Agricultural Research Center, Giza 3725004, Egypt*; Email: amiraelemshaty@yahoo.com

Moses V. M. Chamba − *Department of Physics and Applied Sciences, School of Science and Technology, Malawi University of Business and Applied Sciences, Private Bag 303 Blantyre 3, Malawi*; orcid.org/0000-0003-2860-9259; Email: mchamba@mubas.ac.mw

Authors

Ayah T. Zaidalkilani − *Department of Nutrition, Faculty of Pharmacy and Medical Sciences, University of Petra, Amman 11196, Jordan*

Aman H. Al-Kaby − *Department of Horticulture, Vegetable Physiology, College of Agriculture, University of Al-Muthanna, Al Muthanna 1550, Iraq*

Sadeq K. Alhag − *Biology Department, College of Science and Arts, King Khalid University, Abha 61913, Saudi Arabia*

https://doi.org/10.1021/acsomega.4c08161
ACS Omega 2024, 9, 47788−47801

Laila A. Al-Shuraym − *Biology Department, Faculty of Science, Princess Nourah Bint Abdulrahman University, Riyadh 11671, Saudi Arabia*
Zakaria A. Salih − *Research and Training Station, King Faisal University, Al-Ahsa 31982, Saudi Arabia; Department of Food Engineering and Technology, Faculty of Engineering and Technology, University of Gezira, Wad-Medani 21115, Sudan*
Amro Ahmed Taha − *Research and Training Station, King Faisal University, Al-Ahsa 31982, Saudi Arabia; Bee Research Department, Plant Protection Research Institute, Agricultural Research Center, Giza 3725004, Egypt*
Ammar M. Al-Farga − *Department of Biochemistry, College of Sciences, University of Jeddah, Jeddah 21959, Saudi Arabia*
Ashmawi E. Ashmawi − *Department of Horticulture, Faculty of Agriculture, Al-Azhar University, Nasr City, Cairo 11884, Egypt*
Saleh A. Hamad − *Department of Horticulture, Faculty of Agriculture, Al-Azhar University, Nasr City, Cairo 11884, Egypt*
Hany S. Abd El-Raouf − *Department of Agricultural Botany (General Botany), Faculty of Agriculture, Al-Azhar University, Nasr City, Cairo 11884, Egypt*
Shahinaz E. Ahmed − *Department of Agricultural Botany (General Botany), Faculty of Agriculture, Al-Azhar University, Nasr City, Cairo 11884, Egypt*
Ahmed M. El-Taher − *Department of Agricultural Botany (General Botany), Faculty of Agriculture, Al-Azhar University, Nasr City, Cairo 11884, Egypt*
Taghreed A. Badawi − *Plant Production Department, Faculty of Environmental Agricultural Science, Arish University, Arish 45511, Egypt*

Complete contact information is available at:
https://pubs.acs.org/10.1021/acsomega.4c08161

Author Contributions

All the authors contributed to the conception and design of the study. Conceptualization: A.M.E.-E., A.E.A., and A.M.E.-T.; data curation, A.E.A., A.M.E.-T., A.M.E.-E., A.T.Z., and F.A.; formal analysis, A.E.A., S.A.H., A.M.A., A.H.A.-K., and A.M.E.-T.; funding acquisition: S.K.A., L.A.A.-S.H., and E.A.A.-S.H.; investigation: A.M.E.-E. and A.-M.E.-T.; methodology: M.V.M.C., A.M.E.-E., S.A.H., A.A.-F., S.E.A., and A.M.E.-T.; software: A.A.-F., T.A.B., and A.M.E.-T.; supervision: A.M.E.-E.; validation: M.V.M.C. and A.M.E.-T.; visualization: S.E.A.; writing original draft, writing-review, and editing: M.V.M.C., A.M.E.-E., H.S.E.-R., A.A.-F., and T.A.B. All authors have read and agreed to the published version of the manuscript.

Notes

The authors declare no competing financial interest.
The authors declare that this work does not involve live subjects (humans or animals). They also declare that the manuscript has neither been previously published nor is currently submitted for review to any other journal, and will not be submitted elsewhere before a decision is made by this journal.
All authors whose names appear in the submission consent to participate in the research work in various contributions according to their tasks.
All authors agreed with the content and gave explicit consent for submission with no competing interests.

■ ACKNOWLEDGMENTS

This research was funded by Princess Nourah bint Abdulrahman University Researchers Supporting Project number (PNURSP2024R365), Princess Nourah bint Abdulrahman University, Riyadh, Saudi Arabia. The authors extend their appreciation to the Deanship of Scientific Research at King Khalid University for funding this work through Large Groups Project under grant number (R.G.P.2/60/45).

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https://doi.org/10.1021/acsomega.4c08161
ACS Omega 2024, 9, 47788−47801