

## **BIOFORTIFICATION OF WHEAT PRODUCTIVITY USING HUMIC ACID, ZINC** AND AMINO ACIDS APPLICATION

Seham Mohamed Alzweek<sup>1\*</sup>, Radia Omar Salem<sup>2</sup>

<sup>1.2</sup> Department of Crop Science, Faculty of Agriculture, University of Tripoli, Tripoli, Libya.

s.ezweek@uot.edu.lv

**التعزيز الحيوي لإنتاجية القمح باستخدام حمض الهيوميك والزنك والأحماض الأمينية** سهام محد الزويك<sup>1\*</sup>، راضية عمر سالم<sup>2</sup>

1.2 قسم علوم المحاصيل، كلبة الزر اعة، جامعة طر ابلس، طر ابلس، ليبيا

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#### Abstract

Soil application of humic acids (HA) and foliar fertilization with bio-stimulants as Zn and amino acids, and others can affect plant growth by enhancing resistance to abiotic stress and raising and sustainably increasing grain yield. Consequently, this investigation was carried out to test the response of Bohoth-210 wheat variety to soil-application of HA and foliar application of Zn and amino acids (AA) and their interaction on yield and yield components during the cropping season of 2023/2024. The factorial experiment was designed according to a complete randomized block design (CRBD) in two factors. The first factor was the application of HA as a soil amendment, with variable levels of application set at 0, 7, and 14 kg/ha. The second factor was the foliar spraying levels of (water as a control, Zn at a rate of 1.5 kg/ha, amino acids at a concentration of 1 cm/liter of water), and a combination treatment of Zn and amino acids. These treatments were randomly distributed in three replicates. The findings observed that the application of HA, Zn, and amino acids, along with their interactions, particularly at higher levels has a significant influence on yield characteristics as grain yield, yield components, and the overall protein content of the wheat produced. Such results underscore the significance of utilizing these agricultural inputs effectively to enhance wheat production in Libya, thereby contributing to food security.

Keywords: wheat; yield; yield components; zinc; amino acids; humic acid applications.

الملخص

إن إضافة الأحماض الهيوميكية (HA) إلى التربة والتسميد الورقي بالمحفز ات الحيوية مثل الزنك والأحماض الأمينية وغيرها يمكن أن يؤثر على نمو النبات مُن خلال تحسين مقاومة الاجهادات اللاإحيائية ورفع وزيادة محصول الحبوب بشكل مستدام. لذلك، تم إجراء هذا البحث لدراسة تأثير الإضافة الأرضية لحمض الهيوميك والَّرش الورقى بالزنك والأحماض الأمينيَة وتفاعلهما على محصول الحبوب ومكوناته لصنف القمح بحوث 210 خلال الموسم الزراعي 2024/2023. وصممت التجربة وفقا لتصميم القطاعات الكاملة العشوائية (RCBD) بعاملين، كان العامل الأول هو إضافةً حمض الهيوميك كمحسن للتربة، بمعدلات إضافة متفاوتة تم ضبطها عند 0 و7 و14 كجم / هكتار. سمح هذا بإجراء تحليل شامل لكيفية تأثير الكميات المختلفة من حمضُ الهيوميك على نمو محصول القمح وانتاجه. وكان العامل الثّاني معدلات الرش الورقي (الماء كشاهد، الزنك بمعدل 1.5 كجم/هكتار، والأحماض الأمينية بتركيز [1 سم لكل لتر ماء)، ومعاملة مركبة من الزنك والأحماض الأمينية. وقد تم توزيع هذه المعاملات عشوائياً في ثلاث مكررات. وأشارت النتائج المتحصل عليها إلى أن إضافة حمض الهيوميك والزنك والأحماض الأمينية، إلى جانب التفاعلات بين هذه العناصر، وخاصَّة بمعدلات إضافة عالية، لها تأثير إيجابي على محصول الحبوب ومكوناته ومحتوى البروتين الكلي للقمح المنتج. واكدة هذه النتائج على أهمية الاستفادة من هذه المدخلات الزراعية بشكل فعال لتعزيز إنتاج القمح في ليبيا، وبالتآلي المساهمة في تحقيق الأمن الغذائي.

الكلمات الدالة: القمح، الانتاج، مكونَّات الانتاج، الزنك، الأحماض الأمينية، إضافة حمض الهيوميك.

#### **INTRODUCTION**

Wheat belongs to the Poaceae family; it is the greatest widely grown and consumed and is considered a main food in furthermost country of the world, providing around twenty % of the world's dietary calories (Khalid et al., 2023). In Libya, wheat flour is exclusively used in manufacture of bread, pasta, biscuits and other related industries, and its secondary production is used in animal feed (Faraj et al, 2022). It is one of the utmost chief grain crops widely cultivated in Libya, with a total cultivated area amounting to around 168,770 hectares, and the total production surpassed 130,000 tons (FAO, 2021). To close the gap between production and consumption, efforts should be concentrated toward expanding and enhancing wheat output. There are two ways to increase total wheat production: increasing cultivated area and productivity per unit area. The first scheme comprises using high-yielding wheat varieties to upsurge productivity/unit area, but it needs a long time to breed and evaluate the stability of these new wheat variety (Kandil et al., 2016). Recently, biofortification has emerged as an agricultural strategy that can be used to overcome the main obstructions to reaching wheat production objectives. In addition, biofortified crops can improve food security, yield stability, and climate resistance in addition to improving nutrition, particularly for smallholder farmers in developing nations (Sandhu et al., 2023).

Traditional breeding, genetic manipulation of crops, soil fertilization, or foliar fertilization are the foundations of improving strategies to rise the nutritional value of agricultural products. Biofortification concluded breeding and transgenic tactics be contingent on adequate variation for a assumed attribute. Nevertheless, differences in soil categories, the features of a definite cropping system, and, progressively, climate alteration can limit the applicability of released varieties, particularly for wheat (**Sánchez-Palacios** *et al.*, 2023). Even though agronomic enrichment is the most forthright biofortification technique, its achievement relies on a factor number, comprising the type of nutrient to be provided, uptakes variations, mobility, and plant tissues accumulation, and the soil quality somewhere crop is produced (**Al-Juthery** et al., 2022).

Humic acid (HA) is the primary organic elements of soil (humus). Humic substances (HS) have numerous favorable impacts on physical structure and microbial inhabitants of soil, as well as boosting modification pathways implicated in growth stimulation, cell permeability, nutrient absorption, and yield enrichment (**Celik** *et al.* 2010). HS are classified as HA, fulvic acids (FA), and humus according to solubility in water, acidic or alkaline solutions (**De Melo** *et al.*, 2016). Utilization of HA significantly improved wheat grain protein percentage/yield, and carbohydrate % (**Abd El-Aziz** *et al.*, 2018). HA is organic molecules that play an vital role in enhancing soil properties, growth, agronomic and yield parameters. Soils, lignite, and organic ingredients are the furthermost common marketable sources of HS (**Gollenbeek and Van Der Weide**, 2020; **Yang** *et al.*, 2021). HA produced a significant rise in the growth of plants (root and shoots) in addition to moisture and N uptake of the seedlings (**Malik and Azam**, 1985). **Tahir** *et al.* (2011) and **Nardi** *et al.* (2021) stated that HA spraying raised K levels in soil following crop harvest. The HA chemical and molecular structures, and their sources also the levels of HA are crucial in determining its influences on plants and soil. Significantly, HA using might result in inconsistencies in yield, probably due to the diverse biological sources of HA (**Sible** *et al.*, 2021).

Zn is a mineral that is essential for appropriate growth and development for crops. Also, it is vital in activation of many enzymes and in the certain enzymes creation and hormones for growth. Although total Zn concentrations are adequate in many agricultural locations, accessible Zn concentrations are low due to differences in soil and climatic circumstances. The availability of Zn % in soil is influenced by pH, lime, organic matter %, clay type and %, and P level fertilizer used (**Adiloglu and Adiloglu**, 2006). Zn influences various plant life processes, containing N consumption and metabolism, photosynthetic synthesis, biotic and abiotic stresses tolerance, pollen job, and pollination, on order to Zn act a vital physiological role in their growth (**Cakmak**, 2008, and **Pandey** *et al.*, 2006)

Using Zn and Fe fertilizer as foliar application raised production and quality comparing with the untreated plants. Among treatments, spraying of (Fe + Zn) resulted in the highest seed production and quality (**Habib** 2009). Zn foliar treatment enhanced the number of viable tillers and wheat yield (**Zoz** *et al.*, 2012). The maximum grain production was noticed with the combination of soil and foliar Zn treatment, while the lowest mean values of yield was detected with the untreated plants. The greatest reduction in sterility % was gotten in soil with using Zn. In the same trend, using Zn as basal and foliar applications boost grain yield of wheat (**Firdous** *et al.*, 2018). Wheat needs Zn for development and grain quality (**Arif** *et al.*, 2017). A substantial improvement in growth and yield characteristics recorded with Zn treatment (**Wasaya** *et al.*, 2017). Zn foliar fertilizer significantly improved production and yield components compared to the control, with 0.8% Zn being the optimal treatment (**Hassanein** *et al.*, 2019).

It is widely known that amino acids are utilized as a bio-stimulant with beneficial role in growth and yield, and greatly decrease the impact of biotic stress damage. Foliar spraying with amino acids speedily corrects nutritional shortages since it is easily absorbed and directly used in synthesis in protein. It aids in increasing chlorophyll concentration, resulting in increased photosynthesis and luscious harvests. Amino acids operate as a cytoplasmic osmotic agent in stomatal cells, assisting plants in enrichment absorption of macro and micro- nutrients as well as gases by encouraging in stomatal opening. It facilitates the micronutrients absorption and movement inside the plant. It serves as an equipoise of microbial flora in soil to enhance mineralization of organic matter (OM) and the soil fertility nearby the roots (Ashmead, 1986). The application of HA and amino acids boosted grain and straw yields of wheat, but the treatment combining HA and amino acids recorded the highest yields than using each alone (El-Naggar and El-Ghamry, 2007). Spraying plants with a combination of HA and amino acids, together, achieved the highest grain production of wheat (Kandil et al., 2016). Zn-comprising amino acid treatment enlarged growth and production (Laware and Apparao 2010). Additionally, amino acids have an influence on the yield and growth, either directly or indirectly (Abd El-Aal et al., 2010). The practice of amino acids as foliar application is improving plant function. Amino acid has the importance as component elements of plant proteins in biochemical and physiological processes of plants, their use is critical (Ebrahimi et al., 2014; Shetta and Zayed 2016).

The objective of this investigation was to examine the impact of humic acid (HA) and foliar spray of Zn and amino acids, as well as their interaction, on grain yield and yield components of wheat.

#### MATERIALS AND METHODS

A field experiment was conducted at the Research and Experiment Station of the Faculty of Agriculture, University of Tripoli, Libya, during the 2023/2024 cropping season. The aim of this investigation was to evaluate the influences of soil-applied humic acid (HA) and foliar applications of zinc (Zn) and amino acids (AA) and their interaction on yield and yield components of the wheat variety Bohot-210. A randomized complete blocks design (RCBD) with the 3 replications was used. Each experiment plot included five ridges (70 cm width); each row was 3 m long and 20 cm apart; plot size was 10.5 m<sup>2</sup> (3.5 m x 3.0 m). Bohot-210 variety seeds at the rate of 150 kg/ha were used. Planting date was 20<sup>th</sup> and 25<sup>th</sup> November in the first and the second seasons.

Diammonium phosphate (18/46/0% NPK) at a rate of 150 kg/ha was used as a soil application with the soil preparation before sowing. Nitrogen (N) fertilizer at a rate of 200 kg N/ha was used as urea (46.5% N) in two equal doses; the  $1^{st}$  dose was applied at the tillering stage and the  $2^{nd}$  one at the elongation stage. All other agricultural practices were carried out as per recommendations to the trial throughout the growing season.as recommended.

The soil samples were gathered from different locations at a depth of 0-60 cm before sowing, and some physio-chemical attributes were analyzed in the Faculty of Agriculture, Tripoli University. The soil texture was analyzed using the hydrometer method (**Topp** *et al.*, 1993). Organic matter was measured using a modified version of the Walkley-Black method (**Nelson and Sommers**, 1996). Phosphorus (P) and potassium (K) was assessed using the method described by **Olsen** (1982). Nitrogen (N) levels were determined following **Jackson's** method (1973). Table 1 displays a few of the experimental site's chemical and physical characteristics.

| Particle size distribution       Sandy silt         Soil texture (%)       Sandy silt         pH (1: 2.5 water suspension)       7.80         EC (dSm <sup>-1</sup> )       3.10         Soluble Cations (meq/L.) $(dSm^{++})$ Ca <sup>++</sup> 7.60         Mg <sup>++</sup> 5.20         Na <sup>+</sup> 5.10         K <sup>+</sup> 0.50         Soluble Anions (meq/L.)       HCO <sub>3</sub> <sup>-</sup> HCO <sub>3</sub> <sup>-</sup> 3.00         CI <sup>-</sup> 4.80         SO <sub>4</sub> <sup></sup> 10.30         O.M. (%)       0.82         CaCO <sub>3</sub> (ppm)       5.75         Available Mineral       N (ppm)       9.80         K (mg/Kg)       150.00       Available P (ppm) | Soil characteristics         | Season 2023/2024 |
|--|------------------------------|------------------|
| Soil texture (%)         Sandy silt           pH (1: 2.5 water suspension) $7.80$ EC (dSm <sup>-1</sup> ) $3.10$ Soluble Cations (meq/L.) $Ca^{i+}$ $Ca^{i+}$ $7.60$ Mg <sup>++</sup> $5.20$ Na <sup>+</sup> $5.10$ K <sup>+</sup> $0.50$ Soluble Anions (meq/L.) $HCO_3^-$ HCO_3^- $3.00$ Cl <sup>-</sup> $4.80$ SO <sub>4</sub> <sup></sup> $10.30$ O.M. (%) $0.82$ CaCO <sub>3</sub> (ppm) $5.75$ Available Mineral         N (ppm)           N (ppm) $9.80$ K (mg/Kg) $150.00$   | Particle size distribution   |                  |
| pH (1: 2.5 water suspension)       7.80         EC (dSm <sup>-1</sup> )       3.10         Soluble Cations (meq/L.) $Ca^{++}$ $Ca^{++}$ 7.60         Mg <sup>++</sup> 5.20         Na <sup>+</sup> 5.10         K <sup>+</sup> 0.50         Soluble Anions (meq/L.)       0.50         HCO <sub>3</sub> 3.00         CI       4.80         SO <sub>4</sub> <sup></sup> 10.30         O.M. (%)       0.82         CaCO <sub>3</sub> (ppm)       5.75         Available Mineral       N (ppm)         N (ppm)       9.80         K (mg/Kg)       150.00         Available P (ppm)       16.90  | Soil texture (%)             | Sandy silt       |
| EC (dSm <sup>-1</sup> ) $3.10$ Soluble Cations (meq/L.) $7.60$ $Ca^{++}$ $7.60$ $Mg^{++}$ $5.20$ $Na^{+}$ $5.10$ $K^{+}$ $0.50$ Soluble Anions (meq/L.) $HCO_3^{-}$ $HCO_3^{}$ $3.00$ Cl $4.80$ $SO_4^{+-}$ $10.30$ $O.M.$ (%) $0.82$ CaCO_3 (ppm) $5.75$ Available Mineral $N$ (ppm) $9.80$ K (mg/Kg) $150.00$ Available P (ppm) $16.90$  | pH (1: 2.5 water suspension) | 7.80             |
| Soluble Cations (meq/L.) $Ca^{++}$ 7.60 $Mg^{++}$ 5.20 $Na^{+}$ 5.10 $K^{+}$ 0.50         Soluble Anions (meq/L.)       0.50         HCO <sub>3</sub> <sup>-</sup> 3.00         Cl <sup>-</sup> 4.80         SO <sub>4</sub> <sup>-+</sup> 10.30         O.M. (%)       0.82         CaCO <sub>3</sub> (ppm)       5.75         Available Mineral       9.80         K (mg/Kg)       150.00         Available P (ppm)       16.90  | $EC (dSm^{-1})$              | 3.10             |
| $Ca^{++}$ 7.60 $Mg^{++}$ 5.20 $Na^+$ 5.10 $K^+$ 0.50         Soluble Anions (meq/L.)       10.30 $HCO_3^-$ 3.00 $Cl^-$ 4.80 $SO_4^-$ 10.30 $O.M. (\%)$ 0.82 $CaCO_3$ (ppm)       5.75         Available Mineral       9.80 $K$ (mg/Kg)       150.00         Available P (ppm)       16.90  | Soluble Cations (meq/L.)     |                  |
| $Mg^{++}$ 5.20Na^+5.10K^+0.50Soluble Anions (meq/L.) $HCO_3^ HCO_3^-$ 3.00CI4.80SO_4^-10.30O.M. (%)0.82CaCO_3 (ppm)5.75Available MineralN (ppm)N (ppm)9.80K (mg/Kg)150.00Available P (ppm)16.90  | Ca <sup>++</sup>             | 7.60             |
| $Na^+$ 5.10 $K^+$ 0.50Soluble Anions (meq/L.) $HCO_3^-$ 3.00 $Cl^-$ 4.80 $SO_4^-$ 10.30 $O.M.$ (%)0.82 $CaCO_3$ (ppm)5.75Available MineralN (ppm)9.80K (mg/Kg)150.00Available P (ppm)16.90   | $Mg^{++}$                    | 5.20             |
| K <sup>+</sup> 0.50         Soluble Anions (meq/L.)  | $Na^+$                       | 5.10             |
| Soluble Anions (meq/L.) $HCO_3^ 3.00$ $Cl^ 4.80$ $SO_4^ 10.30$ $O.M.$ (%) $0.82$ $CaCO_3$ (ppm) $5.75$ Available Mineral $N$ (ppm)         N (ppm) $9.80$ K (mg/Kg) $150.00$ Available P (ppm) $16.90$   | $\mathbf{K}^+$               | 0.50             |
| HCO3 <sup>-</sup> 3.00         Cl <sup>-</sup> 4.80         SO4 <sup></sup> 10.30         O.M. (%)       0.82         CaCO3 (ppm)       5.75         Available Mineral       9.80         N (ppm)       9.80         K (mg/Kg)       150.00         Available P (ppm)       16.90  | Soluble Anions (meq/L.)      |                  |
| CI       4.80         SO <sub>4</sub> 10.30         O.M. (%)       0.82         CaCO <sub>3</sub> (ppm)       5.75         Available Mineral       9.80         K (mg/Kg)       150.00         Available P (ppm)       16.90   | HCO <sub>3</sub>             | 3.00             |
| SO4       10.30         O.M. (%)       0.82         CaCO3 (ppm)       5.75         Available Mineral       9.80         K (mg/Kg)       150.00         Available P (ppm)       16.90   | Cl                           | 4.80             |
| O.M. (%)       0.82         CaCO <sub>3</sub> (ppm)       5.75         Available Mineral       9.80         N (ppm)       9.80         K (mg/Kg)       150.00         Available P (ppm)       16.90  | SO <sub>4</sub>              | 10.30            |
| CaCO <sub>3</sub> (ppm)         5.75           Available Mineral         9.80           N (ppm)         9.80           K (mg/Kg)         150.00           Available P (ppm)         16.90  | O.M. (%)                     | 0.82             |
| Available Mineral       9.80         N (ppm)       9.80         K (mg/Kg)       150.00         Available P (ppm)       16.90   | CaCO <sub>3</sub> (ppm)      | 5.75             |
| N (ppm)         9.80           K (mg/Kg)         150.00           Available P (ppm)         16.90  | Available Mineral            |                  |
| K (mg/Kg)         150.00           Available P (ppm)         16.90   | N (ppm)                      | 9.80             |
| Available P (ppm)16.90   | K (mg/Kg)                    | 150.00           |
|  | Available P (ppm)            | 16.90            |

 Table (1). Physical and chemical properties of the experimental soil.

| Annual precipitation rate (mm/year) | 300 |
|-------------------------------------|-----|
|-------------------------------------|-----|

Humic acid (HA) labelled as HABICAR HUMICO WSP was applied to the soil at quantities of 0, 7, and 14 kg/ha after 21 days from planting. Table 2 shows the chemical and physical characteristics of HA compounds (2).

| Chemical   |  |                                  | Physicochem          |                       |                   |       |                             |                |
|------------|--|----------------------------------|----------------------|-----------------------|-------------------|-------|-----------------------------|----------------|
| Properties | Potassium<br>oxide (K <sub>2</sub> O<br>%) | Total<br>Humic<br>extract<br>(%) | Humic<br>acid<br>(%) | Fulvic<br>acid<br>(%) | Appearance        | Color | Solubility                  | P <sup>H</sup> |
| Values     | 12-15                                      | 70                               | 60                   | 10                    | Soluble<br>powder | Black | Totally<br>water<br>soluble | 8-11           |

Zinc (Zn) was obtained from the source zinc sulphate  $ZnSO_4$  7H<sub>2</sub>O, having 24% Zn, and applied as a foliar at a rate of 1.5 kg/ha at 25, 45 and 70 days after planting. Amino acids at a rate of 1 cm/L (water) were applied as a foliar treatment twice, 50 and 70 days after planting. Table 3 summarizes the properties of this fertilizer compound's analysis.

| Table (3). Structure of Amino acids (AA) compound. |           |     |     |     |     |          |          |               |  |
|--|-----------|-----|-----|-----|-----|----------|----------|---------------|--|
| Droportios   | Amino     | Р   | Fe  | Zn  | Mn  | Humic    | Fulvic   | Inert         |  |
| Properties   | acids (%) | (%) | (%) | (%) | (%) | acid (%) | acid (%) | integrant (%) |  |
| Values   | 10        | 4   | 3   | 2   | 2   | 15       | 10       | 54            |  |

## Table (3). Structure of Amino acids (AA) compound.

#### **Studied characteristics:**

- 1. Plant height at harvest (cm) was determined from the soil surface to the top of the plant from 20 plants from each plot at harvest time.
- 2. Grain yield (t/ha) measured from the harvested area (m2) from each plot in terms of kg and converted to t/ha.
- 3. Straw yield (t/ha) is measured by separating straw and spikes of biological yield, then weighing the yield of the straw in kg/m<sup>2</sup> and converting it to t/ha.
- 4. Biological yield (t/ha) was determined from the harvested area (m2) of each plot in terms of kg and converted to t/ha.
- 5. 1000-kernel weight (g): was expressed as an average weight of 1000 clean grains in grams taken from each plot.
- 6. Grains number/spike: was determined as an average grains number of twenty random spike samples from each plot.
- 7. Spikeletes number/spike was determined as an average spikelets number of twenty random spike samples from each plot.

- 8. Spikes number/m2: was determined as a fertile tillers number /m2 from each plot at harvest time.
- 9. Spike length (cm): twenty spikes were randomly taken from each plot to determine spike length (cm).
- 10. Tillers number/m2 was estimated as the fertile and sterile tillers number /m2 from each plot at harvest time.
- 11. The tillering index (%) was calculated according to the following formula:

Tillering index =  $\frac{Number \ of \ spikes/sm}{Number \ of \ tillers/sm} \times 100$ ; Where,  $sm = \ square \ meter$ 

12. Protein content (%) in grains: total nitrogen was measured in digested plant material (wheat grain) calorimetrically by Nessler's method. Reading was achieved using a wavelength of 420 nm, and N was calibrated as a percentage as follows: % N = NH4 % x 0.776485. Protein percentage was determined by estimating the total nitrogen in the grains and multiplying it by 5.75 to obtain the percentage according to AOAC (1995).

## **Data Analysis**

All the collected data during the growing season were subjected to a statistical software package (**CoStat** 2005). The least significant differences test (L.S.D.) at the 0.05 probability level was used to test significances among mean values of each treatment (**Gomez and Gomez**, 1984).

## **RESULTS AND DISCUSSION**

Data in Tables 4, 5, and 6 showed that all the yields and yield related components were significantly affect by application rates of humic acid, foliar treatments, and their interaction during the season 2023/2024. In terms of the influence of HA application on the previous characteristics, Table (4, 5, and 6) revealed that increasing the application of HA up to 14 kg/ha created a considerable rise in growth and yield characters when compared to the other treatments. These results are confirmed by findings reported by **Khan** *et al.* (2010) and **Bakry** *et al.* (2013), who concluded that humic acid increased plant growth, grain output, and wheat quality. In this connection (**Akinremi** *et al.* 2000; **Van Tol de Castro** *et al.*, 2022) suggested that the metabolic activities of plants, such as photosynthesis and total chlorophyll content, are enhanced by humic acid, leading to higher output and quality.

Likewise, the results shown in Tables 4, 5, and 6 indicated that spraying of Zn and AA caused a gradual and significant increase in all characters. The highest values of plant height (103.57 cm), grain yield (3.33 t/ha), straw yield (4.01 t/ha), biological yield (7.46 t/ha), 1000-kernel weight (53.83 and 54.77 g), grains number/spike (54.83 grains), spikeletes number/spike (29.89 spikeletes), spikes number/m2 (365.78 and 366.88 spikes), spike length (13.41 cm), tillers number/m2 (389.00 and 384.89 tillers), tillering index (93.97 %), and grain protein content (10.98%) were recorded under foliar application of Zn and amino acids as compared to the other treatments, but the lowest ones were recorded with control conditions (without using HA, Zn, or AA).

The results clearly illustrated the substantial interaction impact of HA x foliar application on the tested characteristics, where the maximum level of HA with foliar application of Zn and amino acid increased these characters over the control (Tables 4, 5 and 6). The previous results revealed that increasing humic acid rate caused an increase in all the studied characters; that increase may be due to the role of HA in plants as follows: It increases plant biochemical processes like photosynthesis and chlorophyll content, which results in enlarged quality and production (**Akinremi** *et al.*, 2000). Our findings were corroborated by **Khan** et al. (2010), **Tahir** *et al.* (2011), **Nardi** *et al.* (2021), and **Bakry** *et al.* (2013), who concluded that humic acid improved growth and grain production attributes.

The comparatively substantial increase in grain yield and yield components caused by Zn foliar fertilizer demonstrated the importance of Zn and amino acids to plants, both of which play critical roles in many processes in plant organs. Many studies have shown that applying Zn is one of the most applicable methods to enhance cereal crops (**El-Metwally** *et al.*, 2012). **El-Habbasha** *et al.* (2015) and **Esfandiari** *et al.* (2016) showed that spraying of Zn had a significant positive influence on wheat grain yield and its constituents. These findings are consistent with and support the conclusions reached by those other studies such as **El-Dahshouri** *et al.* (2017); **Kandil and Marie** (2017).

Amino acids are effective bio-stimulants that enhance plant growth, yield, and reduce biotic stress damage. When applied as foliar sprays, they quickly address nutrient deficiencies, boost chlorophyll levels for improved photosynthesis, and lead to better harvests. They act as osmotic agents in stomata, facilitating nutrient, and gas absorption. Additionally, amino acids increase enzyme activity related to protein and carbohydrate synthesis, impacting biomass and yield. In wheat, amino acid application improved grain and straw yields, and combining humic substances with amino acids resulted in even higher yields (**El-Naggar and El-Ghamry**, 2007).

The observed enhancement in wheat growth and production attributed to the application of foliar spraying with Zn and AA can be closely related to their essential functions within biological systems. Zn, along with AA, is recognized for its critical role as an integral metal component in various enzymes. Furthermore, amino acids serve not only as creating proteins meanwhile crucial functional, structural, or regulatory co-factors that contribute to the activity and efficiency of a wide array of enzymatic processes. This interconnectedness highlights how the presence of Zn and AA in foliar applications may bolster enzymatic functions, ultimately leading to improved wheat growth and higher production yields. (Hotz and Braun, 2004). Zn is vital in the production of biomass, according to Cakmak (2008); Yavas and Unay (2016) and El-Dahshouri *et al.* (2017).

| Character         | q                    | 2023/2024             |       |            |                  |       |  |
|-------------------|----------------------|-----------------------|-------|------------|------------------|-------|--|
|                   | umic aci<br>kg/ha);A | Foliar Application; B |       |            |                  |       |  |
|                   | H                    | Water                 | Zn    | Amino acid | Zn + Amino acids | Ai    |  |
|                   | 0                    | 77.67                 | 84.41 | 90.33      | 95.17            | 86.90 |  |
| Plant height (cm) | 7                    | 83.95                 | 87.62 | 93.64      | 107.02           | 93.06 |  |
|                   | 14                   | 86.24                 | 88.86 | 98.84      | 108.53           | 95.62 |  |
|                   |                      |                       |       |            |                  |       |  |

 Table 4. Impact of humic acid, foliar application of Zn and amino acids, and their interactions on wheat characteristics in the 2023/2024 season.

| Average (B)           |    | 82.62 | 86.96 | 94.27 | 103.57 |       |
|-----------------------|----|-------|-------|-------|--------|-------|
| LSD at 0.05           |    |       |       |       |        |       |
| А                     |    |       |       | 1.87  |        |       |
| В                     |    |       |       | 2.16  |        |       |
| AB                    |    |       |       | 3.73  |        |       |
|                       | 0  | 2.43  | 2.66  | 2.98  | 3.27   | 2.84  |
| Grain yield (t/ha)    | 7  | 2.37  | 2.86  | 3.08  | 3.23   | 2.89  |
|                       | 14 | 2.47  | 2.80  | 3.04  | 3.50   | 2.95  |
| Average (B)           |    | 2.42  | 2.77  | 3.03  | 3.33   |       |
| LSD at 0.05           |    |       |       |       |        |       |
| А                     |    |       |       | 0.11  |        |       |
| В                     |    |       |       | 0.13  |        |       |
| AB                    |    |       |       | 0.22  |        |       |
|                       | 0  | 2.92  | 3.21  | 3.58  | 3.89   | 3.40  |
| Straw yield (t/ha).   | 7  | 2.81  | 3.44  | 3.69  | 3.93   | 3.47  |
|                       | 14 | 3.03  | 3.36  | 3.70  | 4.20   | 3.57  |
| Average (B)           |    | 2.92  | 3.34  | 3.66  | 4.01   |       |
| LSD at 0.05           |    |       |       |       |        |       |
| A                     |    |       |       | 0.14  |        |       |
| В                     |    |       |       | 0.16  |        |       |
| AB                    |    |       |       | 0.27  |        |       |
| Dialogical wield      | 0  | 5.35  | 5.87  | 6.56  | 7.16   | 6.24  |
| biological yield –    | 7  | 5.18  | 6.3   | 6.77  | 7.16   | 6.35  |
| (t/lia) —             | 14 | 5.50  | 6.16  | 6.74  | 7.7    | 6.53  |
| Average (B)           |    | 5.34  | 6.11  | 6.69  | 7.34   |       |
| LSD at 0.05           |    |       |       |       |        |       |
| A                     |    |       |       | 0.25  |        |       |
| В                     |    |       |       | 0.29  |        |       |
| AB                    |    |       |       | 0.50  |        |       |
| 1000 transal vuoi abt | 0  | 36.17 | 41.29 | 45.13 | 52.65  | 43.81 |
| 1000- kernel weight – | 7  | 35.67 | 43.67 | 48.17 | 54.00  | 45.38 |
| (g) –                 | 14 | 40.15 | 47.08 | 46.50 | 54.83  | 47.14 |
| Average (B)           |    | 37.33 | 44.01 | 46.60 | 53.83  |       |
| LSD at 0.05           |    |       |       |       |        |       |
| А                     |    |       |       | 2.30  |        |       |
| В                     |    |       |       | 2.66  |        |       |
| AB                    |    |       |       | 4.60  |        |       |

# Table 5. Impact of humic acid and foliar applications of Zn and amino acids on wheat yield components in the 2023/2024 season.

| components in the 2023/2024 season. |           |       |             |            |                  |       |  |  |
|-------------------------------------|-----------|-------|-------------|------------|------------------|-------|--|--|
|                                     | Humic     |       |             | a<br>()    |                  |       |  |  |
| Characters                          | acid      |       | ver<br>e (A |            |                  |       |  |  |
|                                     | (kg/ha);A | Water | Zn          | Amino acid | Zn + Amino acids | A go  |  |  |
|                                     | 0         | 39.00 | 43.29       | 48.47      | 55.15            | 46.48 |  |  |
| Grains number/spike                 | 7         | 37.00 | 45.67       | 50.67      | 54.33            | 46.92 |  |  |
|                                     | 14        | 41.65 | 49.91       | 49.33      | 55.00            | 48.97 |  |  |
|                                     |           |       |             |            |                  |       |  |  |

| Average (B)                   |    | 39.22  | 46.29  | 49.49  | 54.83  |        |
|-------------------------------|----|--------|--------|--------|--------|--------|
| LSD at 0.05                   |    |        |        |        |        |        |
| А                             |    |        | 2.70   |        |        |        |
| В                             |    |        | 3.13   |        |        |        |
| AB                            |    |        | 5.42   |        |        |        |
| 0 1 1 4                       | 0  | 19.37  | 22.17  | 24.23  | 27.57  | 23.34  |
| Spikelets                     | 7  | 21.67  | 25.00  | 27.92  | 31.17  | 26.44  |
| number/spike                  | 14 | 23.15  | 25.90  | 28.55  | 30.93  | 27.13  |
| Average (B)                   |    | 21.40  | 24.36  | 26.90  | 29.89  |        |
| LSD at 0.05                   |    |        |        |        |        |        |
| А                             |    |        | 0.68   |        |        |        |
| В                             |    |        | 0.78   |        |        |        |
| AB                            |    |        | 1.35   |        |        |        |
|                               | 0  | 249.57 | 277.45 | 308.75 | 336.67 | 293.11 |
| Spikes number/m <sup>2</sup>  | 7  | 260.87 | 295.72 | 326.01 | 379.67 | 315.57 |
|                               | 14 | 290.95 | 316.30 | 348.68 | 381.00 | 334.23 |
| Average (B)                   |    | 267.13 | 296.49 | 327.81 | 365.78 |        |
| LSD at 0.05                   |    |        |        |        |        |        |
| А                             |    |        | 6.27   |        |        |        |
| В                             |    |        | 7.24   |        |        |        |
| AB                            |    |        | 12.54  |        |        |        |
|                               | 0  | 9.14   | 10.37  | 10.87  | 12.73  | 10.78  |
| Spike length (cm)             | 7  | 9.87   | 10.87  | 11.83  | 12.97  | 11.39  |
|                               | 14 | 10.40  | 11.80  | 12.40  | 14.53  | 12.28  |
| Average (B)                   |    | 9.80   | 11.01  | 11.70  | 13.41  |        |
| LSD at 0.05                   |    |        |        |        |        |        |
| А                             |    |        | 0.30   |        |        |        |
| В                             |    |        | 0.34   |        |        |        |
| AB                            |    |        | 0.59   |        |        |        |
|                               | 0  | 280.29 | 312.79 | 338.88 | 380.67 | 328.16 |
| Tillers number/m <sup>2</sup> | 7  | 298.00 | 327.72 | 354.09 | 392.33 | 343.04 |
|                               | 14 | 323.95 | 349.27 | 381.68 | 394.00 | 362.23 |
| Average (B)                   |    | 300.75 | 329.93 | 358.22 | 389.00 |        |
| LSD at 0.05                   |    |        |        |        |        |        |
| A                             |    |        | 7.35   |        |        |        |
| В                             |    |        | 8.48   |        |        |        |
| AB                            |    |        | 14.70  |        |        |        |

Table 6. Impact of humic acid and foliar application of Zn, amino acids and their interaction on tillering index and Protein content (%) of wheat in 2023/2024 season.

|            | 2023/2024                                 | Đ             |
|------------|---|---------------|
| Characters | Humic<br>(kg/pa)<br>Foliar Application, B | Averag<br>(A) |

|                     |    | Water | Zn    | Amino acid | Zn + Amino acids |       |
|---------------------|----|-------|-------|------------|------------------|-------|
|                     | 0  | 89.03 | 88.71 | 91.13      | 88.42            | 89.32 |
| Tillering index (%) | 7  | 87.76 | 90.24 | 92.11      | 96.77            | 91.72 |
|                     | 14 | 89.81 | 90.56 | 91.35      | 96.71            | 92.11 |
| Average (B)         |    | 88.87 | 89.84 | 91.53      | 93.97            |       |
| LSD at 0.05         |    |       |       |            |                  |       |
| А                   |    |       | 1.93  |            |                  |       |
| В                   |    |       | 2.23  |            |                  |       |
| AB                  |    |       | 3.86  |            |                  |       |
| _                   | 0  | 8.33  | 9.90  | 10.51      | 10.5             | 9.81  |
| Protein content (%) | 7  | 8.60  | 9.64  | 10.21      | 10.44            | 9.72  |
|                     | 14 | 9.36  | 10.14 | 10.89      | 11.99            | 10.60 |
| Average (B)         |    | 8.76  | 9.89  | 10.54      | 10.98            |       |
| LSD at 0.05         |    |       |       |            |                  |       |
| А                   |    |       | 0.33  |            |                  |       |
| В                   |    |       | 0.38  |            |                  |       |
| AB                  |    |       | 0.66  |            |                  |       |

### Conclusion

It has been confirmed through various studies that the application of humic acid (HA) leads to significantly greater yields and enhancements in yield components when compared to control treatments that did not include HA. The positive effects of foliar treatment were particularly noteworthy, as they resulted in substantial improvements in both the growth of wheat and its yield characteristics. When considering the synchronized application of humic acid, zinc (Zn), and amino acids, it was evident that these factors collectively had a pronounced impact on wheat production and the various components that contribute to yield. Based on the observed results, it is reasonable to conclude that the combination of HA with Zn and amino acids effectively enhanced wheat yield parameters. This combination not only resulted in the highest grain yield but also increased the grain protein %. These findings were consistent under the specific conditions of the studied region and were also applicable to similar regions, reinforcing the beneficial effects of this treatment combination in boosting wheat production and nutritional quality.

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