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IMPACT OF ZN-LYSINE CHELATION FOLIAR APPLICATION IN WHEAT PLANTS UNDER DROUGHT STRESS

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ABSTRACT

Drought stress poses a substantial threat to global wheat production, resulting in diminished growth, decreased grain yield, and eventual plant wilting. Wheat plants may also experience physiological changes due to the lack of water, such as altered stomatal behavior and reduced photosynthesis. The purpose of this study was to examine how foliar application of (Zinc) Zn-Lysine chelate affected wheat plants under drought stress. This study was conducted at the Botanical Garden of GCU in Faisalabad, where seeds from the wheat cultivar Millat-11 were used. To compare regular irrigation with water stress situations, the land was divided into two major plots. The experiment was carried out under controlled conditions, using a randomized complete block design with different levels of drought stress and 5 concentrations of Zn-Lysine chelate. (Dry, Hydro, 0.25%, 0.50 and 0.75 Zn-Lysine chelation). The relative leaf water content, cell membrane permeability, chlorophyll contents, flavonoid contents, total reducing sugar, hydrogen peroxide, ascorbic acid, and proline content were determined. Drought stress significantly reduced the biomass of wheat cultivar Millat-11, but foliar application treatment of Zn-Lysine chelation mitigated the influence of drought by improving biochemical and physiological characters. The research showed that Zn-Lysine chelation can successfully lessen the negative consequences of drought stress on wheat plants, favorably affecting a number of morphological, physiological, and biochemical characteristics.

***Keywords*:** Zinc lysine, Wheat, Drought stress, Chelation, Foliar application, Physiological parameters

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1. INTRODUCTION

Pakistan's economy depends on the agriculture sector. This sector accounts for 21.4% of the country's GDP, with 45% of the workers being directly associated with it (Kamal et al. 2019). However, this sector is facing significant challenges in fulfilling the demand for food to sustain Pakistan's growing population. Therefore, there is a need to optimize water usage to ensure the maximum crop yield (Malik 2004; Kamal et al. 2019). Drought significantly influences plant photosynthesis, leading to a decline in yield. Moreover, drought stress results in the reduction of leaf size, limited root propagation, and shoot expansion (Zafar et al. 2023). The water shortage can affect the physiological and biochemical responses of plants which reduces yield. When there's not enough water, plants face challenges like stomata blockage, membrane damage, and enzyme disruption. These factors can reduce the plant's ability to absorb CO2 and produce ATP (Farooq et al. 2009; Zafar et al. 2022).

Plants experience physiological, biochemical, and molecular changes in response to drought. Through these changes, they are better able to build defenses against drought and lessen the damaging effects of water stress. (Arora et al. 2002; Bohnert et al. 2006; Shinozaki and Yamaguchi-Shinozaki 2007; Gholamin et al. 2010). This phenomenon involves sustaining appropriate water levels by strengthening resistance in stomata (Yoo et al. 2009). Plants have developed advanced deep root systems that help them absorb more water, as shown by (Gowda et al. 2011) Additionally, they gather protective substances like glycine, betaine, and proline to cope with water stress, as observed by researchers. To deal with the increased stress, plants employ both enzymatic (involving enzymes) and non-enzymatic antioxidant mechanisms (Zafar et al. 2021). These mechanisms can help to minimize the harmful effects of oxidative stress caused by water stress. Examples include the use of antioxidative compounds like

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glutathione and ascorbate, as well as the production of enzymes like peroxidases and superoxide dismutase to neutralize reactive oxygen species (Carvalho 2008). Transpiration helps to maintain water under drought stress. This helps them to regulate leaf turgor and manage water loss during periods of water stress (Mott and Franks 2001; Mediavilla and Escudero 2004). When plants face water scarcity, they manage the firmness of their leaves by adjusting the concentration of certain substances in their cells. Proline, sucrose, soluble carbohydrates, glycine betaine, and a variety of other substances are included. These modifications allow the plants to absorb more water from the drying soil (Moghaieb et al. 2004; Carvalho 2005).

Drought stress affects wheat growth, causing significant grain production and value loss, especially in dry areas. Plants experience physiological, biochemical, and molecular changes in response to drought (Zafar et al. 2020). Through these changes, they are better able to build defenses against drought and lessen the damaging effects of water stress (Ashraf and Foolad 2007). Punjab produces 80% of the nation's harvest, according to GOP (2007). The remaining three territories contribute 20% of the wheat. Over the years, wheat has consistently had a low yield. In Pakistan's saline areas, there is an average loss of about 65% in wheat production (Shafi et al. 2006). Micronutrients like boron, nickel, iron, manganese, molybdenum, zinc, and copper are crucial in supporting healthy plant growth. In chemical processes within the plant, these micronutrients act as cofactors and directly impact reactions such as oxidation and reduction. A shortage of micronutrients can damage the wheat crop both qualitatively and quantitatively (Narimani et al. 2010).

Around the globe, there's a lack of zinc in soil. A study presented by the Food and Agriculture Organization covering 30 countries revealed that about 30% of the world's agricultural land is deficient in zinc. Similar deficiencies were found in India, Australia, and China, particularly affecting grains grown in dry and semi-dry areas with calcareous soil (Razzaq et al. 2020). Wheat is notably prone to zinc deficiency, and when grown in zinc-deficient soil, not only does it impact plant growth but also reduces grain production. (Nadim et al. 2012). Zinc is a micronutrient that is required by higher plants, particularly oil crops, for protein synthesis, carbohydrate metabolism, and the action of many enzymes such as DNA, RNA, and dehydrogenase. But zinc is also necessary for fertilization, pollen function, and the synthesis of chlorophyll. A lack of Zn also has an impact on carbohydrate metabolism, pollen organization, and net production. Zn is required for plant biochemical processes such as cytochrome and nucleotide synthesis, and enzyme activation (IRRI 2000). Low soil zinc content, high pH, and high concentrations of magnesium (Mg), sodium (Na), phosphate (PO_4^{3-}), and bicarbonate (HCO_3) are the primary causes of zinc shortage in soil (Khan et al. 2012).

Scarcity of water reduces grain productivity. The lack of water is directly causing the grain to lose yield at varying rates. Abiotic stress affects yield and growth in the dry season. In this regard, the dry condition is the fundamental, limitless abiotic stress that has an impact on development and yield. Although foliar Zn application improved wheat yield and ripe tillers, it had little effect on crop agronomic attributes. An important global test is to increase the Iron (Fe) and Zinc (Zn) concentration of food harvesting plants, resulting in increased yield and improved living well-being. Zinc deficiency is prevalent among micronutrients in both yields and people (Welch and Graham 2004). Micronutrients can be applied as foliar applications through soil implementation. Although foliar application is more effective at increasing yield, the strategy is ineffective for -poor farmers due to its high cost, even though any of these techniques improve micronutrient deficiency. (Nawaz et al. 2013). Objectives of the study were to find out the optimum level of foliar implementation to tolerate drought stress, and to identify the most effective levels of Zn-Lysine to enhance micronutrients in wheat plant.

2. MATERIALS AND METHODS

2.1. Growth Conditions and Experimental Design

This study took place at the Botanical Garden of GCU in Faisalabad, where seeds from the wheat cultivar Millat-11 were used. To compare regular irrigation with water stress situations, the land was divided into two major plots. Each plot has been further divided based on Zn-Lysine treatment into subplots. Within each subplot, there were five columns, with rows spaced approximately 14cm apart. Millat-11 wheat seeds were sown on December 17, 2015, and after 8 days of germination, both plots were generously fertilized with urea and Di-ammonium phosphate (DAP). One plot received regular irrigation as needed, while the other plot was irrigated after 45 days. We applied different levels of Zn-Lysine chelation (Control, Hydro, and Zn-Lysine Chelation: 0.25, 0.50, and 0.75%) to assess their effects. After 30 days of the spray, we collected data on various physiological and biochemical attributes. This information helps us understand the influence of different irrigation and Zn-Lysine treatments on wheat health and growth.

2.2. Analysis of Plant Material

2.2.1. Morphological Characteristics: Morphological Characteristics such as the size of plant roots and shoots were measured using an electronic weight balance. The weight was recorded in grams. In addition, the length of the roots and shoots was measured using a scale and recorded in centimeters.

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2.2.2. Physiological Parameters: To determine the Leaf Relative Water Content (LRWC), the researchers employed the following straightforward equation:

RWC = (Fresh Weight - Dry Weight) / (Turgid Weight - Dry Weight) × 100

***2.2.2.1. Cell Membrane Permeability*:** Took 0.5g of fresh leaves, chopped them up, and put them in small plastic containers with 10mL of distilled water. Checked that the initial electrical conductivity (EC0) was at a baseline. Covered the containers and left them overnight at 40°C. The next day measured EC1. Later on, it measured EC2. To find the electrical conductivity rate, use this formula:

CMP = (EC1 - EC0) / (EC2 - EC0) × 100

2.2.2.2. Chlorophyll and its Contents: The chlorophyll content was ascertained by applying Arnon's (1949) method. Plant leaf segments were cut into small pieces and left overnight at 4°C in 80% acetone. A spectrophotometer was used to measure the absorbance at 570, 645 and 663 nm. Using the following equation, the most recent assimilation of chlorophyll a and chlorophyll b was determined.

For fresh leaf weight, Chlorophyll a (mg g^{-1}) = [12.7 (OD 663) - 2.69 (OD 645)] HV/1000×W
For fresh leaf weight, Chlorophyll b (mg g^{-1}) = [22.9 (OD 645) - 4.68 (OD 663)] HV/1000×W
V = The amount of concentration used (in milliliters).
W = New leaf tissue weight (g)

2.3. Biochemical Characteristics

2.3.1. Total Phenolics: Grind 0.1g of plant material in a mill and pestle at 40°C in order to extract plant components. Ten mL of 80% methanol were combined with the ground material. Subsequently, the mixture was put into an Eppendorf tube and centrifuged for a short while at 10,000rpm. The Folin-Ciocalteu technique was employed in this study to monitor the total phenolics (Wolfe et al. 2003). After removing the supernatant, add 1mL of Folin-Ciocalteu reagent and give it a gentle shake. Then, 2.5mL of 20% sodium carbonate (Na_2CO_3) was added, and distilled water was used to reduce the volume to 5mL. At 750nm, we were able to measure the absorbance.

2.3.2. Total Flavonoid: 0.5g of fresh leaf was crushed in 10mL of methanol (80% methanol) at 40°C using a pestle and mortar. After that, the mixture was centrifuged for a brief period at 10,000rpm. The total flavonoid was calculated using Lee et al. (2016). Following the addition of 0.3mL of AlCl3 and 0.3mL of sodium nitrate ($NaNO_2$) to the plant material supernatant. Then, 0.2mL of NaOH was added. A spectrophotometer set up at 510nm was used to measure the arrangement's absorbance.

2.3.3. Total Soluble Sugar: With the aid of a pestle and mortar, 0.5g weight of the fresh leaf was measured, ground in the phenol sulfuric acid reagent in 10mL of methanol (80% methanol) at 40°C, and centrifugal device is used to estimate the material at 10,000rpm. After the plant material was extracted, 0.2mL of phenol sulfuric acid reagent was added, and the mixture was left for a short while. At that stage, a spectrophotometer was used to check the absorbance at 595.

2.3.4. Determination of Ascorbic Acid (Vitamin C): To determine the amount of ASA, use a pestle and mortar to grind 0.50g of plant material in 5mL of Trichloroacetic Acid (TCA) for five minutes at 4°C and 10,000rpm. The ascorbic acid concentrations were monitored. The test tube was covered with a cotton bud, and the plant material that had been separated was mixed with a small amount of thiourea and 0.5mL of di-nitrophenyl hydrazine. The test tubes must then be heated to 90°C in a water bath. 0.7mL of ice-cooled sulfuric acid (H2SO4) was added once the liquid had cooled. At 530nm, the absorbance was measured.

2.3.5. Calculating Hydrogen Peroxide (H_2O_2): Centrifuge the mixture at 1000rpm after homogenizing the plant material (0.50g) in 10mL of trichloroacetic acid (TCA). Measure the absorbance at 390nm after adding 0.5mL of phosphate buffer solution and 1mL of KI to a 0.5mL supernatant.

2.3.6. Calculation of Total Soluble Protein: Bradford reagent (3mL) should be added after the plant material has been homogenized in a phosphate buffer. Dissolved 0.1g of Coomassie Brilliant Blue dye in 50mL of ethanol to make the reagent, then add 100mL of orthophosphoric acid and thoroughly mix it. The tint turned dark brown after 850 milliliters of hydrogen peroxide (H_2O_2) was added. After going through at least five filters, a faint brown tint

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was produced. Add 3mL of Bradford reagent to 0.1mL of plant sample to determine the protein content. After shaking, it settled down at normal room temperature for 10min. The absorbance quality was measured with the photo spectrometer at 595nm.

2.3.7. Total Free Amino Acid: Fresh 0.5g plant material was ground with a pestle and mortar. Centrifugation caused the supplement to separate. To 1mL of enzyme extract, add 1mL each of 2% ninhydrin and 10% pyridine. Boil the mixture for 15min at 90°C in a water bath. Once the mixture had cooled to room temperature, the distilled water was added to maintain the 50mL volume. Determine the absorbance at 570nm.

2.4. Statistical analysis

The significance of collected data was analyzed by applying Analysis of Variance (ANOVA) with the help of Statistix 8.1.

3. RESULTS AND DISCUSSION

In 2015, at Botany Department, Government College University, Faisalabad we conducted experiments. Subplots representing each lysine-chelated zinc treatment were created from the two main plots. Following that, there are 20 seedlings per row and a 14cm gap between rows. One plot received normal irrigation after the initial irrigation, and the other received normal irrigation after 45 days. the external application of foliar Zn-Lysine chelation at varying concentrations (Control, Hydro: 0.25, 0.5, and 0.75%). Following 30 days of spraying, three replicates of each parameter's data were gathered. Findings for the following parameters: physiological, biochemical, and morphological characteristics of plants. Different conditions elicit different responses in plants. The study's primary goal was to improve plant growth and yields in stressful environments. Better adapted to stressful environments, plants responded well, had higher yields, and had better plant growth.

3.1. The Fresh Weight of Shoots

Drought stress significantly ($P<0.01$) decreased the fresh weight of the shoots. The weight of biomass in the wheat cultivar Millat-11 increased when Zn-Lysine was applied topically during a drought, according to the data values as shown in Fig. 1. The fresh weight of shoots in the Millat-11 cultivar decreased as osmotic stress increased, but under drought stress, shoot fresh weight increased significantly when Zn-Lysine chelation was applied topically at concentrations of 0.25% and 0.75%. For this morphological parameter, statistical data indicate a significant increase (Table 1).

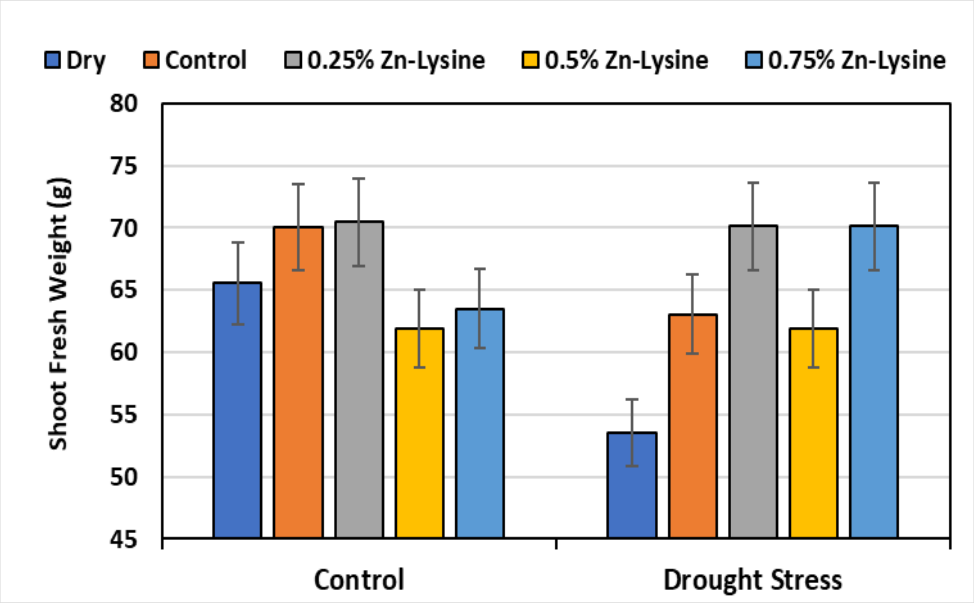

Fig. 1: Impact of applying Zn-Lysine foliar on the fresh weight of wheat shoot (g/plant) during drought stress.

3.2. Dry Weight of Shoot

The data explained the gradual decrease in the dry mass of shoots in wheat (Millat-11) under drought stress. The Millat-11 cultivar's plants are lighter. According to data, control plants of the variety Millat-11 (foliar application of 0.25% Zn-Lysine chelation) exhibited higher shoot dry mass as displayed in Fig. 2. Under drought

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stress, however, the plants treated with 0.5% Zn-Lysine of variety Millat-11 had reduced weight. In comparison to 0.5% and 0.75% Zn-Lysine foliar applications, the Millat-11 control plants treated with level 0.25% Zn-Lysine foliar applications gained more weight. The dry weight of Millat-11 treated with a 0.5% Zn-Lysine foliar application was significantly decreased by drought stress (Table 1). The primary factors and their interactions had a non-significant effect, according to the statistical data for this morphological parameter. Similar results were also observed by Saifullah et al. (2014).

Table 1: ANOVA mean squares for wheat all attributes under drought conditions

SOV	Drought Stress (DS)	Treatment (T)	DS*T	Error
Df	1	4	4	20
SFW	48.90**	109.66**	77.13**	0.7
SDW	13.33**	29.58**	19.10**	0.45
RFW	26.51**	10.64**	2.95**	0.63
RDW	5.55**	2.04**	0.60*	0.16
SL	34.13**	93.58**	50.19**	1.54
RL	17.48**	7.97**	2.22ns	0.83
LA	114.19**	83.77**	39.91*	6.91
RWC	4356.34**	173.11ns	148.93ns	71.18
CM	105.39ns	420.98ns	255.62ns	248.99
CHLa	0.76*	0.02ns	0.95**	0.13
CHLb	0.12ns	0.15ns	0.22ns	0.12
CHLab	9.58**	1.51ns	0.70ns	0.91
FL	38739.9**	7020.5ns	5076.6ns	3945.25
PC	432.5ns	2048.6*	2479.4*	581.96
TRS	18569.4ns	427697.3**	31263.8ns	46878
H_2O_2	107.13**	29.12*	7.07ns	6.35
ASC	29.36*	1.38ns	6.61ns	5.54
AA	7.18ns	0.77ns	10.47ns	10.37
PrC	12.47**	9.19**	0.61ns	1.19

SOV: Source of Variance; Non-significant: NS; Highly significant: **; Significant: *; Fresh Weight of Shoots: SFW; Dry Weight of Shoot: SDW; Fresh Weight of Root: RFW; Dry Weight of Root: DWR; Shoot Length SL; Root Length: RL; Leaf Area: LA; Relative Water Content: RWC; Cell Membrane: CM; Chlorophyl a: CHLa; Chlorophyl b: CHLb; Chlorophyl ab: CHLab; Anthocyanin Contents: AC; Flavonoid Contents: FL; Phenolic Contents: PC; Total Reducing Sugars: TRS; Hydrogen per Oxide: H_2O_2; Ascorbic Acid: ASC; Amino Acid: AA; Proline Contents: PrC.

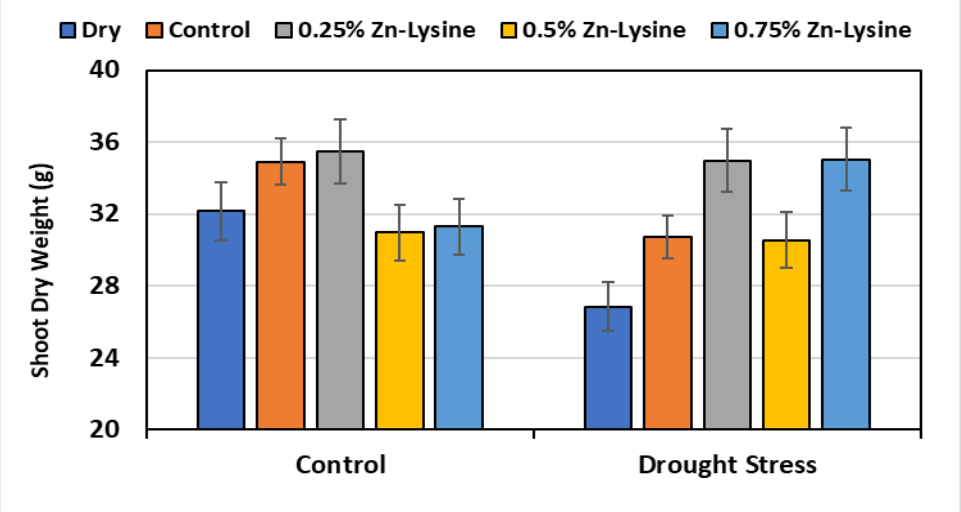

Fig. 2: Impact of foliar ZN-LYSINE application on the dry weight of wheat shoots affected by drought.

3.3. Fresh Weight of Root

The numerical value of the data shows that when Zn-Lysine Chelation was applied topically to drought-stressed roots, the fresh weight of the roots increased (Fig. 3). The plants on Milat-11 were more developed. Millat-11 control plants are given a higher weight than plants that are stressed by drought. Large fresh masses of roots were present in Millat-11 plants that were stressed by drought and received a 0.25% zinc lysine chelation foliar application. Applying a topical solution of 0.25% Zn-Lysine showed good results in Millat-11-controlled plants.

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Application of 0.25% Zn-Lysine foliar solution also enhanced root biomass under drought stress. Based on statistical data, the plant treated with foliar application of Zn-Lysine showed a significant increase in this morphological attribute (Table 1).

3.4. Dry Weight of Root

The experimental value of the data showed that, in drought stress, root dry weight frequently decreased (Millet-11) as shown in Fig. 4. The control plant showed an increase in dry mass, but the plant subjected to drought stress conditions showed low dry mass in all conditions except the plant treated with 0.25% Zn-Lysine foliar application which had comparatively high root dry mass as compared to the other plants of drought stress conditions (Table 1).

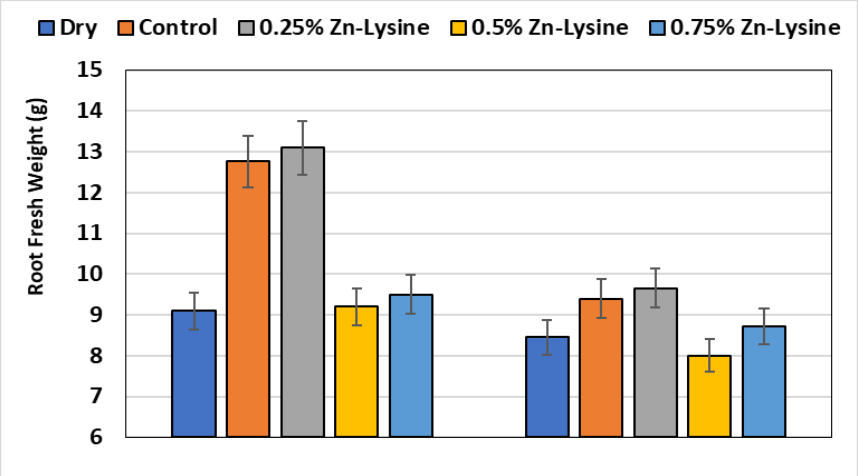

Fig. 3: Impact of foliar Zn-Lysine spray on the fresh weight of roots in wheat plants subjected to drought stress.

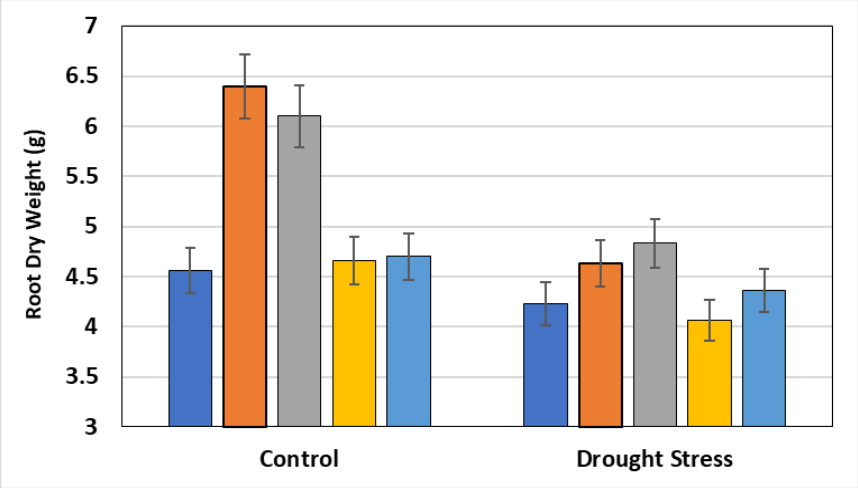

Fig. 4: Impact of foliar Zn-Lysine spray on the root dry weight of drought-stricken wheat plants.

3.5. Length of Shoot

According to the findings, the shoot length of the wheat variety (Millat-11) had a substantial effect on the growth rate under drought stress. Drought stress was reduced in plants treated with Zn-Lysine chelation foliar application. When dry plants were subjected to drought, they had the shortest shoot length (Fig. 5). When compared to control plants, plants treated with 0.75% Zn-Lysine chelation foliar application had increased shoot length in drought conditions. In drought stress conditions, control plants and plants treated with 0.25% Zn-Lysine foliar application have shorter shoot lengths than control plants. The statistical data for this morphological parameter revealed that the main factors had no significant effect, but there was a significant two-way interaction (Table 1). similar trend was also observed by Ali et al. (2022).

3.6. Length of Root

The plant root growth in wheat (Millat-11) reduced the root length when the plant was stressed by drought, according to numerical values about the data (Fig. 6). Under drought stress, root development was found to be reduced in control plants and plants treated with 0.25 and 0.5% Zn-Lysine chelation foliar applications. In

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comparison to drought stress circumstances, Zn-Lysine solution foliar application treatment plants Millat-11 had generally bigger root growth in the control conditions (Table 1).

3.7. Area of Leaf

The Numerical value of data expressed that the leaf area of wheat shows little deviation as shown in Fig. 7. There were non-significant changes observed. In the control condition control plant and 0.25% Zn-Lysine foliar treated plant show increased leaf area as compared to the drought stress condition. Leaf area increase in drought stress condition in plants which were treated with 0.5 and 0.75% Zn-Lysine foliar application as compared to control condition as displayed in Table 1. The results were in accordance with Hussaan et al. (2021) and Ali et al. (2022).

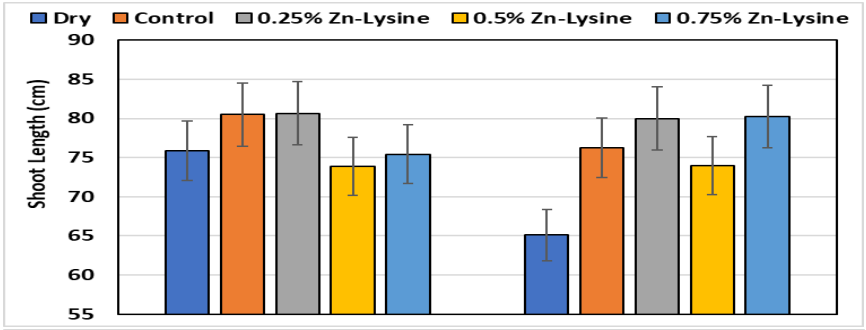

Fig. 5: Effect of foliar Zn-Lysine chelation on wheat shoot length under drought stress.

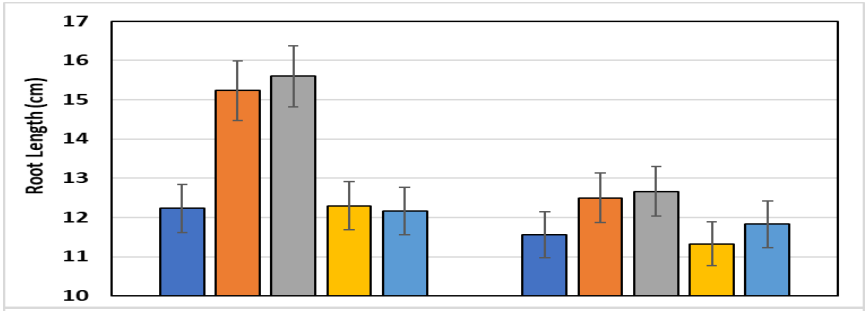

Fig. 6: The effect of foliar Zn-Lysine treatment on wheat root length during drought stress.

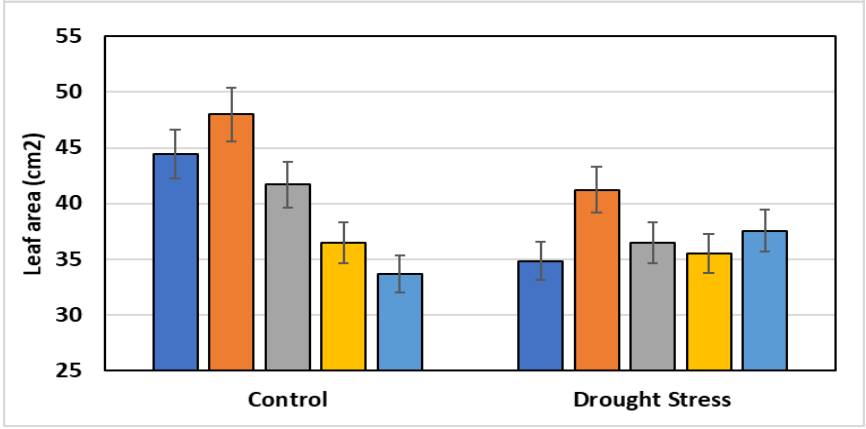

Fig. 7: Impact of applying Zn-Lysine foliar on the leaf area of wheat plants during drought stress.

3.8. Relative Water Contents

The numerical value of the data indicated that, in Millat-11 under drought stress conditions, the relative water contents gradually dropped (Fig. 8). The control plants have higher relative water contents than the drought-stressed plants when exposed to foliar Zn-Lysine chelation. The physiological attribute's statistical data indicated a noteworthy decline during drought stress. A more noticeable drop in relative water content was observed in plants exposed to 0.25 and 0.5% Zn-Lysine chelation foliar application under drought stress compared to plants grown under control conditions (Table 1).

3.9. Permeability of Cell Membrane

The data's numerical value revealed that, in comparison, under drought stress conditions, cell membrane permeability increased (Fig. 9). When Millat-11 dry plant plants were proposed to drought stress, the controlled

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plant and foliar application of 0.75% Zn-Lysine chelation demonstrated increased cell membrane permeability comparatively other controlled conditions. When wheat plants were applied topically with 0.5% Zn-Lysine chelation under control conditions, their cell membrane permeability was higher than when they were under drought stress (Table 1). Similar results were also observed by Saifullah et al. (2014) and Rizwan et al. (2017).

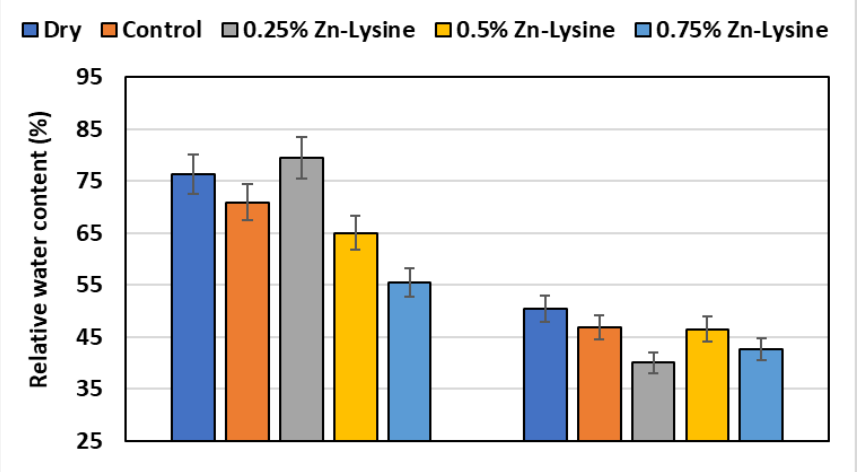

Fig. 8: Effect of ZN-LYSINE foliar application on relative water contents of wheat plants under drought stress.

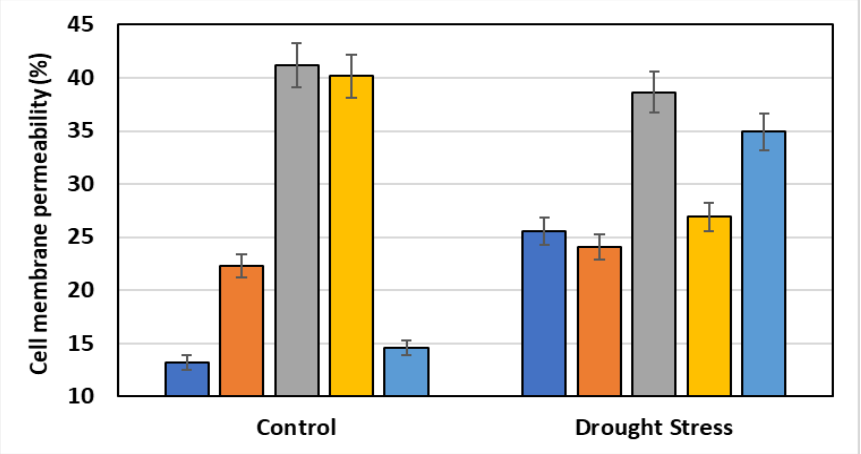

Fig. 9: Effect of ZN-LYSINE foliar application on cell membrane permeability of wheat under drought condition.

3.10. Chlorophyll Contents

A gradual decrease and increase trend were observed in control and drought stress conditions respectively for the chlorophyll content (Fig. 10). However, 0.75% Zn-Lysine foliar spray treatment was observed to be effective in alleviating the drought stress. Accumulation of more chlorophyll contents was observed in dry plants in the normal control condition. While, under drought conditions 0.25, 0.5 and 0.75% Zn-Lysine chelated foliar implicated plants show an increase in chlorophyll content (Table 1).

3.11. Chlorophyll *b* Contents

It was observed from the numerical value of data that chlorophyll *b* contents were greater in controlled plants as compared to drought-stress plants (Fig. 11). In Millat-11 drought stress condition plant exogenesis with 0.75%, Zn-Lysine shows an increase in chlorophyll *b* contents as compared to the control condition plant. In drought stress conditions dry plants, 0.25 and 0.5% Zn-Lysine chelated foliar application plants showed a decrease in chlorophyll *b* content but on the other hand in control conditions, 0.5% and 0.25% Zn-Lysine foliar application plants showed an increase in chlorophyll contents as compared to drought stress plant (Table 1).

3.12. Chlorophyll *ab* Ratio

The Numerical value of data expressed that the *ab* ratio of chlorophyll was higher in drought conditions overall (Fig. 12). The drought stress condition plants treated with foliar application of Zn-Lysine contain a higher ratio than different levels and as compared to the control condition. However, the treated plants of the control condition show no significant rise in the chlorophyll a/b ratio (Table 1).

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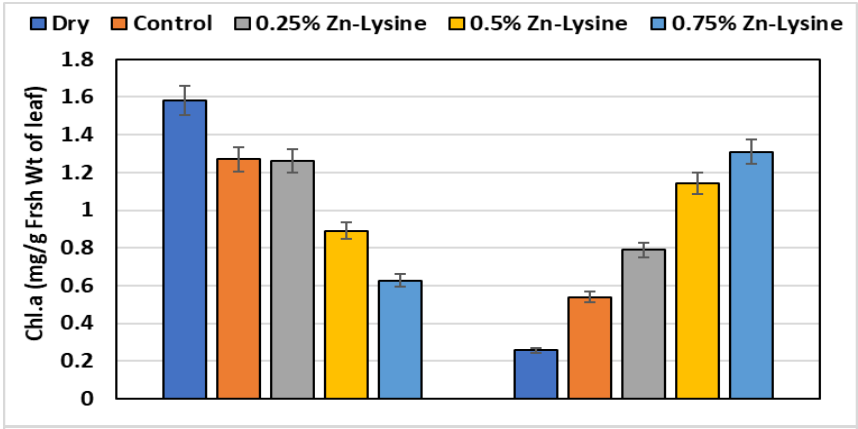

Fig. 10: Influence of Zn-Lysine foliar applications on chlorophyll *a* wheat under drought state.

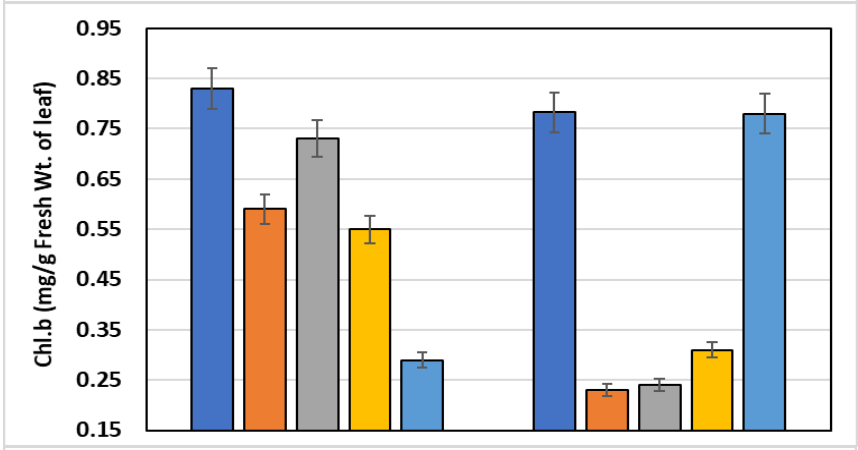

Fig. 11: Impact of Zn-Lysine foliar application on chlorophyll *b* of wheat under drought state.

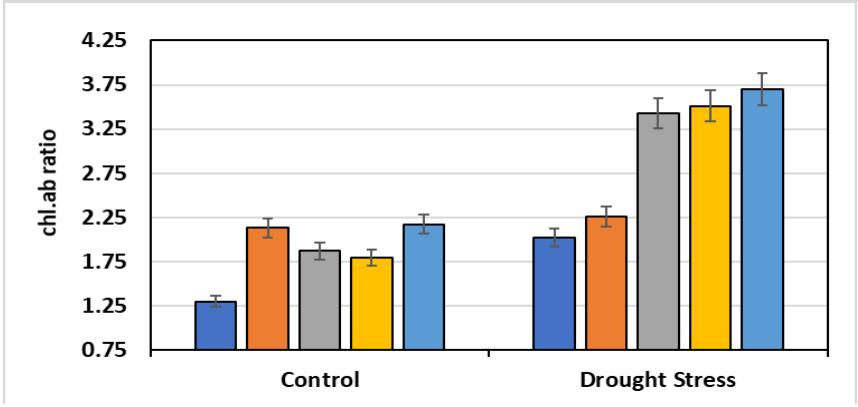

Fig. 12: Impression of Zn-Lysine foliar application on chl. Ab ratio of wheat under drought state.

3.14. Flavonoid Contents

The mathematical value data depicts those flavonoid contents boosted under drought (Fig. 13). It was noticed that the plants have been more developed compared to normal conditions under drought stress (Excessive contents of flavonoid were determined in dry plants under drought stress conditions. However, a huge rise was seen with the increase of Zn-Lysine treatment level (0.25% and 0.5% Zn-Lysine foliar treatment) in drought stress conditions flavonoid contents were increased with an increase in the concentration of Zn-Lysine concentration (Table 1). The plants of Millat-11 showed a good response under drought stress conditions. All treatment levels have increased flavonoid contents to some extent as compared with the control condition.

3.15. Phenolic Contents

Experimental value of data expressed that phenolic contents increased more in dry plants in drought stress conditions (Fig. 14). In the control condition plants of Millat-11 on all treatment levels show an increase in phenolic content as compared to drought conditions. While in drought-stressed conditions plants treated with 0.25 and 0.5% Zn-Lysine foliar application show decrees in phenolic contents as compared to the control-conditioned plant (Table 1).

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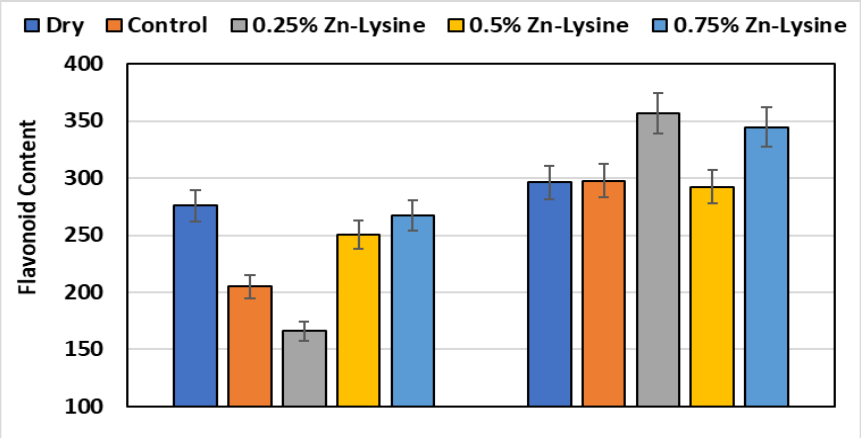

Fig. 13: Impression of Zn-Lysine foliar application on flavonoid of wheat under drought state.

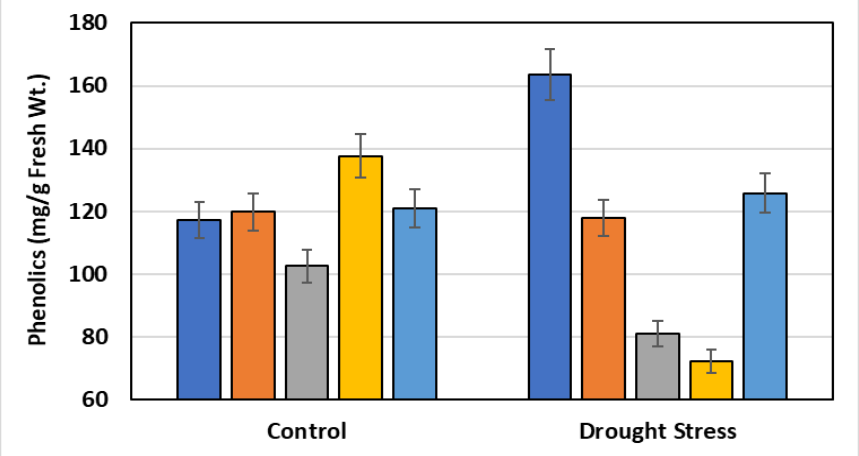

Fig. 14: Impression of Zn-Lysine foliar application on phenolic contents of wheat under drought.

3.16. Total Reducing Sugars

The Accurate value of data represents that the amount of total reducing sugars increased in drought stress conditions (Fig. 15) (control plant and in plants who were treated with 0.75% Zn-Lysine chelate foliar application) in contrast with control condition plants treated with water. However, in treated plants with 0.25 and 0.5% Zn-Lysine foliar application show a little reduced level of total reducing sugars in drought stress plants as compared with the control plant of Millat-11 (Table 1).

3.17. Hydrogen Peroxide Contents

The numerical data revealed that H_2O_2 contents enhanced under drought stress in wheat cultivar (Millat-11) in Fig. 16. However, Zn-Lysine foliar applications alleviate the osmotic effect by reducing its concentration. However, plants of variety Millat-11 when treated with 0.75% solution of Zn-Lysine expressed a large amount of H_2O_2. However, the drought-treated seedling of this same variety had large contents of H_2O_2 when exposed to 0.5% of Zn-Lysine foliar application (Table 1).

3.19. Ascorbic Acid

Numerical value of data showed that in both two conditions drought stress and control conditions of Millat-11 Ascorbic acid contents increased significantly (Fig. 17). In drought conditions plants treated with 0.75 and 0.25% Zn-Lysine foliar application show an increase in Ascorbic acid content as compared to the control condition. Ascorbic acid contents increase in dry plants in drought stress conditions as compared to the control condition plant (Table 1).

3.20. Free Amino Acid

The experimental value expressed that free amino acid contents increased in both control conditions and drought stress conditions (Fig. 18). It was observed that free amino acid contents were higher in drought-stressed conditioned plants as compared to control condition plants. In control conditions plants treated with 0.5% Zn-Lysine foliar application showed an increase in total free amino acid contents in drought stress conditions plants treated with 0.75% dry plant and control plant showed a weighty increase in free amino acid contents (Table 1).

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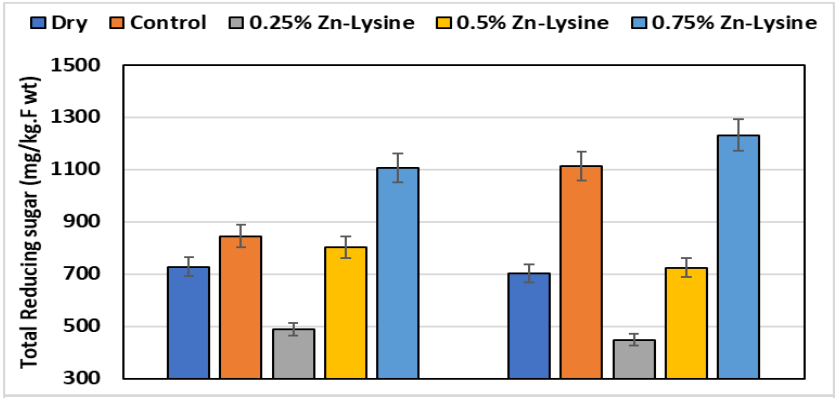

Fig. 15: Impression of Zn-Lysine foliar treatment on total reducing sugars of wheat under drought.

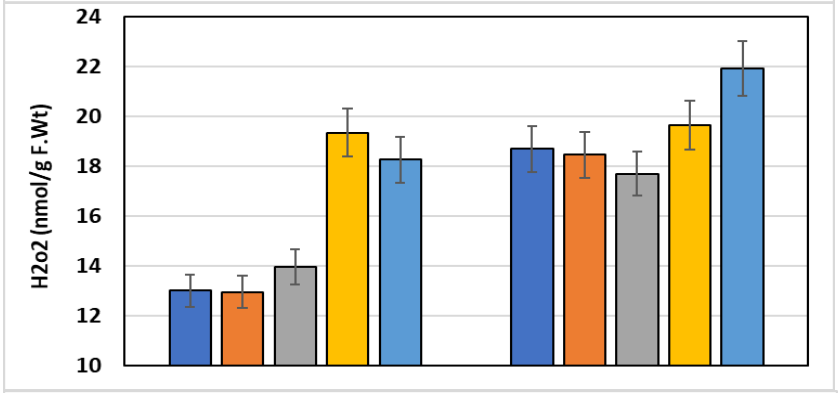

Fig. 16: Impact of Zn-Lysine foliar application on H_2O_2 of the wheat due to drought condition.

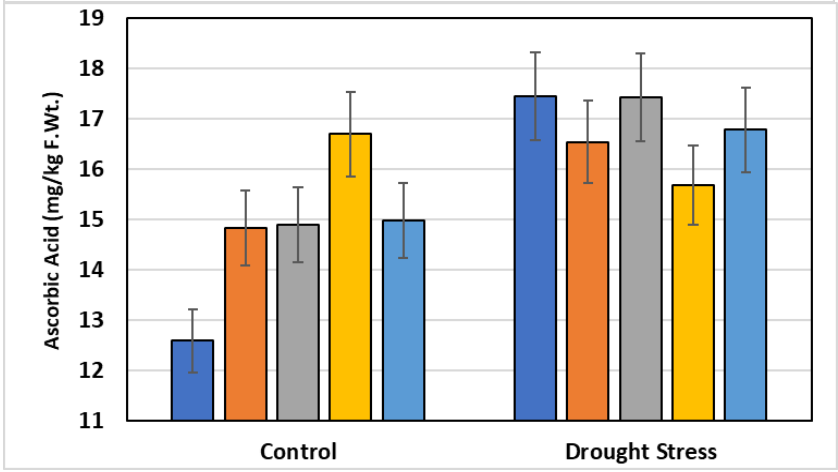

Fig. 17: Impact of Zn-Lysine foliar use on ascorbic acid of wheat during drought.

3.21. Proline Contents

Proline contents enhanced under drought stress conditions plant more significantly as contrast with control condition plant (Fig. 19). In drought stress conditions plants treated with 0.75% Zn-Lysine foliar application showed more increase in Proline contents as compared to the control condition and another treatment level. In the control condition plants treated with 0.75% Zn-Lysine foliar application show an increase in Proline content than other treatment levels but showed less increase in contrast with drought stress condition (Table 1) also stated by Saifullah et al. (2014) and Rizwan et al. (2017).

Drought alters many biochemical and physiological parameters, which in turn affects plant growth and development and ultimately agricultural productivity. Reactive oxygen species (ROS) production, photosynthetic activity, osmotic and hormonal balance, morphological and biochemical changes, and a decrease in the roots' ability to absorb nutrients from the soil are all brought on by drought (Hussaan et al. 2021; Ali et al. 2022). To adapt to these circumstances, plants cause a variety of morphological, biochemical, and physiological modifications. The type and level of stress determine how much the plant has changed (Rizwan et al. 2017). The differences exist both within and between species among different cultivars of the same species. This study aimed to clarify the impacts of Zn-Lysine chelation applied topically on wheat (*Triticum aestivum* L.) plant growth in drought-prone environments.

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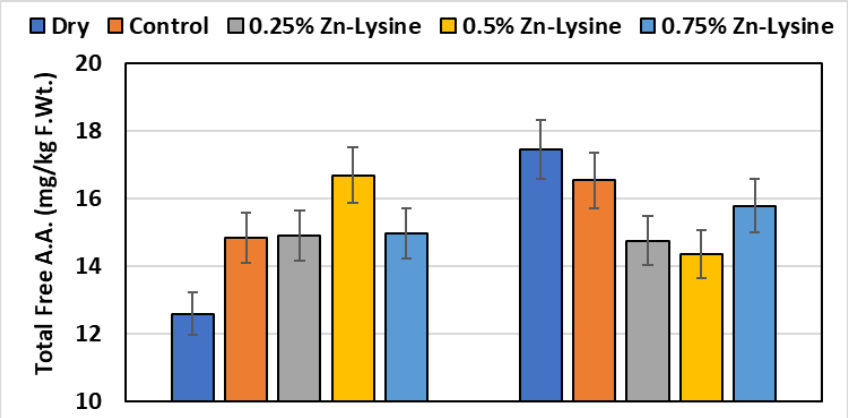

Fig. 18: Influence of Zn-Lysine foliar employment on free amino acid of wheat during drought

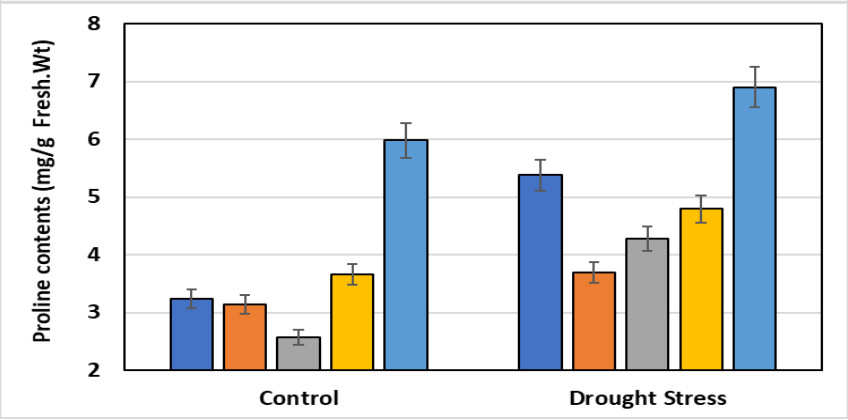

Fig. 19: Foliar Zn-Lysine treatment's effect on wheat plants' Proline during drought stress.

As stated by Rafie et al. (2017), Zn-amino chelate improved the growth parameters of onions. Furthermore, a different study discovered that the growth of wheat plants cultivated in Cd-contaminated soil was enhanced by the topical application of Zn-Lysine (Rizwan et al. 2017). This study provides further evidence, in line with our prior research, that zinc lysine promotes the growth of wheat plants under wastewater stress conditions. Ghasemi et al. (2014) have previously illustrated a comparable favorable reaction to the amino-chelate diet. Potentially due to the combined effects of zinc and lysine, the increase has occurred. Numerous physiological processes and structural components of plants are susceptible to the effects of amino acids (Nasholm et al. 2009; Rizwan et al. 2017). Because of the efficient application of amino-chelate fertilizer, the detrimental effects of effluent on wheat morphological traits have been mitigated. As an amino-chelate fertilizer, Zn-Lysine is widely used due to its capacity to enhance nutrient absorption and provide environmental protection. The combination of hazardous heavy metals with active plant elements significantly reduces the mobility of the metals (Souri 2016). The results of the study indicated that zinc supplementation substantially enhanced wheat plant growth and biomass, whereas dehydration had a profound negative impact on these characteristics. This study indicates that incorporating Zn-Lysine into the diet enhances gas exchange and photosynthetic capacity. Zinc actively participates in the maintenance of chloroplast defence and membrane structural integrity in certain plant species (Zaman et al. 2018; Zaheer et al. 2019). It is also evident that foliar Zn-Lysine administration enhanced photosynthetic and other gas exchange characteristics in the tested plant, given its elevated Cr content. Zaheer et al. (2019) report that Zn-Lysine enhances photosynthetic activity when exposed to abiotic stress. An increase in photosynthetic pigments might be attributable to amino acid complexes comprising micronutrients (Rizwan et al. 2017). Lower chlorophyll concentrations were observed in wheat, spinach, mung legumes, and sunflowers (Ali et al. 2015). These results were corroborated by prior investigations that discovered that substantial zinc ingestion enhanced the photosynthetic efficiency and gas exchange characteristics of the spinach plant. Dryer stress, according to Ahmed et al. (2017), produces reactive oxygen species (ROS), which induce severe oxidative damage. The accumulation of metals has been associated with the detection of oxidative stress in wheat plants (Rizwan et al. 2017; Sattar et al., 2021). Foliar application of Fe-lys and Zn-Lysine decreased oxidative stress in numerous plant species. Oxidative stress in plants has been identified as a consequence of metal toxicity (He et al. 2017). A number of amino acids, including zinc and glutamine, glycine, lysine, and arginine, are necessary to surmount MDA and EL concentrations in various plan. Additionally, amino-chelate fertilizers increased antioxidant activity and decreased oxidative stress, according to the current study. A separate investigation (Rizwan et al. 2017) discovered that wheat plants subjected

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to Cd stress exhibited enhanced antioxidant enzyme activity when foliar-sprayed with Zn-Lysine. A deficiency in zinc, an essential nutrient, may prevent roots from accumulating Cr when soil stress increases, according to Tauqeer et al. (2016). Micronutrient-amino chelators, which increase zinc levels in a variety of plant species, including cabbage, tomatoes, onions, wheat, and spinach, have been the subject of numerous studies. In addition, the amino-chelate fertilizer Zn-Lysine enhances the morpho-physiological characteristics and durability of wheat plants.

5. CONCLUSION

Overall, the study demonstrates that Zn-Lysine chelation, especially at specific concentrations, can effectively mitigate the adverse effects of drought stress on wheat plants, positively influencing various morphological, physiological, and biochemical parameters. These findings provide valuable insights for developing strategies to enhance plant resilience in drought-prone environments, contributing to improved agricultural productivity and crop yield.

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