

## RESEARCH ARTICLE

# Path analysis for growth, yield, and quality traits of sugar beet varieties treated by zinc nanoparticles to alleviate soil salinity stress

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## Abstract

In order to reduce the effects of salinity and increase land productivity, multiple strategies should be used in salt-affected soils.

In two consecutive seasons, the study was conducted in the 2022/23 and 2023/24 seasons at a private farm in Sinnuris, located at latitude 29°17' N and longitude 30°53' E, Fayoum Governorate, Egypt.

The randomized complete block design (RCBD) in a split-plot arrangement in three replications. The results demonstrated that spraying sugar beet varieties with 150 mg/l of nano-zinc during both seasons significantly increased the percentages of total phenol, total chlorophyll, and relative growth rate, as well as root diameter, root length, and root fresh weight/plant. Also, sucrose, extractable sugar, root, and sugar yield/fed. Nano-zinc levels had no discernible marked impact on sodium content in both seasons.

In order to increase root and sugar yields under saline soil conditions, planting a multi-germ variety, Estora-KWS, and sprayed it with 150 mg/l of nano-zinc can be recommended. Estora-KWS and Marwa-KWS multi-germ varieties outperformed all other tested varieties in both seasons, and recorded the highest mean values of root and sugar yields/fed.

There was a strong and positive correlation between root yield/fed and both root diameter and weight. Path coefficient analysis, which compares relationship-based traits, was shown to be reliable and understandable in determining which traits are most helpful in selecting a tolerant sugar beet variety to grow in saline soil.

**Keywords:** *Beta vulgaris*; Zinc Nanoparticle; Sugar yield; Juice quality; Salt stress

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## Introduction

Sugar beets (*Beta vulgaris*, L.) are grown in temperate climates, mostly in the Northern Hemisphere between 30 and 60 degrees north latitude (Abu-Ellail et al. 2021). Nowadays, a significant portion of the sugar industry is made up of sugar beets, which are important produced in subtropical regions (Galal et al. 2022; Ahmed et al. 2023).

Approximately 1.5 million hectares of land are taken out of production each year due to salinization, which accounts for around 7% of all irrigated croplands worldwide (Semida et al. 2015).

Approximately 0.9 million hectares, or 25% of Egypt's total irrigated cultivable croplands, are experiencing salinization issues (FAO, 2022). Sugar beet (*Beta vulgaris* var. *saccharifera*, L.) is one of the most salt-tolerant crops. However, it is considered less tolerant to salinity during the germination, emergence, and seedling stages (Abu-Ellail et al. 2023; Alotaibi et al. 2023). Tolerant crops, such as sugar beet, can be grown successfully on moderately saline soils.

A better understanding of how to promote sugar beet seed emergence in saline field conditions would be helpful if enhancing the germination stage to support sugar beet production in areas where salinity occurs (Kaffka et al. 1999; Aljabri et al. 2023).

Sugar beet plants naturally adapt to withstand salinity up to an electrical conductivity (EC) of 7.0 dSm<sup>-1</sup> without experiencing a significant decrease in yield; each additional EC causes a yield loss of 5.9% per unit (Grieve et al. 2012). Crop growth and survival are negatively impacted by salinity because it causes osmotic and drought stresses, which lead to ionic imbalance because of the high accumulation of sodium (Na<sup>+</sup>) and chlorine (Cl<sup>-</sup>) ions, which cause certain ions to be cytotoxic (Al-Dhumri et al. 2023; Abd-Elrahman et al. 2022).

Zinc plays several physiological functions in plants, including the metabolism of proteins, carbohydrates, lipids, and nucleic acids; photosynthesis; and auxin biosynthesis (Malakoti and Tehrani, 2001). It is a crucial nutrient for sugar beet plants. The primary biochemical and physiological roles of zinc in plants include involvement in the biosynthesis of tryptophan, regulation of carbonic anhydrase, RNA polymerase activation, cytoplasmic membrane stabilization, regulation of oxidative stress via superoxide dismutase, and enhancement of plant resistance to water stress (Hussain et al. 2021).

Zinc oxide (ZnO) is the most widely produced nanomaterial in the world. High catalytic activity, strong photochemical activity, and unique tolerance to biotic and abiotic stress (Sirelkhatim et al. 2015). Additionally, data indicate that ZnO NP accumulation increases stress tolerance in *Beta vulgaris* (Khan and Siddiqui, 2021).

Sugar beet is most sensitive to zinc deficiency in the soil. However, symptoms of deficiency are uncommon (Neamatollahi et al. 2013). Lack of organic manure, a high frequency of sensitive crop cultivation, and excessively high phosphorus (P) rates are some of the factors that can lead to a sugar beet Zn deficiency (Moustafa, 2019). Farahat (2018) studied the effect of Zn nanoparticles spraying sugar beets with 25, 50, 100 ppm on total soluble solids (TSS%) and sucrose sugar beet Oscarpoly cultivar and found that spraying 100 ppm of Zn NPs exhibited a rise in TSS% and sucrose% compared to the control. Amin et al. (2023) used zinc oxide nanoparticles (10, 50, and 100 ppm) as foliar spraying on top of sugar beets and concluded that total chlorophyll, total sugars, and total polyphenols were significantly increased with 100 ppm.

The current work aims to study the possibility of mitigating soil salinity stress on sugar beet varieties, using nano-zinc fertilizer (ZnO) to enhance their

growth and quality traits, as well as to increase root and sugar yields/fed. Also, explore the interrelationships among sugar yield and its related traits using correlation coefficients and path analysis.

## Materials and methods

### Experiments Preparation

The study examined the effects of nano-zinc rates (0, 50, 100, and 150 mg/l) on growth, yield and quality characteristics of five multiterm sugar beet varieties (Estora KWS, Cassiopeia KWS, Marwa KWS, Meralda KWS, and Sugar King) grown in a saline soil in 2022/2023 and 2023/2024 seasons at private farm in in Sinnuris, located at latitude: 29°17' N and longitude: 30°53' E, Fayoum Governorate. Seeds of the tested sugar beet varieties were brought from the Sugar Crops Research Institute, ARC, Giza, Egypt.

The randomized complete block design (RCBD) in a split plot arrangement used to carry out the experiment in three replications. The levels of zinc nanoparticles were allocated in the main plots, and the tested sugar beet varieties were randomly distributed in the subplots. Each nano-zinc treatment was sprayed three times at the four, six, and eight leaf stages. The area of each experimental unit was 15 m<sup>2</sup>, which consisted of five rows of 60 cm apart and 5 m long.

Sugar beet varieties were sown on the 18<sup>th</sup> of September in the 1<sup>st</sup> season and on the 24<sup>th</sup> of September in the 2<sup>nd</sup> one. The previous crop was maize in both seasons. In accordance with Jackson (1973), the experimental soil samples were taken at 0-30 cm depth from the soil surface, prior to cultivation, to determine their physical and chemical characteristics as shown in Table 1. All other agronomic practices for growing the sugar beet crop were done as recommended by the Sugar Crops Research Institute in the region.

**Table 1.** Physical and chemical characteristics of the soil at the experimental site

2022/2023 season							
Particle size distribution %			Texture class	Available nutrients (mg/kg soil)			pH
Sand	Silt	Clay		N	P	K	
22.63	31.98	45.39	Clay loam	49.81	4.28	166	7.08
EC dScm <sup>-1</sup>			Soluble cations and anions (meq/l)				
	Ca <sup>+2</sup>	Mg <sup>+2</sup>	Na <sup>+</sup>	K <sup>+</sup>	HCO <sub>3</sub> <sup>-</sup>	SO <sub>4</sub> <sup>-2</sup>	Cl <sup>-</sup>
4.97	15.47	11.82	20.48	1.93	4.05	22.30	23.35
2023/2024 season							
Particle size distribution %			Texture class	Available nutrients (mg/kg soil)			pH
Sand	Silt	Clay		N	P	K	
21.73	30.25	48.02	Clay loam	51.36	4.64	185	7.04
EC dSm <sup>-1</sup>			Soluble cations and anions (meq/l)				
	Ca <sup>2+</sup>	Mg <sup>2+</sup>	Na <sup>+</sup>	K <sup>+</sup>	HCO <sub>3</sub> <sup>-</sup>	SO <sub>4</sub> <sup>2-</sup>	Cl <sup>-</sup>
4.65	13.61	7.46	21.90	3.53	3.87	21.86	20.77

At harvesting time (210 days after sowing), the following traits were determined in samples of five randomly collected plants:

### Physiological characters

1. Using techniques outlined by Wettstein (1957), the levels of total chlorophyll in the leaves of sugar beet plants were measured at 120 days after thinning.
2. In accordance with Singleton et al. (1999), the total phenolic compounds were measured at a wavelength of 750 nm using a UV/Vis. spectrophotometer manufactured by Jenway, England.
3. It is determined by Hall et al. (1993) that the relative growth rate (RGR) ( $\text{g/g/day}$ ) =  $(\log W_2 - \log W_1) / (t_2 - t_1)$ , where:  $W_1$  and  $W_2$  respectively refer to dry weight at time  $t_1$  and  $t_2$  in days.

### Growth traits

Root diameter (cm), root length (cm) and root fresh weight (kg).

### Quality traits

Root samples were sent to Fayum Sugar Company Laboratories to determine the following quality traits:

1. Sucrose % was measured using a Saccharimeter, according to the method of A.O.A.C. (2005).
2. Impurities % in sugar beet roots (Na, K,  $\alpha$ -amino N) were determined, according to the method of A.O.A.C. (2005).
3. Sugar lost to molasses (SLM%) was calculated as described as Devillers (1988):  

$$\text{SLM\%} = 0.14 (\text{Na} + \text{K}) + 0.25 (\text{Alpha-amino N}) + 0.50$$
4. Extractable sugar (ES %) was estimated as shown by Dexter et al. (1967):  $\text{ES\%} = [\text{sucrose\%} - (\text{SLM\%} + 0.6)]$

### Yield traits

1. Based on root yield/plot (kg), root yield/fed (ton) is determined.
2. Sugar yield/fed (ton) =  $\text{ES\%} \times \text{root yield/fed (ton)} / 100$

### Statistical analysis

The collected data were statistically analyzed using SPSS.15 VR computer program. Means of treatments were compared using LSD at a 5% level of probability, in accordance with Gomez and Gomez (1984). Based on the plot mean, the analysis of variance for the various characters was conducted. Additionally, the correlation coefficient is divided into components of direct and indirect effects using the path coefficient method, which was implemented by Dewey and Lu (1959).

## Results and discussion

### Physiological characters

Data in Table 2 illustrate that increasing the rate of foliar application of nano-zinc up to 150 mg/l considerably and gradually enhanced the total phenol and chlorophyll contents, as well as the relative growth rate (RGR), compared to that left without treatment (control), in the two seasons. These outcomes align with research reported by El-Sayed et al. (2023), who revealed that increasing concentrations of Zn NPs significantly increased levels of chlorophyll, total phenols, and relative growth rate.

The results of this study indicate that ZnO nanoparticles may be applied as a foliar spray to lessen the negative effects of biotic and abiotic stress on sugar beet plants, offering a potential remedy for environmentally friendly and sustainable crop management. Awan et al. (2021) found that the use of micronutrients improves plant metabolism under stress by increasing compounds such as phenols. Zinc spraying increased chlorophyll, demonstrating the important role zinc plays in nitrogen element metabolism and chlorophyll production (Sirelkhatim et al. 2015). Results in the same Table exhibited appreciable variances among the examined beet varieties in the total phenol, chlorophyll, and RGR in both seasons.

The Estora KWS variety scored the highest mean values of total phenol over the other tested sugar beet varieties, in both seasons, there were insignificant variances between Cassiopeia KWS and Meralda KWS as well as between Marwa KWS and Sugar King in the total phenol, in both seasons. Additionally, the data shows that the three varieties, namely Estora KWS, Cassiopeia KWS, and Marwa KWS, substantially recorded higher average values of the total chlorophyll, compared to Meralda KWS and Sugar King, in both seasons. Meanwhile, there were insignificant variances among the three mentioned ones.

Concerning the relative growth rate (RGR), Marwa KWS ranked 1<sup>st</sup> over the other varieties, without a marked difference with Sugar King, in the 1<sup>st</sup> season. In the 2<sup>nd</sup> one, Sugar King was significantly higher than other evaluated varieties in RGR. These results are in line with those found by Abu-Ellail et al. 2023 and 2024, who found that five sugar beet varieties varied considerably in how they responded to stress in terms of physiological traits. The differences between the studied sugar beet varieties in these traits may be attributed to differences in genetic makeup and their interaction with environmental conditions. Islam et al. (2020) clarified that biotic stress also has a negative effect on various parameters of chlorophyll fluorescence, and total polyphenol scavenging activity was remarkably increased in all sugar beet varieties under stress conditions.

Data in Table 2 indicate that total phenol, total chlorophyll, and relative growth rate (RGR) were markedly influenced by the interaction between nano-zinc levels and sugar beet varieties in both seasons. Regarding the results, the roots of Marwa KWS variety markedly contained higher total phenols than those found in Sugar King Variety, when they were sprayed with 0 or 100 mg/l nano-zinc. However, these two varieties did not significantly vary in this trait when they were sprayed with 50 mg/l nano-zinc, in both seasons. Here, there were insignificant variances in the total chlorophyll in leaves of Marwa KWS and Meralda KWS varieties when they were left without nano-zinc. Nevertheless, the two previous

varieties varied substantially when they were given nano-zinc at 100 or 150 mg/l, in both seasons. Regarding RGR, an insignificant difference was detected between Cassiopeia KWS and Marwa KWS in RGR, as they were fertilized with 50 mg/l nano-zinc. However, when the rate of nano-zinc was raised to 100 or 150 mg/l, the variance between the two varieties in RGR amounted to the level of significance, in the 1<sup>st</sup> season. In the second one, an insignificant variance was recorded between Cassiopeia KWS and Sugar King in RGR in this trait, when their tops were sprayed with 150 mg/l, appreciable variances were noticed between them at the rate of nano-zinc, i.e., 50 or 100 mg/l.

**Table 2.** Mean of total phenols, chlorophyll, and relative growth rate (RGR) of five multi-germ sugar beet varieties as affected by nano-zinc rates in 2022/2023 and 2023/2024 seasons.

Treatments		Total phenol		Total chlorophyll		RGR	
		1 <sup>st</sup> season	2 <sup>nd</sup> season	1 <sup>st</sup> season	2 <sup>nd</sup> season	1 <sup>st</sup> season	2 <sup>nd</sup> season
Nano-zinc levels	0 mg/l	132.81	130.13	2.86	2.07	2.15	2.17
	50 mg/l	168.03	165.15	3.69	3.71	2.29	2.37
	100 mg/l	206.72	205.63	4.14	4.07	3.15	3.00
	150 mg/l	229.59	229.86	5.86	4.94	3.76	3.61
<b>LSD 5% level</b>		<b>1.61</b>	<b>1.23</b>	<b>0.68</b>	<b>0.82</b>	<b>0.16</b>	<b>0.21</b>
Sugar beet varieties	Estora KWS	199.12	197.68	4.72	4.43	2.68	2.41
	Cassiopeia KWS	175.21	173.27	4.42	4.47	2.67	3.00
	Marwa KWS	185.47	183.15	4.83	4.08	3.04	2.75
	Meralda KWS	175.47	174.79	3.64	3.17	2.82	2.42
	Sugar King	186.20	184.58	3.10	2.32	2.99	3.36
<b>LSD 5% level</b>		<b>1.63</b>	<b>1.62</b>	<b>0.54</b>	<b>0.67</b>	<b>0.14</b>	<b>0.23</b>
Interaction effect between sugar beet varieties and nano-zinc levels							
Nano-zinc levels	Sugar beet varieties	Total phenol		Total chlorophyll		RGR	
		1 <sup>st</sup> season	2 <sup>nd</sup> season	1 <sup>st</sup> season	2 <sup>nd</sup> season	1 <sup>st</sup> season	2 <sup>nd</sup> season
0 mg/l	Estora KWS	151.85	150.52	2.76	3.53	1.99	5.15
	Cassiopeia KWS	120.38	118.36	2.65	3.48	2.03	5.07
	Marwa KWS	140.11	136.59	2.88	3.55	2.12	4.68
	Meralda KWS	134.18	130.92	2.51	3.34	2.23	3.77
	Sugar King	117.55	114.27	1.64	2.49	2.37	2.92
50 mg/l	Estora KWS	195.09	193.79	6.23	5.46	2.02	2.09
	Cassiopeia KWS	170.65	166.54	5.54	5.07	2.24	2.02
	Marwa KWS	162.45	158.87	5.08	5.24	2.24	2.16
	Meralda KWS	149.12	148.74	4.25	3.46	2.51	2.11
	Sugar King	162.85	157.79	3.03	3.28	2.45	2.45
100 mg/l	Estora KWS	220.03	216.77	6.45	5.59	3.02	2.16
	Cassiopeia KWS	194.76	195.85	7.01	6.17	2.95	2.24
	Marwa KWS	218.39	218.68	6.07	5.62	3.78	2.34
	Meralda KWS	185.07	181.77	4.54	3.74	2.85	2.46
	Sugar King	215.37	215.09	4.08	3.81	3.14	2.65
150 mg/l	Estora KWS	229.49	229.64	5.15	7.42	3.67	2.64
	Cassiopeia KWS	215.03	212.31	5.07	5.32	3.45	3.51
	Marwa KWS	220.93	218.46	4.68	7.29	4.03	2.81
	Meralda KWS	233.49	237.74	3.77	8.38	3.68	2.36
	Sugar King	249.02	251.15	2.92	7.19	3.99	3.69
<b>LSD at 5% level</b>		<b>2.13</b>	<b>1.89</b>	<b>0.87</b>	<b>0.64</b>	<b>0.13</b>	<b>0.21</b>

## Growth traits

The data in Table 3 showed that spraying beets with nano-zinc particles considerably increased root diameter, length, and weight in both seasons compared to the control treatment. Thus, the application of 150 mg/l nano-zinc to beet tops resulted in the highest average values of root diameter, length, and weight in both seasons, and increasing non-zinc levels progressively increased significantly the values of the

three identified traits in both seasons. Teama et al. (2021) showed a gradual increase in the root traits of sugar Beet varieties as the concentration of zinc oxide nanoparticles increased up to 200 mg/l. ZnO NPs effectively improved the fresh root weight, length, and diameter by retaining more water in cells and thus reducing saline stress (Barlóg et al. 2016). Furthermore, the findings show that there were significant differences in root diameter, length, and weight between the tested beet varieties.



Meralda KWS variety surpassed the other examined varieties in root diameter in two seasons, with insignificant variance from Estora KWS in the 1<sup>st</sup> season and from Estora KWS and Cassiopeia KWS in the 2<sup>nd</sup> season. Meralda KWS variety also produced the longest roots with insignificant differences from Cassiopeia KWS and Marwa KWS in the 1<sup>st</sup> season, while in the 2<sup>nd</sup> season, Marwa KWS had the highest root length without marked difference with Meralda KWS and Sugar King Varieties. As for root weight, results showed that Meralda KWS markedly surpassed the other four varieties in root weight in the 1<sup>st</sup> season. While Sugar King produced the heaviest roots with insignificant variance from Estora KWS variety in the 2<sup>nd</sup> one. Also, the interaction between nano-zinc application levels and sugar beet varieties had a significant effect on the root diameter, length, and weight in the first and second seasons (Table 3). Data showed considerable variance in root diameter between Estora KWS and Marwa KWS sprayed with 0, 50, and 100 mg/l nano-zinc, whilst the differences between these varieties were insignificant when treated with 150 mg/l, in the 1<sup>st</sup> season. In the second season, the root diameter of the Meralda KWS variety was markedly higher than that of the Marwa KWS variety when spraying with 50, 100, and 150 mg/l nano-zinc; however, the variance between the same varieties was insignificant under the control treatment.

As for root length, there were insignificant variances between varieties, Cassiopeia KWS and Sugar King when they were left without nano-zinc, but the two varieties significantly differed at 50, and 100 mg/l nano-zinc in the 1<sup>st</sup> season. Insignificant differences in the root length of Estora KWS and Sugar King when they were given nano-zinc at 0 and/or 50 mg/l, in contrast, the same varieties varied substantially when they were given nano-zinc at 100 and/or 150 mg/l, in the 2<sup>nd</sup> season. The results in the same Table pointed to insignificant variances in root weight of Cassiopeia KWS and Marwa KWS when their tops were sprayed with 0, 100, and/or 150 mg/l, however, the two varieties markedly varied in this trait at 50 mg/l nano-zinc in the 1<sup>st</sup> season. In the 2<sup>nd</sup> season, Sugar King and Marwa KWS insignificantly differed in root weight, as they were fertilized with 0, 50, 100 mg/l nano-zinc. However, when the rate of nano-zinc was raised to 150 mg/l, the variance between them in root weight reached the level of significance. The most studied traits, including root fresh weight/plant, root length/plant, and root diameter/plant, increased dramatically with increasing nano-zinc rates (Neamatollahi et al. 2013). The role of Zn in facilitating the utilization of N and P in plants could be responsible for the growth in the top and root fresh weights obtained by Zn application (Mekki 2014).

**Table 3.** Means of root diameter, length and weight of five multi-germ sugar beet varieties as affected by nano-zinc rates in 2022/2023 and 2023/2024 seasons.

Treatments		Root diameter (cm)		Root length (cm)		Root weight (kg)	
		1 <sup>st</sup> season	2 <sup>nd</sup> season	1 <sup>st</sup> season	2 <sup>nd</sup> season	1 <sup>st</sup> season	2 <sup>nd</sup> season
Nano-zinc levels (mg/l)	0 mg/l	11.69	10.42	22.53	21.52	0.77	0.79
	50 mg/l	12.91	12.88	24.46	23.36	0.81	0.83
	100 mg/l	14.34	14.33	26.43	25.00	0.98	0.97
	150 mg/l	16.59	16.35	28.02	27.37	1.04	0.98
<b>LSD at 5% level</b>		<b>0.87</b>	<b>0.69</b>	<b>1.21</b>	<b>1.32</b>	<b>0.09</b>	<b>0.11</b>
Sugar beet varieties	Estora KWS	14.78	13.90	24.69	23.82	0.89	0.94
	Cassiopeia KWS	13.18	13.72	25.57	22.80	0.87	0.85
	Marwa KWS	12.50	13.25	25.67	25.36	0.82	0.84
	Meralda KWS	15.16	14.42	26.00	25.29	1.04	0.88
	Sugar King	13.81	12.20	24.88	24.30	0.89	0.96
<b>LSD at 5% level</b>		<b>1.34</b>	<b>1.02</b>	<b>0.98</b>	<b>1.45</b>	<b>0.07</b>	<b>0.08</b>
Interaction effect between sugar beet varieties and nano-zinc levels							
Nano-zinc levels (mg/l)	Sugar beet varieties	Root diameter (cm)		Root length (cm)		Root weight (kg)	
		1 <sup>st</sup> season	2 <sup>nd</sup> season	1 <sup>st</sup> season	2 <sup>nd</sup> season	1 <sup>st</sup> season	2 <sup>nd</sup> season
0 mg/l	Estora KWS	13.67	12.67	22.33	20.11	0.67	0.85
	Cassiopeia KWS	11.67	11.77	21.67	21.17	0.72	0.79
	Marwa KWS	10.67	9.67	23.67	23.33	0.79	0.73
	Meralda KWS	12.10	9.33	23.83	22.67	0.9	0.76
	Sugar King	10.33	8.67	21.17	20.33	0.77	0.83
50 mg/l	Estora KWS	14.00	13.42	24.18	22.67	0.76	0.81
	Cassiopeia KWS	12.67	12.00	25.50	22.17	0.85	0.76
	Marwa KWS	11.00	12.00	24.64	24.50	0.74	0.82
	Meralda KWS	15.23	14.33	24.33	24.33	0.9	0.81
	Sugar King	11.67	12.67	23.67	23.14	0.82	0.93
100 mg/l	Estora KWS	15.33	14.21	25.83	23.50	1.06	1.07
	Cassiopeia KWS	13.36	15.00	26.45	23.33	0.89	0.91
	Marwa KWS	12.00	13.67	26.87	26.12	0.83	0.92
	Meralda KWS	16.00	15.67	27.50	26.17	1.17	0.99
	Sugar King	15.00	13.12	25.50	25.88	0.94	0.94
150 mg/l	Estora KWS	16.11	15.31	26.41	29.00	1.07	1.01
	Cassiopeia KWS	15.00	16.12	28.67	24.54	1.02	0.93
	Marwa KWS	16.31	17.67	27.50	27.50	0.93	0.90
	Meralda KWS	17.32	18.33	28.33	28.00	1.18	0.95
	Sugar King	18.22	14.34	29.17	27.83	1.01	1.12
<b>LSD at 5% level</b>		<b>0.31</b>	<b>0.64</b>	<b>0.75</b>	<b>0.83</b>	<b>0.10</b>	<b>0.12</b>

## Impurity traits

According to Table 4, the alpha-amino N and K content in both seasons was considerably impacted by the foliar application of nano-zinc. Meanwhile, Na content of root beet was insignificantly influenced by applying nano-zinc levels in both seasons. Whatever, it was observed that the alpha-amino N percentage was substantially decreased when sugar beet tops were sprayed with 50 mg/l nano-zinc in the two seasons.

Additionally, the potassium percentage was markedly reduced by spraying with 150 mg/l nano-zinc compared to that left without treatment (control) in both seasons. Here to, alpha-amino N and K content varied significantly among the tested beet varieties, according to data in the same previous table. Furthermore, the Sugar King variety had the lowest alpha-amino N value, and the Marwa KWS variety produced the lowest K% in both seasons.

These results are similar to those obtained by Heydarzadeh et al. (2021), who found that applying nano-zinc led to a decrease in the Na, K, and amino nitrogen contents of sugar beet up to 150 mg/l. As shown in Table 4 that Na% and K% were not significantly affected by the interaction between sugar beet varieties and nano-zinc levels in both seasons.

However, there was an insignificant difference in alpha-amino N% between Meralda KWS and Sugar King varieties when they were given nano-zinc at 50 mg/l, but the two varieties varied substantially in this trait when they were sprayed with 0, 100, and 150 mg/l nano-zinc in the 2<sup>nd</sup> season. This finding indicates that different sugar beet varieties did not respond similarly to varying concentrations of nano-zinc. In both seasons, the interaction between the tested sugar beet varieties and nano-zinc had a significant impact on alpha-amino N. The impurity trait values were lowest in those treated with 150 mg/l of nano-zinc (Stevens and Mesbah 2005).

**Table 4.** Means of impurities traits of five multi-germ sugar beet varieties as affected by nano-zinc rates in 2022/2023 and 2023/2024 seasons.

Treatments		Alph-amino N		Na		K	
		1 <sup>st</sup>	2 <sup>nd</sup>	1 <sup>st</sup>	2 <sup>nd</sup>	1 <sup>st</sup>	2 <sup>nd</sup>
		season	season	season	season	season	season
Nano-zinc levels (mg/l)	0 mg/l	1.32	1.43	2.41	3.31	1.64	1.76
	50 mg/l	1.17	1.28	2.49	3.39	1.38	1.50
	100 mg/l	1.37	1.48	2.47	3.37	1.48	1.60
	150 mg/l	1.67	1.66	2.57	2.57	1.45	1.45
	<b>LSD at 5% Level</b>	<b>0.02</b>	<b>0.03</b>	<b>NS</b>	<b>NS</b>	<b>0.04</b>	<b>0.05</b>
Sugar beet varieties	Estora KWS	1.47	1.54	2.48	3.16	1.72	1.81
	Cassiopeia KWS	1.23	1.34	2.46	3.13	1.42	1.51
	Marwa KWS	1.73	1.76	2.72	3.39	1.22	1.31
	Meralda KWS	1.35	1.40	2.41	3.08	1.67	1.76
	Sugar King	1.17	1.28	2.38	3.05	1.44	1.53
	<b>LSD at 5% level</b>	<b>0.05</b>	<b>0.02</b>	<b>NS</b>	<b>NS</b>	<b>0.03</b>	<b>0.04</b>
Interaction effect between sugar beet varieties and nano-zinc levels							
Nano-zinc levels (mg/l)	Sugar beet varieties	Alph-amino N		Na		K	
		1 <sup>st</sup>	2 <sup>nd</sup>	1 <sup>st</sup>	2 <sup>nd</sup>	1 <sup>st</sup>	2 <sup>nd</sup>
		season	season	season	season	season	season
0 mg/l	Estora KWS	1.31	1.42	2.48	3.38	1.51	1.63
	Cassiopeia KWS	1.45	1.56	2.28	3.18	1.82	1.94
	Marwa KWS	0.94	1.05	2.65	3.55	1.04	1.16
	Meralda KWS	1.51	1.62	2.27	3.17	2.41	2.53
	Sugar King	1.39	1.50	2.39	3.29	1.45	1.57
50 mg/l	Estora KWS	1.13	1.24	2.53	3.43	1.2	1.32
	Cassiopeia KWS	1.30	1.41	2.45	3.35	1.84	1.96
	Marwa KWS	1.48	1.59	2.23	3.13	0.42	0.54
	Meralda KWS	0.97	1.08	2.63	3.53	1.42	1.54
	Sugar King	1.00	1.11	2.63	3.53	2.05	2.17
100 mg/l	Estora KWS	2.02	2.13	2.61	3.51	2.94	3.06
	Cassiopeia KWS	1.12	1.23	2.53	3.43	1.05	1.17
	Marwa KWS	1.28	1.39	2.39	3.29	1.23	1.35
	Meralda KWS	1.47	1.58	2.23	3.13	1.28	1.40
	Sugar King	0.96	1.07	2.60	3.50	0.91	1.03
150 mg/l	Estora KWS	1.40	1.38	2.31	2.31	1.24	1.24
	Cassiopeia KWS	1.03	1.14	2.57	2.57	0.96	0.96
	Marwa KWS	3.22	3.01	3.59	3.59	2.18	2.18
	Meralda KWS	1.43	1.33	2.50	2.50	1.55	1.55
	Sugar King	1.31	1.43	1.89	1.89	1.36	1.36
	<b>LSD at 5% level</b>	<b>0.03</b>	<b>0.06</b>	<b>NS</b>	<b>NS</b>	<b>NS</b>	<b>NS</b>

## Quality traits

The data in Table 5 showed that spraying with nano-zinc had a significant impact on sucrose percentage, sugar loss to molasses, and extractable sugar in both seasons. It also increased the rate of foliar application of nano-zinc up to 150 mg/l considerably and gradually increased sucrose percentage and extractable sugar percentage in both seasons when compared to the control, which did not receive treatment.

On the other hand, sugar loss to molasses percentage was considerably reduced at 50 mg/l and 150 mg/l nano-zinc compared to the control in the 1<sup>st</sup> and 2<sup>nd</sup> seasons, respectively. Nano-zinc (150 mg/l) attained the highest concentrations of sucrose % in either treated sugar beet varieties. Achieving the highest extracted sugar when sugar beet plants were sprayed with 150 mg/l may be due to the differences in the ability of nano-zinc rates to enhance drought tolerance

at the recommended vital for the plant without appreciably increasing impurity contents (Hefny and Said 2021). Sugar beet varieties significantly differed in sucrose percentage and extractable sugar percentage. In contrast, beet varieties insignificantly differed in sugar loss to molasses in the two seasons.

In 1<sup>st</sup> season, Estora KWS gave the highest sucrose and extractable sugar percentages with insignificant differences from the Marwa KWS variety. In the 2<sup>nd</sup> season, Meralda KWS scored the highest sucrose percentage and extractable sugar percentage over the other tested varieties. Sucrose percentage, sugar loss to molasses%, and extractable sugar% were significantly affected by the interaction between sugar beet varieties and nano-zinc levels in both seasons, except sugar loss to molasses in the 2<sup>nd</sup> season.

**Table 5.** Mean of sucrose%, sugar lost to molasses (SLM %), and extractable sugar% (ES %) of five multi-germ sugar beet varieties as affected by nano-zinc levels in 2022/2023 and 2023/2024 seasons

Treatments		Sucrose%		SLM %		ES%	
		1 <sup>st</sup> season	2 <sup>nd</sup> season	1 <sup>st</sup> season	2 <sup>nd</sup> season	1 <sup>st</sup> season	2 <sup>nd</sup> season
Nano-zinc levels (mg/l)	0 mg/l	16.08	15.05	1.40	1.57	14.08	12.88
	50 mg/l	16.74	16.30	1.34	1.51	14.81	14.19
	100 mg/l	17.46	17.12	1.40	1.57	15.47	14.96
	150 mg/l	18.56	18.12	1.48	1.48	16.47	16.03
	<b>LSD at 5% level</b>	<b>0.58</b>	<b>0.13</b>	<b>0.01</b>	<b>0.03</b>	<b>0.16</b>	<b>0.09</b>
Sugar beet varieties	Estora KWS	17.92	16.46	1.46	1.59	15.87	14.28
	Cassiopeia KWS	16.75	16.39	1.35	1.48	14.80	14.31
	Marwa KWS	17.85	16.88	1.48	1.61	15.77	14.67
	Meralda KWS	16.67	17.68	1.41	1.53	14.67	15.55
	Sugar King	16.85	15.82	1.33	1.46	14.92	13.77
	<b>LSD at 5% level</b>	<b>0.34</b>	<b>0.59</b>	<b>NS</b>	<b>NS</b>	<b>0.14</b>	<b>0.10</b>
Interaction effect between sugar beet varieties and nano-zinc levels							
Nano-zinc levels (mg/l)	Sugar beet varieties	Sucrose%		SLM %		ES %	
		1 <sup>st</sup> season	2 <sup>nd</sup> season	1 <sup>st</sup> season	2 <sup>nd</sup> season	1 <sup>st</sup> season	2 <sup>nd</sup> season
0 mg/l	Estora KWS	16.74	14.95	1.39	1.56	14.75	12.79
	Cassiopeia KWS	15.71	13.87	1.44	1.61	13.67	11.66
	Marwa KWS	16.85	15.63	1.25	1.42	15.00	13.61
	Meralda KWS	15.77	16.46	1.53	1.70	13.64	14.16
	Sugar King	15.31	14.33	1.39	1.56	13.32	12.17
50 mg/l	Estora KWS	17.60	15.69	1.30	1.48	15.70	13.62
	Cassiopeia KWS	16.44	16.54	1.43	1.60	14.41	14.34
	Marwa KWS	17.29	16.66	1.24	1.41	15.45	14.65
	Meralda KWS	16.19	17.22	1.31	1.48	14.28	15.14
	Sugar King	16.20	15.38	1.41	1.58	14.19	13.20
100 mg/l	Estora KWS	17.86	16.94	1.78	1.95	15.48	14.39
	Cassiopeia KWS	16.74	17.25	1.28	1.45	14.86	15.20
	Marwa KWS	18.24	17.12	1.33	1.50	16.31	15.02
	Meralda KWS	16.86	18.19	1.36	1.53	14.90	16.06
	Sugar King	17.61	16.12	1.23	1.40	15.78	14.12
150 mg/l	Estora KWS	19.49	18.26	1.35	1.35	17.54	16.31
	Cassiopeia KWS	18.12	17.89	1.25	1.25	16.27	16.04
	Marwa KWS	19.03	18.11	2.11	2.11	16.32	15.40
	Meralda KWS	17.86	18.86	1.42	1.42	15.84	16.84
	Sugar King	18.28	17.46	1.28	1.28	16.40	15.58
	<b>LSD at 5% level</b>	<b>0.94</b>	<b>1.01</b>	<b>0.03</b>	<b>NS</b>	<b>0.11</b>	<b>0.17</b>

In the 1<sup>st</sup> season, an insignificant variance was noticed between Cassiopeia KWS and Marwa KWS varieties in sucrose % when they were sprayed with 50 and/or 150 mg/l, but the two varieties varied markedly at the level of 0 and/or 100 mg/l nano-zinc.

However, in the 1<sup>st</sup> one, an appreciable variance was recorded between Estora KWS and Sugar King in sugar loss to molasses % when their tops were sprayed with 50, 100, and/or 150 mg/l, but they did not differ markedly when they were left without treatment (control). According to the results, there was a significant difference in the percentage of extracted sugar between the Cassiopeia and Meralda KWS varieties when 50 and/or 150 mg/l nano-zinc was applied. However, when they were sprayed with 0 and/or 100 mg/l nano-zinc, there was no significant difference in this trait in the first season.

The superiority of varieties in achieving the highest extracted sugar when sugar beet plants were fed with 150 mg/l may be due to the ability of nano-zinc rates to enhance drought tolerance at the recommended rate necessary for the requirements of the plant without appreciably increasing impurities content. Significant differences among the tested varieties in the quality characteristics of sugar beet grown under newly reclaimed saline soil (Piskin, 2017). These findings are consistent with those reviewed by Abu-Ellail et al. (2023), who claimed that sugar beet varieties differed in terms of their sucrose content; as a result, their extractable sugar and differences were primarily caused by their different maturity states, which were impacted by their make-up.

## Yield traits

Data in Table 6 showed that increasing the rate of nano-zinc foliar application up to 150 mg/l significantly and gradually increased the root and sugar yield. Results in the same previous table showed appreciable variances among the sugar beet varieties in the root and sugar yield in both seasons.

Additionally, the data indicated that in the first season, Meralda KWS variety had the highest root yield, followed by Estora KWS and Marwa KWS; meanwhile, there were insignificant variances between the last two varieties. In the second season, Estora KWS variety recorded the highest value of this trait, with negligible differences from the Marwa KWS variety. The variations among the sugar beet varieties under investigation may result from differences in the gene composition and how each variety reacts to its surroundings. The employment of micronutrients such as manganese, zinc, and iron in an equilibrium can enhance and increase sugar beet productivity of root and sugar yields (Hassnein et al. 2019).

On the other hand, the three varieties, namely Estora KWS, Marwa KWS, and Meralda KWS, substantially scored higher values of sugar yield, compared to Cassiopeia KWS and Sugar King, in both seasons. Meanwhile, there were insignificant variances among the three mentioned ones.

The findings presented in Table 6 demonstrated that the combination of the nano-zinc treatment and the varieties in the two seasons had a significant impact on the yield traits. Concerning root yield, there were insignificant differences in root yield between Estora KWS and Marwa KWS varieties when they were treated with nano-zinc at 0 and/or 50 mg/l, while the two varieties were markedly different when they were sprayed with nano-zinc at 100 and/or 150 mg/l in the 1<sup>st</sup> season.

The results showed that the roots of Estora KWS variety markedly contained higher root yield/fed than that found in Marwa KWS variety, when they were sprayed with 150 mg/l nano-zinc. However, these two varieties did not markedly differ in this trait when they were sprayed with 0, 50, and 100 mg/l nano-zinc in the 2<sup>nd</sup> season.

The superiority of these varieties is due to their better root traits, i.e., diameter, length, and fresh weight root (Table 3). With respect to the sugar yield trait, an insignificant variance in the sugar yield value of Marwa KWS and Meralda KWS varieties when they were sprayed with nano-zinc at 50 and/or 150 mg/l; however, the two varieties varied substantially when they were given nano-zinc at 0 and/or 100 mg/l in the 1<sup>st</sup> season.

In the 2<sup>nd</sup> season, Cassiopeia KWS and Sugar King varieties did not differ markedly in their sugar yields by applying 50 mg/l nano-zinc, while the two varieties varied substantially when they were given nano-zinc at 0, 100, and 150 mg/l. It was noticed that Estora KWS variety exhibited the highest value of sugar yield over the other tested sugar beet varieties at 150 mg/l nano-zinc in both seasons and without appreciable variance with Marwa KWS variety in the 1<sup>st</sup> one.

These results may be due to the main components of sugar yield, i.e., extractable sugar% % (Table 5) and root yield /fed (Table 6). Furthermore, a range of differences in root parameters were observed by variations in sugar beet root yield by varieties that were highly significant when applying nano-zinc enhanced root yield and associated characteristics, according to Barlóg et al. (2016) and Mekdad a Rady (2016) who showed that supplement mixtures of micronutrient (Fe, Zn and Mn) improved yield and its components of sugar beet.



**Table 6.** Means of root and sugar yields of five multi-germ sugar beet varieties as affected by nano-zinc levels in 2022/2023 and 2023/2024 seasons

Treatments		Root yield/fed (ton)		Sugar yield/fed (ton)	
		1 <sup>st</sup> season	2 <sup>nd</sup> season	1 <sup>st</sup> season	2 <sup>nd</sup> season
Nano-zinc levels (mg/l)	0 mg/l	21.86	23.11	3.08	2.99
	50 mg/l	23.84	24.49	3.53	3.48
	100 mg/l	26.67	25.48	4.13	3.81
	150 mg/l	29.92	27.97	4.93	4.48
	<b>LSD at 5% level</b>	<b>1.13</b>	<b>1.17</b>	<b>0.17</b>	<b>0.12</b>
Sugar Beet varieties	Estora KWS	26.08	27.54	4.16	3.96
	Cassiopeia KWS	21.40	23.44	3.21	3.40
	Marwa KWS	26.53	26.40	4.20	3.88
	Meralda KWS	28.32	25.20	4.17	3.93
	Sugar King	25.53	23.73	3.85	3.29
	<b>LSD at 5% level</b>	<b>1.08</b>	<b>1.21</b>	<b>0.11</b>	<b>0.13</b>
Interaction effect between sugar beet varieties and nano-zinc levels					
Nano-zinc levels (mg/l)	Sugar beet Varieties	Root yield/fed (ton)		Sugar yield/fed (ton)	
		1 <sup>st</sup> season	2 <sup>nd</sup> season	1 <sup>st</sup> season	2 <sup>nd</sup> season
0 mg/l	Estora KWS	22.72	25.03	3.35	3.20
	Cassiopeia KWS	16.45	19.87	2.25	2.32
	Marwa KWS	21.90	25.07	3.28	3.41
	Meralda KWS	26.29	23.57	3.59	3.34
	Sugar King	21.92	21.99	2.92	2.68
50 mg/l	Estora KWS	25.20	26.66	3.96	3.63
	Cassiopeia KWS	18.97	21.81	2.73	3.13
	Marwa KWS	24.40	25.77	3.77	3.77
	Meralda KWS	26.29	25.24	3.75	3.82
	Sugar King	24.34	22.96	3.46	3.03
100 mg/l	Estora KWS	26.81	27.74	4.15	3.99
	Cassiopeia KWS	21.92	23.57	3.26	3.58
	Marwa KWS	28.81	26.62	4.70	4.00
	Meralda KWS	29.46	25.57	4.39	4.11
	Sugar King	26.34	23.89	4.16	3.37
150 mg/l	Estora KWS	29.59	30.74	5.19	5.01
	Cassiopeia KWS	28.27	28.49	4.60	4.57
	Marwa KWS	31.00	28.13	5.06	4.33
	Meralda KWS	31.23	26.40	4.95	4.44
	Sugar King	29.52	26.07	4.84	4.06
	<b>LSD at 5% level</b>	<b>1.23</b>	<b>1.34</b>	<b>0.15</b>	<b>0.13</b>

## Correlation coefficient analysis

At the 1% probability level, positive and highly significant correlations were found between root yield and root diameter ( $r = 0.982^{**}$ ), root fresh weight/plant ( $r = 0.964^{**}$ ), sucrose% ( $r = 0.861^{**}$ ), and extractable sugar% ( $r = 0.821^{**}$ ) over the two seasons.

This follows the correlation analysis of some of the traits (Table 7). At the 1% probability level, there were strong positive correlations between the fresh weight of the roots per plant and the root diameter ( $r = 0.897^{**}$ ), root length ( $r = 0.744^{**}$ ), sucrose percentage ( $r = 0.634^{*}$ ), extractable sugar percentage ( $0.591^{*}$ ), sugar yield ( $r = 0.869^{**}$ ), and root yield ( $r = 0.964^{**}$ ) in the two seasons. In addition, at a 1% probability level, sugar yield/fed was positively correlated with each of the following: sucrose percentage ( $r = 0.963^{**}$ ), root diameter ( $0.913^{**}$ ), root weight/plant

( $r = 0.869^{**}$ ), and root yield/fed ( $r = 0.902^{**}$ ) for two seasons. On the contrary, a negative significant correlation was detected between SLM percentage and each of sucrose percentage ( $-0.647^{*}$ ), extractable sugar percentage ( $-0.571$ ), and sugar yield ( $-0.801^{**}$ ).

These outcomes were consistent with the findings published by Abu-Ellail et al. (2023) and Nassar et al. (2023) reported that a significant and positive correlation exists between sucrose percentage, root, and sugar yields. One selection criterion for high yield is root circumference, which has a positive correlation with root yield.

Additionally, a positive correlation between root yield and root weight was discovered, and Alawad et al. (2024) recorded that root yield is an important variable relating to sugar beet yield.

**Table 7.** Correlation coefficient analysis for root and sugar yields and its related traits under different levels of nano zinc through over two years.

Traits	RL	RD	RW	SU%	ES %	S LM%	RY
RL							
RD	0.684*						
RW	0.744**	0.897**					
SU%	0.354	0.469	0.634*				
ES%	0.465	0.391	0.591*	0.935**			
SLM%	0.374	0.235	0.341	- 0.647*	- 0.571*		
RY	0.698*	0.982**	0.964**	0.861**	0.821**	- 0.239	
SY	0.321	0.913**	0.869**	0.963**	0.936**	- 0.801**	0.902**

Abbreviations: RL (root length), RD (root diameter), RW (root weight), SU% (sucrose %), ES% (extractable sugar %), SLM% (sugar lost to molasses, RY (root yield), SY (sugar yield).

\* and \*\* mean significant at 5 and 1% level of probability, respectively

### Path coefficient analysis

The pairwise correlation coefficients between the sugar yield (as a dependent variable) and each attribute (as an independent variable) were divided into direct and indirect effects on the sugar yield using the path coefficient analysis technique to specify the causal interrelationships.

In the current study, sugar yield was the dependent variable, and the other attributes were made independent variables.

Figure 1 displays a matrix of the yield-related traits' individual and combined effects on sugar beet yield. It is noteworthy that, except for the top yield and root length, the diagonal values, which indicate direct effects, were positive when they were both less than one.

The root yield (0.607) had the biggest direct impact, followed by the root diameter (0.251), the sucrose percentage (0.290), and the extractable sugar percentage (0.303).

This example clearly shows how beneficial path-coefficient analysis is. The data indicated a negative, nearly zero relationship between root length and sugar yield at the level of the simple correlation coefficient (-0.102).

Nevertheless, the direct effect appears to be a somewhat negative relationship between top and sugar yields (-0.319) when the indirect effects are isolated from the simple correlation coefficient using path analysis. Root length had moderately positive indirect effects through extractable sugar percentage and sucrose percentage (0.283 and 0.264), and it was positively correlated with top yield (0.021).

However, when viewed indirectly through the other traits, it had negative correlations that may have been negligible.

Through a strong correlation (0.418), the only significant indirect effect component was identified in root diameter via root yield.

Root diameter was 0.233, and the indirect effects of top yield via root yield were positive and strong (0.502); however, its indirect impacts were negative through the remaining characteristics and might be insignificant. Indirect effects on extractable sugar percentage through sucrose percentage were noted on the same side (0.297).

Additionally, the other characteristics demonstrate that sucrose percentage had 0.314 indirect impacts, whereas its association with extractable sugar percentage had negligible and negative indirect effects.

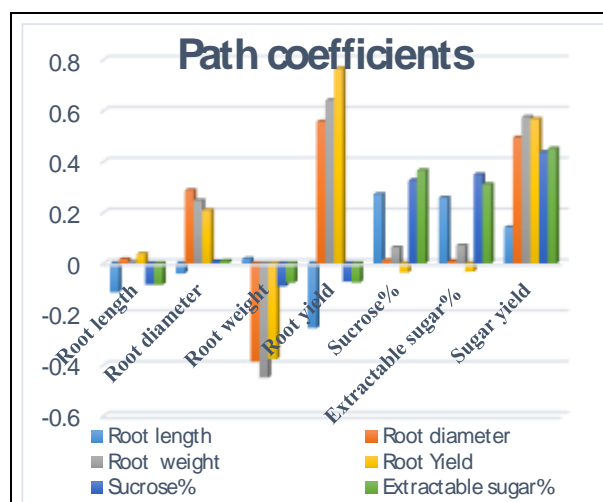
The importance of indirect selection may be considered when direct effects are either nonexistent or of negligible importance.

The examined traits of the tested beet varieties varied considerably, according to Makhoul et al. (2021).

There were notable differences between sugar beet varieties in terms of sucrose percentage, extractable sugar percentage, and root fresh weight per plant, and root and sugar yields (El-Kady et al. 2021).

The present findings were consistent with those of Yan and Kang (2003), who highlighted the value of path analysis in determining selection criteria based on sugar beet yield components.

Taking root diameter and top yield into account, the indirect effect components were more significant than the direct effect components. Indirectly increasing sugar yield in sugar beet would be possible through simultaneous selection, which considers these pairs of traits.



**Figure1.** Path coefficients represent the direct and joint effects of yield along with related traits

## Conclusions

In general, it can be said that applying 150 mg/l of nano-zinc micronutrients greatly enhanced the root and sugar yields of sugar beet varieties. The studied varieties were close in their performance with an increasing rate of spraying with nano-zinc, but the varieties (Estora-KWS and Marwa-KWS) were better than others in most of the studied traits. Root diameter and weight at harvest are highly correlated with crop root yield. Root yield is closely associated with the weight and diameter of the crop roots at harvest. Moreover, a significant correlation exists between the percentage of sucrose and extractable sugar and sugar yield.

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