

## Effect of Nano-NPK and Proline application on reducing salt stress on the vegetative and chemical traits of Pomegranate Seedlings Wonderful cultivar

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### Abstract

The experiment was conducted in lathhouse of the Department of Plant Production Techniques / Musayyib Technical College, during the growing seasons 2021 and 2022 on pomegranate seedlings cultivar WONDERFUL at the age of one year. To study the effect of nano NPK application and proline spraying on growth and phenolic compounds of WONDERFUL cultivar pomegranate seedlings under salt stress. The experiment included three factors, the first factor was irrigation the seedlings with four levels of saline water (9, 6, 3, 1.3 dSm<sup>-1</sup>), the second factor included three concentrations of NPK Nano fertilizer (0,75, 150 mg L<sup>-1</sup>), The third factor was spraying with the amino acid proline at a concentration of (0,300 mg L<sup>-1</sup>) and the interactions between them. The vegetative and chemical measurements were performed (leaf area, chlorophyll, protein percentage, dry matter percentage in leaves, carbohydrates percentage), The results showed that the salinity level of 1.3 dS.m<sup>-1</sup> in most of the traits represented (leaf area, chlorophyll, carbohydrates) compared to the salinity level of 9 dSm m<sup>-1</sup>, which gave the lowest averages. chlorophyll, carbohydrates, protein), and for the effect of spraying proline, the concentration of 300 mg L<sup>-1</sup> in some traits (chlorophyll, carbohydrates) excelled .The research was carried out according to a completely randomized DESIGN (CRD) with three replicates, each replicate contains 24 treatments with three seedlings for the experimental unit.

### Introduction

Pomegranate belongs to the family Punicaceae and it is *Punica granatum* L. It is located among deciduous fruit trees and is widespread in temperate regions. Pomegranate is believed. It is located among deciduous fruit trees and is widespread in temperate regions. It is believed to be native to Iran, southwest Asia and Iraq (18), There are many local cultivars in Iraq, but the wonderful American cultivar has recently spread and gained the attention of researchers because of its strong vegetative growth traits, abundant production, and its suitability for the conditions of Iraq (1). Pomegranate production was estimated at 241,671 tons for the summer season 2020, at an average of 94.9%. Diyala ranked first in terms of production, as it reached 132767 tons or 54.94%, Followed by Salah El-Din province, where its production amounted to 9,203 tons, with an average of 28.38%. Karbala province ranked third, with its production reaching 8454 tons, or 50.3%, while the rest of the province accounted for

3.28% of the total production of Iraq, and the production of pomegranate represented 28.16% of the total production of summer fruit trees in Iraq (10). Pomegranate leaves contain active compounds such as tannins, flavonoids, and phenols that have been shown to be effective in treating pathogens (6);(20).Salinity, whether soil salinity or the salinity of irrigation water, is one of the most important challenges facing the agricultural sector, especially in arid and semi-arid areas, and it negatively affects the growth and productivity of plants, where its accumulation leads to inhibition of the activity of enzymes as well as affects all metabolic and physiological processes. Iraq is at the forefront of the Arab countries affected by salinity (5). Because of the damage caused by salinity, it is necessary to use anti-saline agents to reduce its damage to the plant, including proline. It is a natural amino acid in the plant that accumulates in the plant when exposed to salt stress. It stimulates plant growth and increases its response to fertilizers and salinity tolerance. This acid plays a role in controlling

the osmotic effort and stability of cell membranes. It is considered a source of nitrogen and carbon needed for growth. Proline is one of the important means that reduce the harmful effects of salinity (22). Fertilizer is the technology of mixing soluble fertilizers with irrigation systems whether it is drip, sprinkler irrigation or Addition to water tanks (11). This technique is one of the means that maintains a constant level of nutrients in the soil, improves their distribution in the root zone, and increases soil fertility (21). Nanotechnology is one of the modern technologies in agricultural production, and the word “nano” is used in science to refer to a unit of measurement of one-millionth of a millimetre. This technology provides the possibility of radical changes in agriculture, improving the ability of plants to absorb nutrients and enhancing fertilizer efficiency. Nanofertilizers are the best alternative to traditional fertilizers due to their unique characteristics of small size, large surface area and lower melting point than in their natural state (2). In view of the lack of studies on the management of nano-nutrients and the spraying of proline on seedlings of Wonderful pomegranate under salt stress, this study was conducted with the aim of:

- 1) Knowing the tolerance of pomegranate wonderful seedlings to salinity levels of irrigation water.
- 2) Possibility of improving the salinity tolerance of the pomegranate wonderful seedlings by using the application technique and spraying Proline separately.
- 3) Knowledge of the combined effects of Nano NPK and proline application on the tolerance of pomegranate wonderful seedlings to a salinity of irrigation water.

### Materials and methods

The seedlings were obtained from the Najaf private nursery / Department of Horticulture of the Ministry of Agriculture in September 2021, as homogeneous as possible, at the age of one year, and they were prepared for the

experiment, where they were planted with polyethylene bags. 3: Service operations were conducted for all seedlings, from watering and removing the weeds, the floor of the lathouse was furnished with nylon to prevent the growth of the weed and to avoid the descent of salty irrigation water to the floor of the lathouse and to cover the surface of the canopy with saran to protect the seedlings from the heat and harmful sunlight. Factors ( $4 * 3 * 2$ ) and three replicates with 24 experimental units (3 seedlings for the experimental unit). The factors were as follows: The first factor: - NPK nano fertilizer was used (12:12:36) and according to the recommendation, three concentrations (150,75,0) mg liter<sup>-1</sup> were added to the ground after dissolving these levels with saline concentrations and watering the seedlings according to the required concentrations. The second factor: - Irrigation with four levels of saline water (9,6,3,1.3) Ds.m<sup>-1</sup>, and the saline irrigation water was prepared by dissolving NaCl salt in tanks of 150 liters for each salt level according to the concentrations shown above. The concentrations were measured with an Ec meter. The third factor: - spraying the shoots with the amino acid Proline at two levels (300.0) mg L<sup>-1</sup> using a 2-liter sprayer. These concentrations are within the recommended limits. The seedlings were sprayed with eight sprays, 4 sprays in the first season and 4 sprays in the second season. The experiment was completed a month after the last spray was scheduled. The samples were taken for analysis and the results are taken for analysis using the GENSTAT program and the differences between the means were compared according to the least significant difference test (LSD) at the level of 0.05).

### Studied traits:

**Leaf area (cm):** This trait was calculated on the basis of the leaf area, as 6 full-width leaves were taken from each seedling and printed on white A4 paper, and then the reading was taken with a planimeter. The device's lens was passed over the paper and the process was repeated three times, after which the average leaf area was calculated for each seedling and

the leaf area rate was calculated according to the following equation.

The leaf area of a seedling = average leaf area x average number of leaves per seedling

#### **Chlorophyll content of leaves SPAD UNIT:**

Chlorophyll was measured in pomegranate leaves for each seedling at the end of the experiment with a SPAD device. Unit and calculate the rate for each experimental unit.

**The percentage of carbohydrates in the leaves:** The carbohydrates in the leaves was estimated according to the method (14) in the laboratory of Al-Fadil / Babylon.

**Protein percentage:** The protein content of leaves was estimated based on their nitrogen content according to the following equation:

Percentage of protein in leaves = Percentage of nitrogen x 6.25

**Percentage of dry matter in leaves:** Samples were taken from pomegranate leaves for each seedling, then the wet weight of samples was taken using a sensitive scale.

The percentage of dry matter in the leaves =  $\frac{\text{dry sample weight}}{\text{fresh sample weight}} \times 100$

Then it was dried in the oven at 65 °C until the weight was stable (23), then measured the percentage of dry matter for the leaves according to the equation:

## **Results**

The data in Table (1) indicate that the salinity of the irrigation water showed significant effects on the leaf area. The salinity treatment was superior to 1.3 ds m<sup>-1</sup> by giving the highest average, which amounted to 1048 cm<sup>2</sup>, while the salinity treatment of 9 ds m<sup>-1</sup> gave the lowest average for leaf area. Where it

reached 218 cm<sup>2</sup>. As for the nanoNPK, no significant differences appeared. The treatment with a concentration of 150 mg L<sup>-1</sup> gave the highest rate of 646 cm<sup>2</sup> and the concentration of 75 gave the lowest average leaf area of 609 cm<sup>2</sup>. While the spraying of Proline had a significant effect on the leaf area, where the concentration of 0 mg L<sup>-1</sup> gave the highest average of 686 cm<sup>2</sup>. While the concentration of 300 mg.L<sup>-1</sup> gave the lowest average of 577.25 cm<sup>2</sup>. As for the interaction between the salinity levels of irrigation water and the nano-NPK fertilizer, it showed significant differences. While the interaction treatment (9 dSm<sup>-1</sup> + nano NPK at a concentration of 0 mg L<sup>-1</sup>) had the lowest average of 191 cm<sup>2</sup>. The interaction between salinity levels and proline showed significant differences, where the interaction treatment (1.3 dS m<sup>-1</sup> + proline at a concentration of 0 mg L<sup>-1</sup>) was excelled, giving the highest average of 1081 cm<sup>2</sup>, while the interaction treatment gave (9 dS m<sup>-1</sup> + proline at a concentration of 300 mg L<sup>-1</sup>) The lowest rate was 188 cm<sup>2</sup>. The results of the interaction between proline and nano-NPK showed significant differences, where the interaction treatment (NPK at a concentration of 150 mg L<sup>-1</sup> + proline at a concentration of 300 mg L<sup>-1</sup>) gave the highest rate of 715 cm<sup>2</sup>. While the interaction treatment (NPK at a concentration of 0 mg L<sup>-1</sup> + Proline at a concentration of 300 mg L<sup>-1</sup>) the lowest average was 575 cm<sup>2</sup>. The results of the triple interaction indicated that there were significant differences, as the interaction treatment (1.3 dSm<sup>-1</sup> + NPK nanoparticles at a concentration of 150 mg L<sup>-1</sup> + Proline 0 mg L<sup>-1</sup>) gave the highest average of 1196 cm<sup>2</sup>. Whereas, the interaction treatment (9 dSm<sup>-1</sup> + NPK at a concentration of 150 mg L<sup>-1</sup> + Proline 300 mg L<sup>-1</sup>) gave the lowest average leaf area of 161 cm<sup>2</sup>.

**Table (1) Effect of Nano NPK application and spraying of Proline under saline stress on leaf area cm<sup>2</sup> of pomegranate seedling**

	Proline mg L <sup>-1</sup>		NPK nano mg.L <sup>-1</sup>	salinity levels (ds.m <sup>-1</sup> )
N×S	P2(300)	P1 (0)		
1005	924	1086	N1 (0)	S1 1.3 (dS.m <sup>-1</sup> )
1018	1074	962	N2 (75)	
1123	1050	1196	N3 (150)	
842	784	900	N1 (0)	S2 3 (dS.m <sup>-1</sup> )
838	720	956	N2 (75)	
844	794	895	N3 (150)	
547	391	704	N1 (0)	S3 6 (dS.m <sup>-1</sup> )
329	332	325	N2 (75)	
381	296	465	N3 (150)	
191	212	170	N1 (0)	S4 9 (dS.m <sup>-1</sup> )
250	189	310	N2 (75)	
212	161	262	N3 (150)	
632	577.25	686	average P	
S×N	P	S×N×P	L.S.D <sub>(0.05)</sub>	
157.5	64.3	222.8		
S average	Salinity and Proline levels interaction			
1048	1016	1081	S1	S × P
842	766	917	S2	
419	340	498	S3	
218	188	248	S4	
91.0	128.6			L.S.D <sub>(0.05)</sub>
N average	interaction of nano NPK and proline			
640	575	705	N1	N×P
609	579	638	N2	
646	715	578	N3	
78.8	111.4			L.S.D <sub>(0.05)</sub>

The results of Table (2) show that salinity levels have no significant effect on the chlorophyll content of the leaves, where the salinity treatment with a concentration of 3 dSm<sup>-1</sup> gave the highest rate of SPAD 30.58, while the salinity treatment with a concentration of 9 dSm<sup>-1</sup> gave the lowest chlorophyll content of SPAD 26.51. As for the NPK nano, there were no significant differences, as the concentration of 150 mg L<sup>-1</sup> gave the highest chlorophyll content of 29.75 SPAD, while the concentration of 0 mg L<sup>-1</sup> gave the lowest average chlorophyll content in the leaves was 27.22 SPADs for the proline

spraying, it did not show any significant differences. The treatment without the proline spray gave the lowest average chlorophyll content of 27.61 SPAD. While spraying treatment with a concentration of 300 mg L<sup>-1</sup> gave the highest average of chlorophyll content of 29.01 SPAD. As for the effect of the interaction between salinity levels and nanoscale NPK concentrations, it showed significant differences. The interaction treatment (3 dSm<sup>-1</sup> + NPK at a concentration of 75 mg L<sup>-1</sup>) was excelled and gave the highest average of chlorophyll

content of SPAD 33.08. While the interaction treatment ( $9 \text{ dSm}^{-1}$  + NPK at a concentration of  $150 \text{ mg L}^{-1}$ ) gave the lowest average of 23.85 SPAD. As for the interaction between salinity levels and proline, it showed significant differences in the content of chlorophyll, where the interaction treatment ( $3 \text{ dsm}^{-1}$  + proline at a concentration of  $300 \text{ mg L}^{-1}$ ) gave the highest concentration of chlorophyll that reached 32.44 SPAD, Whereas, the interaction treatment ( $9 \text{ dSm}^{-1}$  + proline at a concentration of  $0 \text{ mg L}^{-1}$ ) gave the lowest average of 25.16 SPAD. The interaction between nano-NPK and proline did not give significant differences in the chlorophyll content. The interaction treatment (NPK at a concentration of  $150 \text{ mg L}^{-1}$  +

proline at a concentration of  $300 \text{ mg L}^{-1}$ ) gave the highest concentration of chlorophyll reaching 31.01 SPAD, While the interaction treatment (NPK at a concentration of  $75 \text{ mg L}^{-1}$  + Proline at a concentration of  $0 \text{ mg L}^{-1}$ ) the lowest average of chlorophyll content in the leaves was 26.38 SPAD. As for the triple interaction, it gave significant differences in the chlorophyll content, where the interaction treatment consisting of ( $3 \text{ dsm}^{-1}$  + at a concentration of  $75 \text{ mg L}^{-1}$  + proline at a concentration of  $300 \text{ mg L}^{-1}$ ) gave the highest chlorophyll content of 38.03 SPAD, While the interaction treatment ( $9 \text{ dSm}^{-1}$  + at a concentration of  $150 \text{ mg L}^{-1}$  + proline at a concentration of  $300 \text{ mg L}^{-1}$ ) gave the lowest average of 22.37 SPAD.

**Table (7) The effect of NPK nanoparticle application and spraying of Proline under saline stress on the chlorophyll content of ( SPAD leaves. Unit ) of pomegranate seedling.**

N×S	Proline mg L <sup>-1</sup>		NPK nano mg.L <sup>-1</sup>	salinity (ds.m <sup>-1</sup> )	levels
	P2(300)	P1 (0)			
32.52	33.60	31.43	N1 (0)	S1 1.3 (dS.m <sup>-1</sup> )	
28.45	26.83	30.07	N2 (75)		
27.47	26.37	28.57	N3 (150)		
27.57	31.17	23.97	N1 (0)	S2 3 (dS.m <sup>-1</sup> )	
33.08	38.03	28.13	N2 (75)		
31.08	28.13	34.03	N3 (150)		
29.42	26.73	32.10	N1 (0)	S3 6 (dS.m <sup>-1</sup> )	
24.15	24.63	23.67	N2 (75)		
26.48	29.07	23.90	N3 (150)		
29.50	32.53	26.47	N1 (0)	S4 9 (dS.m <sup>-1</sup> )	
26.17	28.67	23.67	N2 (75)		
23.85	22.37	25.33	N3 (150)		
28.31	29.01	27.61	average P		
S×N	P	S×N×P	L.S.D <sub>(0.05)</sub>		
8.304	3.390	11.744			
S average	Salinity and Proline levels interaction				
29.48	28.93	30.02	S1	S × P	
30.58	32.44	28.71	S2		
26.68	26.81	26.56	S3		
26.51	27.68	25.16	S4		
4.794	6.780			L.S.D <sub>(0.05)</sub>	
N average	interaction of nano NPK and proline				

27.22	26.48	27.96	N1	N×P
27.96	29.54	26.38	N2	
29.75	31.01	28.49	N3	
4.152	5.872			L.S.D <sub>(0.05)</sub>

The results in Table (3) show that the salinity levels of irrigation water had a significant effect on the percentage of carbohydrates, where the level of 1.3 dS. m<sup>-1</sup> gave the highest percentage of 44.63%, while the level of 9 dS. m<sup>-1</sup> gave the lowest percentage of 31.49%. As for the effect of the nano NPK fertilizer, it showed a significant effect, where the concentration of 150 mg L<sup>-1</sup> gave the highest percentage, reaching 39.10%, While the concentration of 0 mg L<sup>-1</sup> was the lowest, which amounted to 36.66%. As for spraying Proline, it did not show a significant effect on the percentage of carbohydrates, so the concentration of 300 mg L<sup>-1</sup> gave the highest percentage of 38.18%, while concentration of 0 mg L<sup>-1</sup> gave the lowest percentage of 37.69%. As for the effect of the interaction between salinity levels and nano-NPK, it had a significant effect. The interaction treatment (1.3 dSm<sup>-1</sup> + nano-NPK at a concentration of 150 mg L<sup>-1</sup>) gave the highest percentage of 44.82%, while the interaction treatment gave (9 dSm<sup>-1</sup> + nano-NPK at a concentration of 0 mg L<sup>-1</sup>) The lowest percentage is 30.89%. The

results of the interaction between salinity levels and proline spray showed significant differences in the percentage of carbohydrates, Where the interaction treatment (1.3 dS m<sup>-1</sup> + proline at a concentration of 300 mg L<sup>-1</sup>) gave the highest percentage of 45.02%, and the interaction treatment (9 dS m<sup>-1</sup> + proline at a concentration of 300 mg L<sup>-1</sup>) gave the lowest percentage, which was 30.93%. Regarding the interaction between nano-NPK and proline spray, it had a significant effect on the percentage of carbohydrates. While the interaction treatment (Nano-NPK at a concentration of 0 mg L<sup>-1</sup> + Proline at a concentration of 0 mg L<sup>-1</sup>) gave the lowest percentage of 36.58%. As for the triple interaction, the interaction treatment (1.3 dSm<sup>-1</sup> + Nano NPK at a concentration of 0 mg L<sup>-1</sup> + Proline at a concentration of 300 mg L<sup>-1</sup>) gave the highest average of 46.42%, While the interaction treatment (9 dSm<sup>-1</sup> + Nano NPK at a concentration of 0 mg L<sup>-1</sup> + Proline at a concentration of 300 mg L<sup>-1</sup>) gave the lowest percentage of 27.87%.

**Table (3) Effect of NPK nanoparticle application and spraying of Proline under salt stress on the carbohydrate content of leaves of pomegranate seedling.**

N×S	Proline mg L <sup>-1</sup>		NPK nano mg.L <sup>-1</sup>	salinity (ds.m <sup>-1</sup> ) levels
	P2(300)	P1 (0)		
44.77	46.42	43.13	N1 (0)	S1 1.3 (dS.m <sup>-1</sup> )
44.29	43.85	44.74	N2 (75)	
44.82	44.78	44.85	N3 (150)	
38.22	38.17	38.26	N1 (0)	S2 3 (dS.m <sup>-1</sup> )
40.46	41.90	39.02	N2 (75)	
44.38	44.35	44.41	N3 (150)	
32.76	34.52	31.00	N1 (0)	S3 6 (dS.m <sup>-1</sup> )
36.50	36.98	36.03	N2 (75)	
34.59	34.46	34.73	N3 (150)	
30.89	27.87	33.92	N1 (0)	S4 9 (dS.m <sup>-1</sup> )
30.96	31.45	30.47	N2 (75)	

32.62	33.47	31.78	N3 (150)	
37.94	38.18	37.69	average P	
S×N	P	S×N×P	L.S.D <sub>(0.05)</sub>	
3.577	1.460	5.058		
S average	Salinity and Proline levels interaction			
44.63	45.02	44.24	S1	S × P
41.02	41.47	40.56	S2	
34.62	35.32	33.92	S3	
31.49	30.93	32.06	S4	
2.065	2.920			L.S.D <sub>(0.05)</sub>
N average	interaction of nano NPK and proline			
36.66	36.75	36.58	N1	N×P
38.05	38.54	37.56	N2	
39.10	39.26	38.94	N3	
1.788	2.529			L.S.D <sub>(0.05)</sub>

The data in Table (4) indicate that there are significant differences in average the levels of salinity of irrigation water in the percentage of protein, where the treatment 3 dSm<sup>-1</sup> gave the highest percentage of protein amounted to 7.468, while the level of 6 dSm<sup>-1</sup> gave the lowest average of protein amounted to 6.687. While the effect of nano-NPK gave a concentration of 150 mg L<sup>-1</sup> the highest average of protein, reaching 7.876, while a concentration of 75 mg L<sup>-1</sup> gave the lowest protein percentage, which amounted to 6.613. As for spraying the amino acid proline, it did not show a significant effect on the percentage of protein, where the treatment without spraying 0 mg L<sup>-1</sup> gave the highest protein rate of 7.148, while the concentration of 300 mg L<sup>-1</sup> gave the lowest concentration of 6.938. As for the interaction between the salinity levels of nano-NPK irrigation water, the interaction treatment (3 dSm<sup>-1</sup> + nano-NPK at a concentration of 150 mg L<sup>-1</sup>) was superior, giving the highest rate of 8.213, while the interaction treatment (6 dSm<sup>-1</sup> + NPK at a concentration of 75. mg L<sup>-1</sup>) was excelled the lowest average was 6.080. While the results of the interaction between salinity and proline

spray showed that there were significant differences between the treatments, the interaction treatment (3 dSm<sup>-1</sup> + proline 0 mg L<sup>-1</sup>) outperformed by giving the highest protein percentage of 7.572, while the interaction treatment (6 dS m<sup>-1</sup> + proline concentration 300 mg L<sup>-1</sup>) gave the lowest concentration of 6.633. The results of the interaction between nano NPK and proline showed a significant effect between the treatments in the proportion of protein, the interaction treatment (75 mg L<sup>-1</sup> + proline at a concentration of 0 mg L<sup>-1</sup>) was significantly excelled on the interaction treatment, which gave the highest average of 8.163, While the interaction treatment (75 mg L<sup>-1</sup> + Proline at a concentration of 300 mg L<sup>-1</sup>) gave the lowest rate of 5.063. As for the triple interaction treatment, the interaction treatment (1.3 dSm<sup>-1</sup> + NPK at a concentration of 150 proline at a concentration of 300 mg L<sup>-1</sup>) gave the highest protein percentage of 8.477, While the interaction treatment (6 dsm<sup>-1</sup> + NPK at a concentration of 75 mg L<sup>-1</sup> + Proline at a concentration of 300 mg L<sup>-1</sup>) gave the lowest average of 4.307.

**Table (4) Effect of NPK nanoparticle application and spraying of Proline under salt stress on protein percentage of pomegranate seedling.**

	Proline mg L <sup>-1</sup>		NPK nano	salinity	levels
N×S	P2(300)	P1 (0)	mg.L <sup>-1</sup>	(ds.m <sup>-1</sup> )	
6.393	7.540	5.247	N1 (0)	S1 1.3 (dS.m <sup>-1</sup> )	
6.497	4.830	8.163	N2 (75)		
7.955	8.477	7.433	N3 (150)		
7.225	7.953	6.497	N1 (0)	S2 3 (dS.m <sup>-1</sup> )	
6.965	5.663	8.267	N2 (75)		
8.213	8.473	7.953	N3 (150)		
6.130	7.537	4.723	N1 (0)	S3 6 (dS.m <sup>-1</sup> )	
6.080	4.307	7.853	N2 (75)		
7.850	8.057	7.643	N3 (150)		
6.810	7.433	6.187	N1 (0)	S4 9 (dS.m <sup>-1</sup> )	
6.912	5.453	8.370	N2 (75)		
7.485	7.537	7.433	N3 (150)		
7.043	6.938	7.148	average P		
S×N	P	S×N×P	L.S.D <sub>(0.05)</sub>		
0.7258	0.2963	1.0265			
S average	Salinity and Proline levels interaction				
6.948	6.949	6.948	S1	S × P	
7.468	7.363	7.572	S2		
6.687	6.633	6.740	S3		
7.069	6.808	7.330	S4		
0.4190	0.5926			L.S.D <sub>(0.05)</sub>	
N average	interaction of nano NPK and proline				
6.640	7.616	5.663	N1	N×P	
6.613	5.063	8.163	N2		
7.069	8.136	7.616	N3		
0.3629	0.5132			L.S.D <sub>(0.05)</sub>	

The data in Table (5) showed that salinity levels had a significant effect on the percentage of dry matter, where the level of salinity 9 dS m<sup>-1</sup> gave the lowest dry matter percentage in the leaves amounted to 34.91%, while the level of 3 ds m<sup>-1</sup> gave the highest average amounting to 37.23%. As for the effect of NPK nanoparticles, there were no significant differences between the treatments, where the concentration of 0 mg L<sup>-1</sup> gave the highest average amounting to 36.83%, While the concentration of 015 mg L<sup>-1</sup> gave the lowest percentage of dry matter in the leaves, which was 34.38%. While the spraying of the amino acid proline had no significant effect on

the percentage of dry matter, where the concentration of 0 mg L<sup>-1</sup> gave the highest average of 36.46%, While the concentration of 300 mg L<sup>-1</sup> gave the lowest average of 35.41%. The results of the interaction between salinity levels and nano-NPK showed significant differences in the percentage of dry matter in the leaves. The interaction treatment (3 dsm<sup>-1</sup> + nano-NPK at a concentration of 0 mg L<sup>-1</sup> gave the highest average of 38.57%, Whereas, the interaction treatment (6 dSm<sup>-1</sup> + Nano NPK at a concentration of 150 mg L<sup>-1</sup>) gave the lowest mean of 32.40%. Regarding the interaction between the levels of salinity of irrigation water and



proline, there are significant differences in the percentage of dry matter. The interaction treatment (3 dm-1 + proline at a concentration of 0 mg L-1) excelled, giving the highest average of 39.61%. While the interaction treatment (6 dsm-1 + proline at a concentration of 0 mg L-1) gave the lowest percentage, which was 34.13%. The results of the interaction between nano-NPK and proline showed a significant effect on the percentage of dry matter. The interaction treatment (75 mg L-1 + proline at a concentration of 0 mg L-1) gave the highest percentage of 38.58%,

While the interaction treatment (150 mg L-1 + Proline at a concentration of 0 mg L-1) gave the lowest percentage of 34.18%. As for the triple interaction among the study factors, the interaction treatment (3 dSm-1 + Nano NPK at a concentration of 0 + Proline at a concentration of 0 mg L-1) excelled and gave the highest average of 44.00%, While the interaction treatment (9 dSm-1 + Nano NPK at a concentration of 0 + Proline at a concentration of 0 mg L-1) gave the lowest rate of 30.73%.

**Table (5) Effect of nano NPK application and spraying of Proline under salt stress on the dry matter percentage in leaves of pomegranate seedling.**

N×S	Proline mg L <sup>-1</sup>		NPK nano mg.L <sup>-1</sup>	salinity (ds.m <sup>-1</sup> )	levels
	P2(300)	P1 (0)			
37.87	37.40	38.33	N1 (0)	S1 1.3 (dS.m <sup>-1</sup> )	
37.28	35.97	38.60	N2 (75)		
34.92	34.43	35.40	N3 (150)		
38.57	33.13	44.00	N1 (0)	S2 3 (dS.m <sup>-1</sup> )	
38.22	36.47	39.97	N2 (75)		
34.40	34.93	34.87	N3 (150)		
36.95	40.43	33.47	N1 (0)	S3 6 (dS.m <sup>-1</sup> )	
35.40	32.93	37.87	N2 (75)		
32.40	33.73	31.07	N3 (150)		
33.93	37.13	30.73	N1 (0)	S4 9 (dS.m <sup>-1</sup> )	
35.48	33.07	37.90	N2 (75)		
35.30	35.23	35.37	N3 (150)		
35.93	35.41	36.46	average P		
S×N	P	S×N×P	L.S.D <sub>(0.05)</sub>		
5.745	2.345	8.125			
S average	Salinity and Proline levels interaction				
36.69	35.93	37.44	S1	S × P	
37.23	34.84	39.61	S2		
34.92	35.70	34.13	S3		
34.91	35.14	34.67	S4		
3.317	4.691			L.S.D <sub>(0.05)</sub>	
N average	interaction of nano NPK and proline				
36.83	37.03	36.63	N1	N×P	
36.60	34.61	38.58	N2		
34.38	34.58	34.18	N3		
2.872	4.062			L.S.D <sub>(0.05)</sub>	

## Discussion

Tables (5-1) show that the levels of saline irrigation water had a significant effect on the traits (leaf area, chlorophyll, protein, carbohydrates, percentage of dry matter in leaves). Low concentrations of salinity in irrigation water ( $1.3 \text{ dm}^{-1}$ ) led to a significant increase in the above-mentioned traits, while high concentrations of salinity caused a decrease in all growth indicators mentioned above. Decreased leaf area (Table 1) is due to the negative effect on leaf respiration (9) and high levels of salinity lead to a defect in leaf growth and small size, thus reducing the efficiency of the photosynthesis process. As well as the low amount of water absorbed affected the elongation of the leaf cells and then the decrease in the leaf area. The effect of salinity on chlorophyll in Table (2) is referred to by (4) (3) As the high concentrations of salinity lead to the reduction of chlorophyll because it leads to senescence of the leaves due to the production of growth inhibitors (ABA and ethylene), which accelerate the decomposition of chlorophyll (8). The reason for the decrease may be due to the active radicals ROS, which leads to the disruption of the ionic balance and the reduction of the size of Stroma, which reduces the formation of chlorophyll. The reason for the decrease in carbohydrates in Table (3) at salinity may be due to its inhibitory effect on chlorophyll, which in turn reduces the formation of carbohydrates, and the reason may be due to disturbance of nitrogen metabolism or inhibition of nitrate absorption (12) and this is consistent with (17). The 'Wonderful' pomegranate seedlings had reduced chlorophyll and carbohydrate content when irrigated with salt water. Nano fertilizers are nutrients that are developed using raw materials with nano scale dimensions ranging from 1–100 nm. These nutrients have a high surface area and the ability to retain nutrients, which increases plant efficiency (16). and the excellence in chlorophyll in pomegranate leaves (Table 2) The reason may be due to the fact that nano-fertilizers increase the availability of nutrients for a longer period.

This result is consistent with what was reached (15). The reason for the increase in leaf area in Table (1) as a result of treatment with NPK nano fertilizer is due to the increase in enzymatic activities and the rate of their reactions, which leads to the production of raw materials to increase cell divisions and then increase the leaf area (13). The increase in the percentage of dry matter in the leaves may be due to the effect of nano-NPK (Table 5) due to the large leaf area and the increase in metabolic reactions in the plant, which in turn leads to an increase in the efficiency of the photosynthesis process and the manufacture of carbohydrates that increase growth and accumulate in the vegetative system, which leads to an increase in the substance Dry (7) As for the increase in the percentage of protein (table 4), it may be due to the fact that nitrogen is the main component of organic compounds, including proteins. It is the main component of the amino acids necessary for the formation of proteins, or it may be attributed to the fact that the nano-fertilizer led to the activity of vital processes and thus increased the acids and enzymes responsible for the formation of protein.

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