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Article

Effectiveness of Proline (B), Humic Acid (H), and Salinity (S) on the Vegetative Indicators and Chemical Properties of Wormwood Plants

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Abstract: This study was conducted during the 2023 summer season In the wooden pergola belonging to the Department of Horticulture and Landscape Engineering - College of Agriculture - Tikrit University The aim was to study the effect of proline, humic acid, and salinity on the vegetative indicators and chemical properties of the wormwood plant. The results of this work accorded that spraying proline at a concentration of 200 mg/L significantly affected all the studied vegetative and chemical characteristics in this experiment, except for the percentage of dry matter. The addition of humic acid had a positive role in most of the traits studied in this study, as the 3 g L-1 level significantly increased plant height, number of leaves, nitrogen, phosphorus, potassium and carbohydrates. The effectiveness of sodium chloride (S) had a significant effect on wormwood plants, exceeding the 10 g/L level in nitrogen, phosphorus, potassium, and carbohydrates. The interfere between (B) treatment(200 mg L^{-1}) and (S) treatment (10 g L^{-1}) significantly affected all chemical properties except phosphorus. vegetative properties not affected by interfere between (B) treatment and (S) treatment. The interfere between (B) treatment (200 mg L^{-1}) and the control treatment also had a significant effect.

Keywords: Wormwood, Proline, Humic Acid, Salinity, Vegetative Growth

Introduction

Wormwood *Artemisia herba-_alba* From the Asteraceae family of plants. Studies have indicated the medicinal importance of the plant due to its content of many active substances and compounds, such as volatile oils, alkaloids, glucosides, saponins, and tannins [1].

The plant is used in folk medicine to treat fever, as a stimulant for the gastric glands, as an antiseptic, and to treat respiratory and digestive disorders [2].

Salinity is one of the most significant challenges facing agricultural production, and leads to a decrease in the productivity of plant species [3]. These are abiotic factors that negatively affect plants worldwide [4].

Salinity significantly impairs plant growth due to the increased concentration of sodium chloride and the resulting osmotic pressure in the early stages of plant life. This is primarily caused by high concentrations of ions in the soil, making it difficult for the plant to absorb water from the saline environment [5].

Some substances with growth-stimulating effects have been used, directly or indirectly, to improve plant growth and increase their ability to withstand and resist environmental factors that cause stress or strain on those plants [6].

Amino acids are very important materials that contribute to the structure of plant organs and to growth. They are involved in the synthesis of proteins and other organic compounds such as alkaloids, pigments, and enzymes. They also stimulate cell growth and act as an acid buffer in plant cells due to their content of basic and acidic groups. Furthermore, they play a crucial role in protecting cells from excess ammonia, which can cause toxicity [7].

Despite the efficiency of mineral fertilizers, their wide range of uses, and their importance in increasing agricultural production and improving plant growth, their excessive application without adhering to approved fertilizer recommendations causes environmental pollution and Plant diseases, animals, and humans, in addition to being economically costly [8].

Using organic fertilizers is a suitable alternative to reduce chemical pollution problems.

One of the most important of these substances is humic acid, as it enhances soils poor in organic matter, improves their physical and chemical properties, contributes to soil aeration and water retention, and provides nutrients to plants [9], [10], [11].

The present study aimed to evaluate the effect of proline, humic acid, and salinity on the vegetative indicators and Chemical properties of the Wormwood plant

Materials and Methods

This study was conducted during 2023 at the wooden pergola belonging to the Department of Horticulture and Landscape Engineering/College of Agriculture - Tikrit University to study the effect of proline, humic acid, and salinity on the vegetative indicators and Chemical characteristics of Wormwood plants.

Preparing seedlings and planting

The seedlings were prepared from a nursery in Baghdad Governorate on 2/28/2023, and the experiment began on the first day of March (knowing that the plants were one year and two months old). The seedlings were transferred to plastic pots with a diameter of 22 cm and a volume of 9.5 liters containing soil and peat moss in a ratio of 1:2.

Treatments

The first factor :

The first factor included spraying with the amino acid proline (B) in the following proportions

- Control without addition (B0)
- 100 mg L-1 (B1)
- 200 mg L-1 (B2)

The second factor :

Is the addition of humic acid (H) at two levels.

- Control without addition (H0)
- 3 g L-1 (H1)

The third factor :

Adding sodium chloride salt, symbolized by (S), at the following levels.

- Control without addition (S0)
- 5 g L-1 (S1)
- 10 g L-1 (S2)

Statistical analysis

The treatments were arranged in a (RCBD) design. Using a factorial experiment system within the schizont, proline acid was placed in the primary sections, humic acid in the secondary sections, and a salinity factor in the sub-secondary sections [12].

Study characteristics

Vegetative growth characteristics :

1. Plant height (cm): - Measurements were taken using a measuring tape from the soil surface of the pots to the top of the plant.
2. Number of leaves:- The number of plant leaves was counted at the end of the season for three plants in each experimental unit, and their average was taken.
3. Dry weight of the vegetative parts:- The stems and leaves were washed to remove any dirt and dried in an electric oven at a certain temperature, 65-70 c For 72 hours until the weight stabilized. It was weighed using a sensitive scale.

Chemical properties of the plant :

1. Percentage of nitrogen in leaves:- The percentage of nitrogen in the digested samples was measured using a distillation apparatus (Microkjeldhal) [13].
2. Estimating the percentage of phosphorus in leaves:- The percentage of phosphorus in digested leaf plant samples was estimated using the colorimetric method with a spectrophotometer (Libra-Bichron 2005 UK-S22). To measure the visible density of phosphorus at a wavelength of 620 nanometers [14].
3. Estimating the percentage of potassium in the leaves:- The percentage of potassium in the digested samples was estimated according to the method Using a device Atomic Absorption spectrophotometer [15].
4. Estimating the percentage of total carbohydrates in the leaves:- The total carbohydrate content in the leaves was estimated, and the readings were recorded using a device, a Spectrophotometer, at a wavelength of 490 nanometers [16].

Results

Table 1. The effect of salinity, proline, and humic acids and their interactions on the height of wormwood plants.

B X H	Salinity			Humic	Proline
	S_0	S_1	S_2		
29.223d	44.216a	22.013i	21.440i	$_0$humic	proline$_0$
32.806c	44.663a	25.796h	27.960g	humic$_1$	
28.812d	33.013e	28.440g	24.983h	humic$_0$	Proline$_1$
35.281b	42.546b	35.533d	27.763g	hhumic$_1$	
35.353b	41.096b	34.540d	30.423f	humic$_0$	proline $_2$
41.430a	45.430a	39.426c	39.433c	humic$_1$	
Average Proline					
31.015c	24.700g	23.905g	44.440a	proline$_0$	B X S
32.046b	26.373f	31.986e	37.780c	proline$_1$	
38.391a	34.928d	36.983 c	43.263b	proline$_2$	
Average Humic					
31.129b	25.615f	28.331e	39.442b	H_0	S X H
36.505a	31.718d	33.585c	44.213a	H_1	
	28.667c	30.958b	41.827a	average S	

Means with the same letter are not significantly different

Results in Table 1. Showed Significant differences were found due to the factors under study, with the treatment being superior B2 The best value was given to the plant height attribute (38.391 cm) compared with B0 treatment (31.015 cm), As for humic acid, the H1 treatment was significantly superior and gave a value of (36.505 cm) compared with H0 treatment (31.129 cm), As for the effect of salt stress, treatment S0 was superior and recorded 41.827 cm compared to treatment S2 (28.677 cm).

The dual interference treatment between proline and salt stress (B0S0) was significantly superior, giving a plant height of 44.440 cm compared to the B0S2 treatment, which gave 24.700 cm. As for the dual interaction between proline and humic acid, the H1B2 treatment was superior, with a plant height of 41.430 cm compared to the B1H0 treatment, which gave a plant height of 28.812 cm. As for the dual interference between salt stress and humic acid, the H1S0 treatment was significantly superior, with a plant height of 44.213 cm compared to the H0S2 treatment (25.615 cm).

The table shows that the triple interference treatment B2H1S0 was significantly superior and gave the highest plant height of 45.430 compared to the treatment B0H0S2, which recorded 21.013 cm.

Table 2. The effect of salinity, proline, and humic acids and their interactions on the number of leaves of the wormwood plant.

B X H	Salinity			Humic	Proline
	S_0	S_1	S_2		
1075.270f	1154.960g	1090.070k	980.780o	$_0$humic	proline$_0$
1239.575b	1453.413c	1154.893h	1110.420i	humic$_1$	
1087.481e	1409.526d	1005.363m	847.553r	humic$_0$	Proline$_1$
1173.317c	1537.926b	1092.003j	890.023q	hhumic$_1$	
1091.156d	1367.006e	1000.016n	906.446p	humic$_0$	proline $_2$
1361.803a	1625.056a	1075.426l	1249.926f	humic$_1$	
Average Proline					
1157.422b	1045.600g	1122.481d	1304.186c	B_0	B X S
1130.339c	869.788i	1048.683f	1473.726b	B_1	
1203.980a	1078.186e	1037.721h	1496.031a	B_2	
Average Humic					
1084.635b	911.593f	1031.816e	1310.497b	H_0	S X H
1243.231a	1083.456d	1107.441c	1538.798a	H_1	
	997.858c	1069.628b	1424.648a	average S	

Means with the same letter are not significantly different

As shown in Table 2. The treatment with proline gave the best value for the number of leaves trait, as the best results were recorded in treatment B2 (1203.980 leaves per plant) compared to treatment B1 (1130.732 leaves per plant), Regarding the effect of humic acid, the H1 treatment was superior and gave 1243.545 plant leaves compared to the H0 treatment (1084.635 plant leaves), while the effect of the S0 treatment was superior with 1424.648 plant leaves compared to the S2 treatment with 997.858 plant leaves.

As for the bilateral interfere between proline acid and salinity, the B2S0 treatment showed a significant superiority in the number of leaves, recording 1496.031 leaves per plant -1 compared to the B1S2 treatment (869.788 leaves per plant -1), Regarding the binary interfere between proline and humic acid, the H1B2 treatment (1361.803 plant leaves -1) was superior compared to the B0H0 treatment (1075.270 plant leaves -1). In the binary interaction between salinity and humic acid, the H1S0 treatment showed superiority and reached 1538.798 plant leaves -1 compared to the S2H0 treatment (911.593 plant leaves 1). Regarding the triple interference, significant differences were recorded between the treatments, as the B2H1S0 treatment was superior (1625.056 plant leaves 1) compared to the B1H0S2 treatment, which reached 847.553 plant leaves -1.

Table 3. Effect of salinity, proline, and humic acid and their interactions on the percentage of dry matter of the vegetative parts (%) in wormwood leaves.

B X H	salinity			humic	proline
	S_0	S_1	S_2		
27.390e	23.620o	28.193n	30.356j	$_0$humic	proline$_0$
31.365c	34.410e	30.980h	28.706m	humic$_1$	
37.794a	43.603a	34.176f	35.603c	humic$_0$	Proline$_1$
33.314b	35.473c	36.136b	28.333n	hhumic$_1$	
33.433b	34.740d	30.763i	34.796d	humic$_0$	proline$_2$
30.537d	32.136g	29.913k	29.563l	humic$_1$	
Average Proline					
29.377c	29.531g	29.586g	29.015h	B_0	B X S
35.554a	31.968e	35.156b	39.538a	B_1	
31.985b	32.180d	30.338f	33.438c	B_2	
Average Humic					
32.872a	33.585b	31.044d	33.987a	H_0	S X H
31.738b	28.867e	32.343c	34.006a	H_1	
	31.226c	31.693b	33.997a	average S	

Means with the same letter are not significantly different

Data presented in Table 3. Treatment B1 gave the best value in the percentage of dry matter % of the vegetative body (%), as the best results were recorded in treatment B1 (35.554%) compared to treatment B0, which gave 29.377%. As for the humic effect, treatment H0 was superior and reached 32.872% compared to treatment H1 (31.728%). As for the salinity effect, treatment S0 was superior and recorded 33.997% compared to treatment S2 (31.226%).

In the binary interference between proline and humic acid, treatment B1H0 showed a significant advantage in the percentage of dry matter of the vegetative body, giving 37.794% compared to treatment B0H0 (27.390%). As for the binary interference between proline and salinity, treatment B1S0 was superior, recording 39.538% compared to treatment B0S0 (29.015%). In the binary interference between salinity and humic acid, treatment H1S0 was superior, giving 34.006% compared to treatment H1S2 (28.867%). Likewise, in the triple interference, significant differences were recorded between the treatments, with treatment B1H0S0 being superior (43.603%) compared to treatment B0H0S0, which reached 23.620%.

Table 4. The effect of salinity, proline, and humic acids and their interactions on the nitrogen content (%) of wormwood leaves.

B X H	Salinity			humic	proline
	S_0	S_1	S_2		
1.507d	1.090g	1.373gf	2.060de	$_0$humic	proline$_0$
1.747c	1.250g	1.870e	2.123dce	humic$_1$	
1.433d	1.133g	1.743fe	1.423gf	humic$_0$	Proline$_1$
2.163b	1.430gf	2.053de	3.006b	hhumic$_1$	
2.064b	1.250g	2.426dc	2.516c	humic$_0$	proline$_2$
2.614a	1.913e	2.516c	3.413a	humic$_1$	
Average Proline					
1.627c	2.091dc	1.621e	1.170f	B_0	B X S

1.798b	2.215c	1.898d	1.281f	B_1	
2.339a	2.965a	2.471b	1.581e	B_2	
Humic Average					
1.562b	2.000bc	1.531d	1.157e	H_0	S X H
2.280a	2.847a	2.146b	1.847c	H_1	
	2.423a	1.996b	1.344c	average S	

Means with the same letter are not significantly different

Data in Table 4 showed that Treatment B2 recorded the best results, which amounted to 2.339% compared to Treatment B0 (1.627%). As for the effect of humic acid (H), treatment H1 was superior, as it reached 2.280% compared to treatment H0 (1.562%). As for the effect of salinity (S), treatment S2 was superior, giving 2.423% compared to treatment S0 (1.344%).

In the binary interference between (B) proline and (S) salinity, the S2B2 treatment recorded a significant effect in the nitrogen content %, which reached 2.965%, compared to the B0S0 treatment, which recorded 1.170%. As for the binary interaction between (B) and (H), the H1B2 treatment was superior at 2.614%, compared to the B1H0 treatment (1.433%). As for the binary interference between salinity and humic acid, the H1S2 treatment was superior, as it gave 2.847%, compared to the H0S0 treatment, which reached 1.157%. In the triple interference, significant differences were recorded between the treatments, as the B2H1S2 treatment was superior (3.413%), compared to the B0H0S0 treatment, which recorded 1.090%.

Table 5. Effect of salinity, proline, and humic acids and their interactions on the phosphorus content (%) of wormwood leaves.

B X H	Salinity			Humic	Proline
	S_0	S_1	S_2		
0.46a	0.28a	0.51a	0.58a	$_0$humic	$proline_0$
0.48a	0.54a	0.59a	0.31a	$humic_1$	
0.43a	0.39a	0.52a	0.36a	$humic_0$	$Proline_1$
0.66a	0.61a	0.50a	0.88a	$hhumic_1$	
0.86a	0.34a	0.45a	1.80a	$humic_0$	$proline_2$
0.38a	0.38a	0.16a	0.61a	$humic_1$	
Average Proline					
0.46c	0.44a	0.55a	0.41a	B_0	B X S
0.54b	0.62a	0.51a	0.50a	B_1	
0.62a	1.21a	0.31a	0.36a	B_2	
Average Humic					
0.58a	0.91a	0.50a	0.34a	H_0	S X H
0.51b	0.60a	0.42a	0.51a	H_1	
	0.75a	0.45b	0.42c	average S	

Means with the same letter are not significantly different

As shown in Table 5. Treatment B2 recorded the best results at 0.62% compared to treatment B0 (0.46%). Regarding the effect of (H) humic acid, treatment H0 (0.58%) was superior to treatment H1 (0.51%). As for the effect of (S) salinity, treatment 2S was superior, yielding 0.75% compared to treatment S0 (0.42%).

However, no significant differences were observed between the two- and three-treatment combinations in leaf phosphorus concentration.

Table 6. Effect of salinity, proline, and humic acid and their interactions on the potassium content (%) of wormwood leaves.

B X H	Salinity			Humic	Proline
	S_0	S_1	S_2		
1.464f	1.073q	1.363p	1.956g	$_0$humic	proline$_0$
1.827e	1.683m	1.796l	2.003i	humic$_1$	
1.965d	1.646n	2.283f	1.966g	humic$_0$	Proline$_1$
2.301b	2.710c	2.133g	2.060h	hhumic$_1$	
2.230c	1.426o	2.450d	2.813b	humic$_0$	proline $_2$
2.433a	1.850k	2.356e	3.093a	humic$_1$	
Average Proline					
1.646c	1.980f	1.580h	1.378i	B_0	B X S
2.133b	2.013e	2.208c	2.178d	B_1	
2.331a	2.953a	2.403b	1.638g	B_2	
Average Humic					
1.886b	2.245b	2.032e	1.382f	H_0	S X H
2.187a	2.385a	2.095c	2.081d	H_1	
	2.315a	2.063b	1.731c	average S	

Means with the same letter are not significantly different

Table 6. Shows the results, treatment B2 recorded the best results at 2.331% compared to treatment B0 (1.646%). As for the effect of humic acid, it was significantly superior with treatment H1 (2.187%) compared to treatment H0, which gave 1.886%. As for the effect of salinity, it was significantly superior with treatment S2 (2.315%) compared to treatment S0, which gave 1.731%.

In the binary interaction between proline and humic acid, the H1 B2 treatment showed a significant advantage, recording 2.433%, compared to the B0H0 treatment (1.464%). As for the binary interaction between proline and salinity, the B2S2 treatment was superior, recording 2.953%, compared to the B0S0 treatment, which gave 1.378%. As for the effect of the binary interaction between salinity and humic acid, the H1S2 treatment was superior, reaching 2.385%, compared to the H0S0 treatment (1.382%). In the triple interaction, significant differences were recorded between the treatments, with the B2H1S2 treatment being superior, giving 3.093%, compared to the B0H0S0 treatment (1.073%).

Table 7. Effect of salinity, proline, and humic acids and their interactions on the carbohydrate content (%) of wormwood leaves.

B X H	Salinity			Humic	Proline
	S_0	S_1	S_2		
6.551f	5.423o	7.010n	7.220m	$_0$humic	proline$_0$
8.694d	7.773l	8.526g	9.783g	humic$_1$	
8.562e	8.053k	8.866i	8.766i	humic$_0$	Proline$_1$
10.067c	10.063f	9.116h	11.023d	hhumic$_1$	
10.754b	9.120h	10.570e	12.573c	humic$_0$	proline $_2$
13.221a	11.080d	14.713a	13.870b	humic$_1$	
Average Proline					
7.622c	8.501f	7.768g	6.598h	B_0	B X S

9.314b	9.895d	8.991e	9.058e	B_1	
11.987a	13.221a	12.641b	10.100c	B_2	
Average Humic					
8.622b	9.520d	8.815e	7.532f	H_0	S X H
10.660a	11.558a	10.785b	9.638c	H_1	
	10.539a	9.800b	8.583c	average S	

Means with the same letter are not significantly different

The results are shown in Table 7. Treatment B2 recorded the best results for the percentage of carbohydrates (11.987%) compared to treatment B0, which reached 6.598%. As for the humic effect, treatment H1 was superior and recorded 10.660% compared to treatment H0, which reached 8.622%. As for the salinity effect, treatment S2 was superior, reaching 10.539% compared to treatment S0 (8.583%). As for the binary interaction between proline and humic, treatment H1 B2 was superior, reaching 13.221% compared to treatment B0H0, which gave 6.551%.

Regarding the effect of the binary interaction between proline and salinity, the S2 B2 interaction treatment gave the highest concentration (13.221) compared to the S0 B0 treatment, which gave the lowest concentration (6.598). As for the binary interaction between salinity and humic acid, the H1S2 treatment was superior (11.558%) compared to the H0S0 treatment, which reached 7.632%. In the triple interference treatment, we note that significant differences were recorded between the treatments, as the B2H1S1 treatment was superior, reaching 14.713% compared to the B0H0S0 treatment, which reached 5.423%.

Discussions

Salt stress significantly affected the rate of most of the vegetative growth characteristics under study. This may be attributed to the effect of salinity on physiological activity, particularly photosynthesis, which is the main reason for reduced plant growth [17]. This may be due to a negative relationship between photosynthetic activity and the sodium and chloride content in the leaves, as salinity reduces chlorophyll content. This reduction in photosynthesis depends on the intensity of salinity and the plant's tolerance [18]. Several studies on the effect of salinity on chlorophyll content have shown that salts affect chloroplast membranes.

Salinity also affects plant growth by causing morphological changes in the plant, represented by a reduction in the vegetative mass [19], [20]. This is evident through a decrease in plant height, number of branches, and number of leaves, which is attributed to the cumulative effect of salts during this experiment. Salinity inhibits plant growth through four main mechanisms: osmotic pressure, specific ion toxicity, oxidation, and hormonal imbalance [21]. Salinity alters the water and ionic balance of tissues at the leaf level, and this phenomenon is associated with a decrease in turgor pressure (swelling) [22]. The decrease in water potential exchange between the plant and the environment is also a contributing factor [23]. Salinity also leads to an increased accumulation of mineral ions such as Cl- and Na+ in the tissues at toxic concentrations (ionic stress) [24], [25].

Likewise, the decrease in the percentage of dry matter of the vegetative system when the concentration of salts (NaCl) increases may be attributed to the water potential in the root system's growth medium, which reduces the plant's ability to absorb water and also affects the absorption of nutrients by plant cells, thus reducing the fresh and dry weight of the vegetative system [26]. On the other hand, salinity has a biological effect, as it inhibits growth hormones such as gibberellins and cytokinins, in conjunction with the activation of growth inhibitors such as abscisic acid, which is reflected negatively on plant growth in general [27].

Spray treatment with proline at a concentration of 200 mg.L-1 reduced the negative effects of salinity, and proline helps to enhance plant defenses against salt stress by improving absorption and nutritional balance [28].

Proline helps the plant adapt to environmental stress because it is a means of regulating osmotic potential by reducing the water potential of leaf cells, causing water to enter them [29].

Emphasized the role of proline as an osmotic buffer, and many researchers have indicated that the accumulation of these compounds is important as a source of energy for overcoming stress and returning to normal [30]. Furthermore, the effectiveness of foliar spraying with proline depends on its optimal concentration, the plant species, and the growth stage at which the spraying takes place. The response of plants exposed to proline spraying may be attributed to the fact that salinity causes proline to accumulate in the cytoplasm at high concentrations, thus reducing the water potential of the cytoplasm. This balances the low water potential of the vacuole sap, which causes the accumulation of salinity-causing ions, thereby maintaining plant growth under high saline conditions. This explains the positive results observed in most vegetative growth characteristics of plants sprayed with proline.

We also observe the positive effectiveness of addition humic acid on most of the vegetative growth characteristics under study, as humic acid improves soil fertility and increases the availability of nutrients, in addition to reducing the negative effects of salinity, thus increasing plant growth and productivity [31]. Furthermore, humic acid binds rapidly and enters root tissue cells within a short time (a few hours).

Using radioactive carbon, indicated that humic acid binds to root tissue cells and becomes soluble within 3-18 hours, and consequently plays a role in increasing root system growth and activity [32].

We observe that the values of nitrogen, potassium, and carbohydrates, respectively, were unaffected by salinity during the first season. This may be attributed to the resistance of the wormwood plant to the salinity concentrations under study.

This may also be due to the essential role of chlorine in photosynthesis, particularly in the light-dependent reactions, where it acts as a cofactor in splitting water molecules. This resistance positively impacted the concentrations of the aforementioned traits. Furthermore, the plant's needle-like leaves, containing small leaflets, contribute to its resistance to environmental stresses.

The lack of effect of these traits, and some vegetative growth characteristics, on salinity concentrations may also be attributed to the plant's high proline content, which regulates intracellular osmosis [33].

(Muralitharan et al. 1990) found that using radioactive Na ions and monitoring their movement within the cell revealed that sodium ions accumulate more within the vacuole than in the cytoplasm or cell wall, thus maintaining the cell's osmotic balance.

In the second experiment, we observed that the plant was clearly affected by salinity and most of the studied traits, especially with increasing Na concentration. This may be due to the increased salt concentration causing a decrease in chlorophyll.

Conclusion

This study assessed the effects of proline, humic acid, and salinity on the vegetative and chemical properties of wormwood plants, focusing on key growth characteristics such as plant height, leaf number, and dry matter percentage, as well as the content of nitrogen, phosphorus, potassium, and carbohydrates. The results demonstrated that proline, particularly at a concentration of 200 mg/L, had a significant positive impact on most plant traits, improving both vegetative and chemical properties. Humic acid at a concentration of 3 g/L also contributed positively to growth, especially in terms of plant height and nutrient content.

Salinity, however, was detrimental to plant growth, as expected, with higher concentrations of sodium chloride (S2) resulting in decreased vegetative growth and nutrient content. The interaction between proline and salinity, particularly at lower concentrations of salinity (S0), showed potential in mitigating the negative effects of salt stress, enhancing plant height and leaf number. The combined effect of proline and humic acid also resulted in superior growth compared to individual treatments, underscoring the synergistic potential of these substances in improving plant resilience under adverse conditions.

While the study demonstrated the benefits of proline and humic acid, it also highlighted the need for further investigation into the optimal concentrations and timing of application for maximizing plant growth and stress tolerance. The limitations of this study include the short duration of the experiment and the limited scope of salinity levels tested. Future research could explore a wider range of salinity conditions, longer experimental periods, and additional plant species to develop a more comprehensive understanding of how these treatments interact and affect plant health.

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