## RESPONSE OF VARIOUS ZINC APPLICATION METHODS AND CONCENTRATIONS OF PHYTOHORMONES ON YIELD AND GRAIN ENRICHMENT OF ZINC IN EARLY AND LATE SOWN WHEAT (*Triticum aestivum* L.) VARIETIES

Thesis Submitted for the Award of the Degree of

### **DOCTOR OF PHILOSOPHY**

in

Agronomy

By

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LOVELY PROFESSIONAL UNIVERSITY, PUNJAB 2024

#### DECLARATION

I, hereby declared that the presented work in the thesis entitled "Response of various zinc application methods and concentrations of phytohormones on yield and grain enrichment of zinc in early and late sown wheat (*Triticum aestivum* L.) Varieties" in fulfilment of degree of Doctor of Philosophy (Ph. D.) is outcome of research work carried out by me under the supervision of Dr. Bhupendra Mathpal (20525), working as Associate Professor, in the Department of Agronomy, School of Agriculture of Lovely Professional University, Phagwara, Punjab, India. In keeping with general practice of reporting scientific observations, due acknowledgements have been made whenever work described here has been based on findings of other investigator. This work has not been submitted in part or full to any other University or Institute for the award of any degree.

Simarystingh

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#### CERTIFICATE

This is to certify that the work reported in the Ph. D. thesis entitled "Response of various zinc application methods and concentrations of phytohormones on yield and grain enrichment of zinc in early and late sown wheat (*Triticum aestivum* L.) Varieties" submitted in fulfillment of the requirement for the reward of degree of Doctor of Philosophy (Ph.D.) in the Department of Agronomy, School of Agriculture, is a research work carried out by Simarjot Singh (Registration no: 12014329), is bonafide record of his original work carried out under my supervision and that no part of thesis has been submitted for any other degree, diploma or equivalent course.

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#### ABSTRACT

**Thesis title:** "Response of various zinc application methods and concentrations of phytohormones on yield and grain enrichment of zinc in early and late sown wheat (*Triticum aestivum* L.) varieties"

The field experiment was performed at agriculture farm, School of Agriculture, Lovely Professional University, Phagwara, Punjab during the rabi seasons of 2021-22 and 2022-23. The first objective was to study the effect of various zinc (Zn) application methods on Zn uptake, grain yield and quality of early and late sown wheat varieties. The second objective was to evaluate the impact of various concentrations of phytohormones on uptake of Zn, its transport and grain quality of early and late sown wheat varieties. The third objective was to study the comparative economics of various zinc application methods and concentrations of phytohormones on early and late sown wheat varieties. While the last objective was to evaluate the effect of various Zn application methods on nutrient status of the soil. The design of experiment was split-split plot replicated thrice for each treatment. The main plots consisted of two varieties, namely V725 (PBW 725) and V752 (PBW 752) that were split into three sub plots: Zn<sub>0</sub> (no zinc application), Zn<sub>62.5</sub> (62.5 kg/ha of zinc sulphate applied to the soil) and Zn<sub>31.25,F</sub> (31.25 kg/ha of zinc sulphate applied to the soil together with a foliar spray of 0.5% zinc sulphate solution). The subplots were further split into four sub-sub plots, namely  $H_0$  (no hormone application),  $H_1$  (10 mg/L GA), H<sub>2</sub> (10 mg/L cytokinin) and H<sub>3</sub> (5 mg/L GA plus 5 mg/L cytokinin). For growth parameters, maximum height of plant and accumulation of dry matter were recorded under treatment V<sub>725</sub>+Zn<sub>31.25,F</sub>+H<sub>1</sub> while number of tillers and leaf area index were recorded maximum under treatment V<sub>725</sub>+Zn<sub>31.25,F</sub>+H<sub>2</sub>. Among the yield contributing attributes, the treatment V<sub>725</sub>+Zn<sub>31.25,F</sub>+H<sub>2</sub> reported maximum number of effective tillers, spike length, 1000-grain weight and number of grains per spike. The grain yield was recorded maximum under treatment V725+Zn31.25,F+H2 while the straw yield and harvest index were recorded maximum under treatment V<sub>725</sub>+Zn<sub>31.25,F</sub>+H<sub>1</sub>. On accessing the quality of wheat, grain Zn content and protein content was maximum under treatment V<sub>725</sub>+Zn<sub>31.25,F</sub>+H<sub>2</sub>. The Zn content in stem and leaves were recorded maximum under treatment V<sub>725</sub>+Zn<sub>31.25,F</sub>+H<sub>1</sub>. The grain phytic acid was recorded minimum under treatment V<sub>725</sub>+Zn<sub>31.25,F</sub>+H<sub>2</sub>. The soil Zn content was found maximum under treatment V725+Zn62.5+H3. Maximum benefit-cost ratio was found

under treatment  $V_{725}$ + $Zn_{31.25,F}$ + $H_2$ . Identical outcomes were reported for both the 2021-22 and 2022-23 seasons.

Keywords: Wheat, zinc, cytokinin, gibberellic acid and fertilizer application methods

Dr. Bhupendra Mathpal Advisor Simarjot Singh Author

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### List of abbreviations

Abbreviations	Full name
Al	Aluminium
ANOVA	Analysis of variance
ATP	Adenosine triphosphate
B:C	Benefit: cost
BAP	Benzyl aminopurine
BR	Brassinosteroids
BSA	Bovine serum albumin
С	Carbon
CD	Critical difference
СКХ	Cytokinin oxidase/dehydrogenase
CRI	Crown root initiation
Cu	Copper
DAP	Diammonium phosphate
DAS	Days after sowing
DNA	Deoxyribonucleic acid
EC	Electric conductivity
Fe	Iron
GA	Gibberellic acid
Н	Hormone
HI	Harvest index
HMA	Heavy metal ATPase
IPT	Isopentenyl transferase
K	Potassium
LAI	Leaf area index
Mn	Manganese
МОР	Muriate of potash
N	Nitrogen
NCD	Non-communicable diseases
NP	Nano particle
Р	Phosphorus
рН	Negative logarithm of H <sup>+</sup> ions

RDF	Recommended dose of fertilizer
RNA	Ribonucleic acid
ROS	Reactive oxygen species
TaCKX	Triticum aestivum cytokinin oxidase/dehydrogenase
V	Variety
YSL	Yellow stripe-like
ZIP	Zn-regulated, Fe-regulated transporter-like proteins
ZmIPT	Zea mays isopentenyl transferase
Zn	Zinc

## List of units

Units	Full name
%	Percent
cm	Centimetre
cm <sup>2</sup>	Centimetre square
ds/m	Deci siemens per metre
g	Gram
Kg/ha	Kilogram per hectare
m <sup>2</sup>	Metre square
mg	Milligram
mg/kg	Milligram per kilogram
mg/L	Milligram per litre
ml	Millilitre
ml/kg	Millilitre per kilogram
mM	Millimolar
N	Normality
nm	Nanometre
q/ha	Quintal per hectare
rpm	Revolutions per minute

# List of chemical compounds

Chemical formula	Chemical name
CaCO <sub>3</sub>	Calcium carbonate

EDTA	Ethylene diamine tetra acetic acid
FeCl <sub>3</sub> .6H <sub>2</sub> O	Ferric chloride
Fritted glass	Zinc frits
H <sub>2</sub> SO <sub>4</sub>	Sulphuric acid
HCl	Hydrochloric acid
HClO <sub>4</sub>	Perchloric acid
HEDTA	Hydroxy ethyl ethylene diamine tri acetic acid
HNO <sub>3</sub>	Nitric acid
KNO <sub>3</sub>	Potassium nitrate
MgSO <sub>4</sub>	Magnesium sulphate
Na <sub>2</sub> ZnEDTA	Disodium Zn EDTA
NaCl	Sodium chloride
NaH <sub>2</sub> PO <sub>4</sub>	Mono sodium phosphate/ sodium dihydrogen phosphate
NaZnEDTA	Sodium Zn EDTA
NaZnHEDTA	Sodium Zn HEDTA
Zn(NH <sub>3</sub> ) <sub>4</sub> SO <sub>4</sub>	Ammoniated Zn
Zn(NO <sub>3</sub> ) <sub>2</sub> .3H <sub>2</sub> O	Zinc nitrate
$Zn_3(PO_4)_2$	Zinc phosphate
ZnCl <sub>2</sub>	Zinc chloride
ZnCl <sub>2</sub>	Zinc chloride
ZnCO <sub>3</sub>	Zinc carbonate
ZnO	Zinc oxide
ZnO.ZnSO <sub>4</sub>	Zinc oxysulphate
ZnSO <sub>4</sub>	Zinc sulphate
ZnSO <sub>4</sub> .4Zn(OH) <sub>2</sub>	Basic zinc sulphate
ZnSO <sub>4</sub> .7H <sub>2</sub> O	Zinc sulphate heptahydrate
ZnSO <sub>4</sub> .H <sub>2</sub> O	Zinc sulphate monohydrate

#### CHAPTER – I

#### **INTRODUCTION**

Triticum aestivum L., Triticum durum L. and Triticum dicoccum L. are species of wheat which are cultivated in the Indian subcontinent from ancient times. The data from the Ohalo sites in Israel showed that wheat is under cultivation since 19,000 years (Kislev et al., 1992). Researchers and historians claim that the wheat cultivation may be traced back to a time period between 3000 BC and 5000 BC. According to Hasan et al. (2016), wheat is the most widely consumed crop in the world and is number one in terms of production among the major cereals. Its worldwide production was greater than 800 million tonnes in the year 2022-23. In terms of production, it ranks second in India after rice. The production reached a record-breaking 108 million tonnes in the same year (Anonymous, 2023). Around fifty percent of the total calories consumed daily in India are derived from wheat. Micronutrient deficiencies are more widespread in various regions of the world compared to low-calorie consumption and inadequate food quality (Stewart et al., 2010). Zinc (Zn), iron (Fe) and Vitamin A deficiency is accountable for about 20% of fatality in children below the age five (Prentice et al., 2008). Considering the fact that it is a low-cost food resource, it is important to improve wheat quality.

Zinc is a mineral nutrient that is required for all living beings. Todd, Elvehjem and Hart in 1933, discovered it as essential for growth and nourishment of fungus *Aspergillus niper* and subsequently it was recognised as essential for mice and rats. It was in 1960s that scientists recognised Zn as an essential trace element for human health (**Zhao** *et al.*, **2021**). Zinc is a crucial component of a variety of proteins & enzymes and it plays a catalytic, functional, structural and regulatory role in different immunological, clinical and biochemical processes. These processes include immune function, transcription of DNA, synthesis of blood etc (**Kambe** *et al.*, **2015**). According to **Noulas** *et al.* (**2018**), plants require Zn at a concentration between 30 and 100 mg/kg for their normal development.

Both quantity and quality of wheat are greatly impacted when there is a deficiency of Zn. This is because Zn is an essential component for biosynthesis of auxin, synthesis of proteins, development of pollen grains and maintenance of ion transport systems. If Zn concentrations is less than 15-20 mg/kg in plants, they display brownish spots on their uppermost leaves, interveinal chlorosis on their young leaves and midribs, as well as retard the growth and induce sterility of spikelets

(Sinclair and Kramer, 2012). Zinc application to crops resulted in an increase in the count of tillers, accumulation of dry matter and leaf area index. The wheat crop showed an increased number of productive tillers and number of ears, which led to increased yield and productivity. In developing nations, Zn deficiency is sixth largest cause of sickness and mortality. Approximately there are 3 billion people globally who are suffering from Fe and Zn deficiency (Cakmak, 2002). The skin, gastrointestinal tract, immunological system, central nervous system and reproductive system are all affected by Zn deficiency in humans.

Because of its superior agronomic flexibility, nutritional content, ease of storage and palatability, wheat is main source of sustenance for around 33% of global population spread over forty different countries. Wheat comprises 28% of the total edible dry matter produce, sixty percent of daily calorie intake and 20 % of protein that is consumed per person on a worldwide scale (**Long, 2019**). Although, wheat has a relatively low content of Zn, it is nonetheless an important component of the diets of those living in developing nations. For fulfilling the requirements of human body, Zn content in wheat grains should be at least 45 mg/kg. However, according to statistical analysis, the average wheat grain Zn content over the world is merely 28 mg/kg, which is an amount less than the quantity that is necessary for growth and development (**Wang, 2011**). According to **Wang et al.** (**2020**), one of the most important strategies for addressing Zn deficiency is to increase the daily intake of Zn through the consumption of wheat-based processed food items.

Wheat endosperm has a lower Zn level than the seed coat because of several processes that prevent Zn transportation from seed coat into the endosperm. Phytic acid is also responsible for the sequestration of Zn as insoluble ligands by aleurone cells (**Persson et al., 2016**). Transporter proteins like Zn-regulated, Fe-regulated transporter-like proteins (ZIP) and yellow stripe-like (YSL) as well as chelating molecules such as 20-deoxymugineic acid and nicotianamine are present in the seed's external tissue area, also known as bran. It is during germination that Zn is transported to the inner parts of the endosperm (**Walker and Waters, 2011**). Consequently, the minimum activity of the transporters and small number of the chelating molecules cause a reduction in the concentration of Zn that is transported from the bran to endosperm during the reproductive stage of crop (**Zhao et al., 2021**). The endosperm cavity is responsible for isolating the endosperm crease and this dis-continuity in the channel of transport may serve as an additional barrier to the transfer of Zn into

endosperm (**Persson** *et al.*, **2016**; **Ajiboye** *et al.*, **2015**). Furthermore, the number of transporters and their activity in the transfer cells that govern efflux and/or uptake of Zn in modified aleurone cells are very low, which has the limiting effect on accumulation process of Zn into wheat endosperm (**Wang** *et al.*, **2011**).

Therefore, many methods have been used to enhance the Zn content in wheat where in, Zn biofortification through agronomic or genetic engineering techniques are regarded as effective (Nakandalage *et al.*, 2016). Agronomic biofortification with Zn fertilisation, more especially foliar application of Zn, is commonly believed to be an instant and economically feasible solution to the problem of Zn deficiency in wheat grains (Ram *et al.*, 2016). Previous research has showed that foliar spray of Zn has the potential to improve the concentration of Zn in wheat grains (Wang *et al.*, 2012). When it comes to determine the concentration of Zn in wheat grain, the type of fertiliser, concentration and the time of the foliar application are all extremely important factors. As, anthesis is considered as an optimal stage for enhancing the content of Zn in wheat grains hence, foliar application at this stage is advisable (Saha *et al.*, 2017).

The term "plant growth regulators" is widely used to refer the phytohormones, are chemical substances that are found in plant body in relatively low concentrations. These include auxin, cytokinin, gibberellin, abscisic acid and ethylene. Cytokinin is produced in the roots and play a substantial role in the process of aiding cell division, boosting germination and delaying senescence (**Honig et al., 2018**). In addition, it controls the amount of leaf primordia that are produced as well as the size of the shoot meristem. These functions of cytokinin help the plants to stay green during different stress conditions boosting assimilation and biomass production. Further, the transport of cytokinin takes place through the xylem demonstrating its role in improving availability of nutrients. Gibberellin, which is often referred to as gibberellic acid (GA), accelerates the process of cellular elongation, increases the size of fruits and hastens seed germination (**Niharika et al., 2021**). The hormones not only help plants to achieve optimal growth and functioning, but they also help plants in dealing with stressful situations.

Cytokinin and GA plays a crucial role in the process of Zn translocation from straw to grain as well as in the synthesis of proteins through a variety of direct and indirect modes of action. Cytokinin application promotes the gene expression of TaCKX (*Triticum aestivum* cytokinin oxidase/dehydrogenase). The TaCKX plays a role in controlling the cytokinin concentration inside the wheat plant and increases nutrient translocation especially of Zn to the grains (**Zarea** *et al.*, **2023**). Cytokinin enhance the sink strength by promoting the endospermic cell division which results in greater assimilate translocation leading to heavier and nutrient rich grains. According to **Gao** *et al.* (**2019**), both hormones are responsible for regulating ZIP and heavy metal ATPase (HMA) proteins that assists the translocation of Zn throughout the plant body. Increasing the concentrations of these hormones through exogenous application results in increased Zn in the grains of the wheat crop (**Ren** *et al.*, **2023**). In addition, the hormones activate the production of RNA, which is associated with an increased protein content in the grains (**Karunadasa** *et al.*, **2020**; **Prasad**, **2022**). Taking into consideration the points presented above, the current study hypothesizes that application of Zn and phytohormones (GA and cytokines) play vital role in improving the growth, yield and quality of wheat.

The present study was carried out with following objectives.

#### **Objectives:**

- **1.** To study the effect of various methods of zinc application on zinc uptake, yield and quality of early and late sown wheat varieties.
- **2.** To evaluate the impact of various concentrations of phytohormones on zinc uptake, transport and quality of early and late sown wheat varieties.
- **3.** To study the comparative economics of various methods of zinc application and concentrations of phytohormones on early and late sown wheat varieties.
- **4.** To evaluate the effect of various methods of zinc application on Zn nutrient status of the soil.

#### **2.1 Wheat characteristics**

Wheat is an imperative staple food crop for 1/3<sup>rd</sup> of the global population (**Grote** *et al.*, 2021). It provides more proteins and calories to the global diet than the rest of the cereal crops (**Abdel-Aal** *et al.*, 1998; **Adam** *et al.*, 2002; **Shewry** *et al.*, 2009). It possesses high nutritional value, has convenient storage and transportation capabilities and may be transformed into numerous food items through processing. Wheat is regarded as a valuable source of minerals, proteins, vitamin B-complex and dietary fibres (**Iqbal** *et al.*,2022; **Shewry** *et al.*, 2007; **Simmonds** *et al.*, 1989). However, the nutritional content of wheat grains including its bran coating, vitamins and minerals can be influenced by environmental circumstances. Despite this, wheat remains an exceptional food for promoting good health. Wheat flour is utilised in the production of bread, cookies, confectionery items, noodles and essential wheat gluten or seitan (**Guzman** *et al.*, 2022). Wheat has several purposes including being utilised as animal feed, for the manufacturing of ethanol, in beer brewing, as cosmetics raw material, as meat substitute protein and in the creation of wheat straw composites (**Belcar** *et al.*, 2022).

Wheat germ and bran are beneficial for their high dietary fibre content which aids in preventing and treating certain digestive diseases and consuming whole-wheat bread is beneficial for one's overall health (**Joye, 2020**). Undoubtedly, the success of wheat may be attributed to its adaptability and high yields. However, these factors alone do not fully explain its current dominance in the temperate regions. One distinguishing feature that gives wheat an edge over other crops in temperate regions is the exceptional qualities of dough made from wheat flours. These qualities enable it to be transformed into many types of baked goods (such as cakes and cookies), pasta, noodles, breads and various other processed meals (**Guzman** *et al.*, **2022**). The features of wheat proteins are determined by their structures and interactions, which collectively constitute the gluten protein fraction. Lutein is the most abundant carotenoid found in wheat (**Guan** *et al.*, **2022**). Lutein, in conjunction with zeaxanthin, plays a crucial role in maintaining the well-being of the human skin and eye. The bran/germ portions of wheat have higher levels of antioxidants and carotenoids compared to the endosperm portions (**Baublis** *et al.*, **2000**).

Nutrients	Wheat bran	Wheat flour	Wheat germ
Carbohydrate (g)	26.8	68.5	44.7
Fat (g)	5.5	2.0	9.2
Protein (g)	14.1	12.6	26.7
Starch (g)	2.0	66.8	28.7
Total sugar (g)	3.8	1.7	16.0
Riboflavin (mg)	0.36	0.07	0.72
Vitamin E (mg)	2.6	0.6	22.0
Thiamine (mg)	0.89	0.30	2.01
Niacin (mg)	39.6	1.7	45.0
Folate (ug)	260.0	51.0	-

 Table 2.1: The nutritional value of different parts of wheat per 100 gram edible portion.

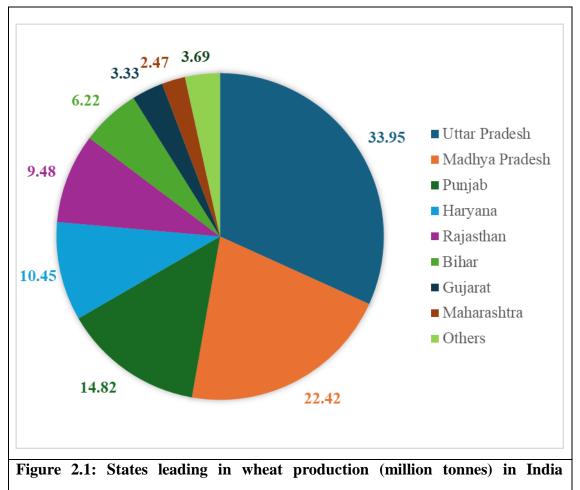
The most recent version of the USDA Diet Guidelines explicitly specifies that all adults must consume a minimum of three servings of whole grains on a daily basis. It aids in the prevention of both cardiovascular illnesses and cancer, hence reducing mortality rates. The prevention of heart disease might happen from the consumption of whole grains, phytochemicals, vitamins, antioxidants, fibre and trace minerals (Slavin et al., 2001). Whole grains aid in the prevention of diabetes by enhancing insulin sensitivity and reducing the impaired insulin activity associated with metabolic syndrome (Kumar et al., 2011). The identical compounds found in whole grains which provide protection against heart-related diseases, also have a preventive effect on several types of cancer (Slavin, 2004). The fibre and specific types of carbohydrates found in whole grains undergo fermentation in the colon, resulting in the production of compounds that have the potential to inhibit the cancer-promoting properties of bile acids. In addition, experts hypothesise that certain compounds included in whole grains might influence level of hormones and perhaps reduce the likelihood of hormone related malignancies such as breast cancer. Wheat has several forms such as wheat berries, wheat bran, grouts and cracked wheat. These forms are rich in fibre and can help lower the risk of colon cancer (Francavilla et al., 2020;

**Guo** *et al.*, **2022**). Consuming wheat decreases estrogen levels in the bloodstream, hence decreasing the likelihood of developing prostate and breast cancers. Whole grains also include antioxidants that are linked to a decreased risk of certain types of cancer.

#### 2.1.1 Nutritional importance of wheat

There are billions of people globally who heavily depend on wheat as a significant component of their diet. Hence, the significance of wheat proteins in terms of nutrition should not be undervalued, especially in underdeveloped nations where bread, noodles and other food items may contribute significantly to the overall diet (Adams et al., 2002). Wheat contributes around 55% of the total carbohydrate intake and supplies around 20% of the total calories consumed in meals. The composition of wheat consists of 78.10% carbohydrates, 12.70% proteins, 2.10% fat, and 2.10% minerals (Iqbal et al., 2022). Additionally, it includes significant amounts of vitamins (namely vitamin-B and thiamine) as well as minerals such as iron (Fe). Wheat grain is a valuable source of trace elements such as selenium (Se) and magnesium (Mg), which are crucial for maintaining good health (Shewry et al., 2006; Fraley, 2003). The wheat grain is referred to as caryopsis, which is composed of fruit or pericarp and true seed. Seed endosperm stores proteins (around 72 %), accounting for 8-15% of the total protein per weight of grain. Wheat grains contain significant amounts of sugars, riboflavin, pantothenic acid and other minerals. The barn is composed of aleurone, pericarp and testa that serves as a nutritional source of fibre, phosphorus (P), potassium (K), calcium (Ca), magnesium (Mg) and niacin in limited amounts (Topping et al., 2007).

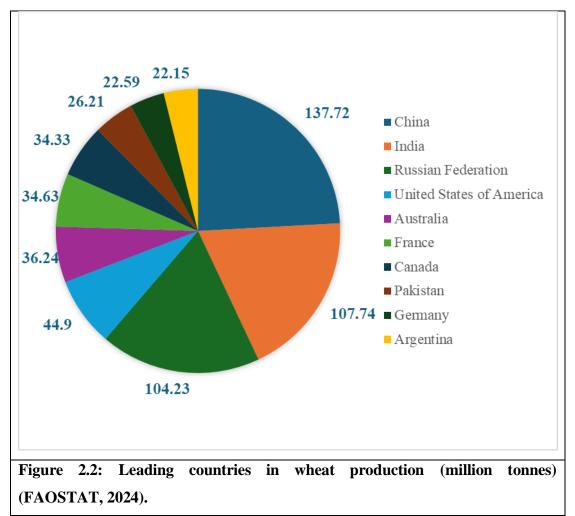
The wheat kernel contains a reservoir of vital nutrients that are necessary for the human diet. The endosperm constitutes approximately 83% of the kernel weight and serves as the primary source of white wheat flour. The kernel endosperm includes the highest proportion of carbohydrates, protein, Fe and various B-complex vitamins, including niacin, riboflavin and thiamine. Bran constitutes approximately 14.5 % of the weight of kernel (**Drankhan** *et al.*, 2003; Blechl *et al.*, 2007; Uauy *et al.*, 2006). Bran is also a constituent of whole-wheat flour and it contains a little quantity of protein, significant amounts of B-complex vitamins, indigestible cellulose material and trace minerals. Wheat germ is the embryonic part of the kernel has a comparatively high amount of protein, lipids and numerous B-complex vitamins (Adams *et al.*, 2002). The endosperm's exterior layers and the aleurone have a greater abundance of vitamins, phytic acid and proteins compared to the interior endosperm. The interior endosperm of the grain contains the majority of protein content and starch. Table 2.1 displays the nutritional makeup of several wheat products. Wheat germ is devoid of salt and cholesterol and it is highly concentrated in nutrients. The grain has high levels of Mg, vitamin E, pantothenic acid, thiamin, phosphorus (P) and niacin. Additionally, it serves as a provider of PABA (para-aminobenzoic acid) and coenzyme Q10 (ubiquinone) (Shewry, 2007; Shewry, 2009). Wheat germ is rich in fibre with an estimated content of 1 gram fibre per tablespoon. A fibre-rich diet can effectively regulate bowel function by minimising constipation. It is often advised for individuals who are at risk of developing heart disease, colon disease and diabetes.



#### (Anonymous, 2024).

Wheat played a vital role in the achievement of green revolution and tremendously aided in transforming our country from a state where we relied on food imports to being self-sufficient. Despite the significant increase in wheat production during the "Green Revolution" (**Evenson, 2003**), the current yield is projected to be just 64% of its maximum potential (**Neumann** *et al.*, **2010**). The current status of wheat production in India and world are presented in figure 2.1 and 2.2. It serves as

an affordable source of both calories and protein (**Zhuang, 2003**). It accounts for around 50 % of daily intake of calories in many developing nations (**Cakmak, 2008; Ghasal** *et al.*, **2017a**).



Wheat is a main component of the human diet and fulfils a high nutrition requirement by supplying over 60% of the protein and calories (**Khalil and Jan**, **2002; Arshad, 2021; Adil et al., 2022**). Recurrent consumption of wheat-based goods might result in deficiency of Zn due to the low wheat grain Zn content and are high in phytate, which reduces the availability of Zn (**Cakmak, 2008**). Given the inter- and intra-specific diversity, it is plausible to identify wheat cultivars that exhibit higher input-use efficiency. The lack of nitrogen and phosphorus, in addition to Zn, has a significant negative effect on the average yield of wheat, leading to a decrease in productivity (**Hotz and Braun, 2004**).

#### 2.2 Status of Zn in soil

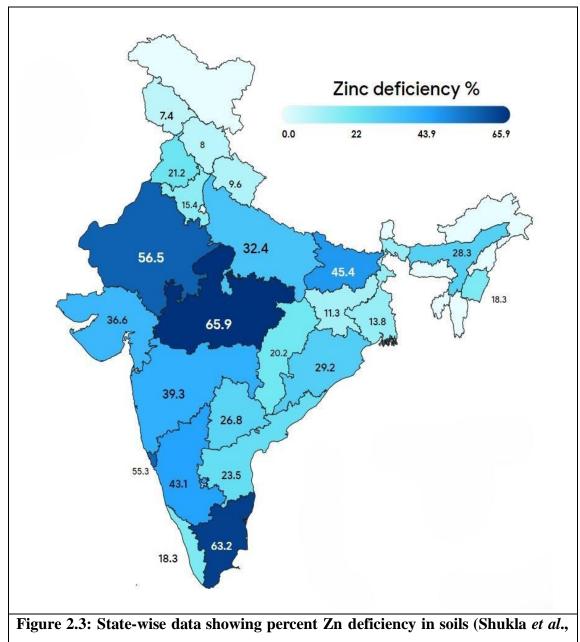
Zinc (Zn) is the 2<sup>nd</sup> most abundant metal and 23<sup>rd</sup> most prevalent element in the lithosphere (earth's crust) and is commonly discovered in polymetallic mines (**Kaur** *et al.*, **2024**). Zinc in the soil occurs in several forms, such as water-soluble,

organically adsorbed, chelated, exchangeable and Zn soil solution. Zinc bioavailability in the soil is significantly impacted by parent material of the soil (Jalal et al., 2024). The natural soil Zn concentration varies from 10 - 300 mg/kg, with a average value of 50 mg/kg. Volcanic rocks have a soil Zn concentration of around 70 - 130 mg/kg, whereas sandstone and carbonated rocks have soil Zn around 16 and 20 mg/kg, respectively (Alloway, 2009). Large-scale analysis of Indian soils showed varying degree of Zn deficiency across the country as shown in figure 2.3. Approximately 50 % of the soil in cereal-growing regions of whole world has limited Zn availability (Alloway, 2008; Cakmak, 2008; Zou et al., 2008). Figure 2.5 shows that Zn shortage continues to be prevalent in soils in tropical regions, as evidenced by the worldwide Zn deficiency index. Plants may access soil Zn in the form of  $Zn^{2+}$ , ZnOH<sup>+</sup> and soluble organic Zn complexes. However, the availability of these forms can be influenced by factors such as high levels of bicarbonates, carbonates, soil pH, content of phosphorous and unbalanced macronutrient fertilisers (Leite et al., 2019).

#### 2.2.1 Soil factors leading to Zn deficiency

Zn availability to plants and the process of leaching might differ depending on the organic matter level and clay content in the soil. The adsorption of Zn to clay particles is not uniform, this is due to the occurrence of high levels of aluminium (Al) and Fe oxides as well as organic materials, which reduce Zn availability. The Zn availability for plant uptake in soil solution is negatively correlated to the pH of soil. Increased soil pH inhibits the release of Zn bound to soil organic matter and clay particles, resulting in a decrease in the availability of  $Zn^{2+}$  ions to the plants (Knijnenburg et al., 2019). Zinc deficiency arises from improper distribution and solubility of Zn in soil solutions, resulting in a worldwide lack of Zn in agricultural plants. This deficiency causes poor growth and deformed leaves, that leads to reduced crop productivity and the production of undernourished grains (Sudha et al., 2017). Zinc is a nutrient that does not move easily in the soil solution. Its ability to move is limited by factors such as high pH of soil, Ca content, intense agricultural methods, excessive use of fertilisers and adsorption to particles of soil colloid. The most common micronutrient deficiency seen in tropical agricultural systems is Zn deficiency, which limits grain production. Tropical soils often experience significant weathering, which can lead to the breakdown of primary minerals found in igneous and sedimentary rocks, resulting in a deficiency of both micro and macro-nutrients in the soil. Rising demand for phosphorus (P) in tropical agriculture poses a significant

obstacle to the availability and functioning of many micronutrients, particularly Zn. This can lead to an increase of reactive oxygen species (ROS) and negatively impact crop development and production (**Alloway**, **2008**).



2016).

#### **2.2.2 Importance of Zinc for plant**

Zinc is an essential nutrient that is necessary for the consistent crop growth and development. A modest quantity of Zn is needed for the appropriate physiological, metabolic and enzymatic processes that occur in plants (**Khatun** *et al.*, **2018**). Zinc serves as a cofactor and structural component for various proteins and enzymes involved in multiple metabolic processes, including glucose synthesis, photosynthesis, metabolism of proteins, pollen production, cell membranes, and pathogenic resistance (**Khatun** *et al.*, **2018**; **Suganya** *et al.*, **2020**). Zinc has an impact on the growth of roots, their structure and plant biomass. It is a beneficial nutrient that improves the process of germination, stability of cell membranes, movement of stomata, photosynthesis, and regulation of respiration. It also interacts with protein synthesis and antioxidant enzymes, that ultimately improves plant health and productivity. However, a lack of Zn can result in reduced protein synthesis and chlorophyll content (**Hassan** *et al.*, **2020**). Zinc homeostasis in the cytosol involves the participation of low molecular weight compounds, chelates, and proteins, as well as the storage of Zn in intracellular compartments. Zinc homeostasis is an intricate process that occurs inside many subcellular compartments, including vacuoles (which provide protection against Zn toxicity), vesicles and cell walls. The mobilisation of Zn from subcellular compartments is essential during periods of shortage, senescence or abscission (**Sinclair** *et al.*, **2012**; **Gupta** *et al.*, **2016**).

Critical concentration of Zn required for the earliest emerged leaves of wheat is 14 mg/kg dry weight (**Brennan, 2001**). At the tillering stage, the critical concentration is 16.5 mg/kg of dry weight while at anthesis it is 7 mg/kg dry weight (**Riley** *et al.*, **1992**). For grains, the critical concentration is 10 mg/kg (**Riley** *et al.*, **1992**; **Rengel and Graham, 1995**). Zinc plays a crucial role in several physiological processes in wheat, such as activation of enzymes, synthesis of proteins, metabolism of nucleic acids and carbohydrates (**Cakmak, 2000; Palmer and Guerinot, 2009**). Zinc finger proteins which regulate cell differentiation and proliferation have Zn as a structural component (**Palmer and Guerinot, 2009**). Application of Zn boosts the concentration of glutenin, globulin, albumin and gliadins thus increases the quality and quantity of proteins in wheat grain (**Liu** *et al.*, **2015**). Moreover, the Zn concentration is strongly associated with the gluten content in the grain (**Peck** *et al.*, **2008**). It also regulates the small proportion of polymeric protein that can be extracted by sodium dodecyl sulphate (SDS) (**Peck** *et al.*, **2008**).

Zinc deficiency impairs protein synthesis by reducing biosynthesis of RNA and/or by causing deformity in ribosomes (Alloway, 2008). Therefore, Zn is necessary for protein synthesis and improves wheat quality. It also mitigates heat stress by sustaining the activity of superoxide dismutase, which helps detoxify ROS and maintains the plant's ability to perform photosynthesis at high temperatures (Cakmak, 2000). Zinc has a crucial role in protecting membrane lipids, proteins, DNA and other essential cellular constituents (Cakmak, 2000). It aids in preserving

the structural integrity of cellular membranes by shielding proteins and lipids from superoxide radicals and other ROS (Cakmak and Marschner, 1988; Rehman *et al.*, 2018).

#### 2.2.3 Importance of Zn for humans

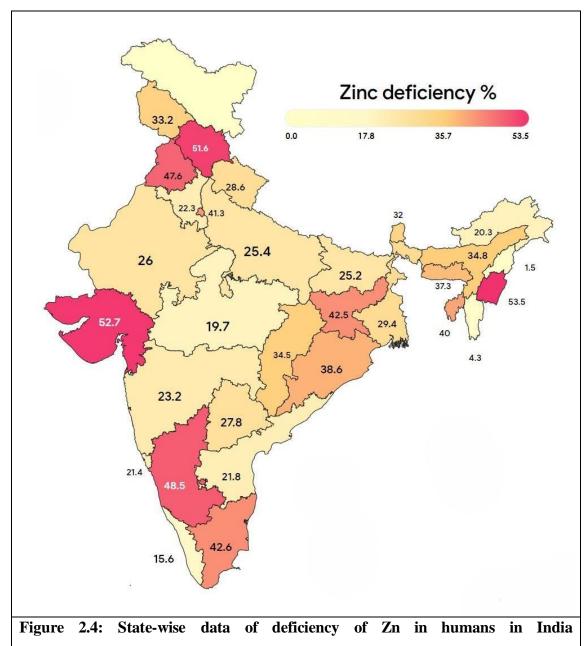
Zinc has a more significant role in critical life functions compared to any other micronutrient. It acts as a catalyst and regulator in many metabolic processes, expression of genes, hormone function, immunological defence systems, and other significant functions (Wessels et al., 2021; Lowe et al., 2024). This makes it extremely important for overall health and growth throughout life (Livingstone, **2015; King, 2011)**. Zinc deficiency is a prevalent kind of micronutrient malnutrition worldwide, yet it is still not well acknowledged. Approximately 17% of global population is at danger of having deficiency of Zn with the rates being as high as 19% in Asian and 24% in African continents (Wessells et al., 2012; Kumssa et al., 2015). Status of Zn deficiency in Indian population is shown in the figure 2.4. The symptoms of Zn deficiency are not specific and their severity depends on factors like duration, age and the existence of other underlying disorders (Shah et al., 2016). It acts as a major constraint for growth in children residing in low to middle income countries (Carducci et al., 2021), leading to stunted growth in both apparently healthy (Nakamura et al., 1993) and malnourished children (Ruz et al., 1997). Severe Zn deficiency not only increases the likelihood of several childhood health problems, but also leads to recurrent infections, cognitive impairment, diarrhea and delay in wound healing. Deficiency of Zn in adolescents and adults adversely impacts reproduction (Bernhardt et al., 2012; Kawade, 2012).

There is an increasing body of research that also establishes a connection between Zn deficiency and an elevated risk of cardiometabolic diseases (**Tamura**, **2021**). Zinc has a significant impact on the release of insulin and the regulation of glucose levels in the body. When Zn level is low, it has been linked to more severe cases of type 2 diabetes mellitus. The status of Zn also has an influence on lipid metabolism, which includes the process of absorbing, synthesising, and metabolising fatty acids. This, in turn, changes the lipid profile in the bloodstream and may potentially raise the risk of cardiovascular disease (**Olechnowicz** *et al.*, **2018**; **Banaszak** *et al.*, **2021**). Zinc has a variety of important roles in vital health functions, particularly in relation to immunocompetence. One example is the process of T-cell activation, which is an important step in the immune response that is necessary for eliminating virus-infected cells and neoplasms. This activation relies on the complex formation between Lck protein and T-cell coreceptor CD8 or CD4, which is reliant on the presence of Zn. Zinc have an imperative role in controlling dephosphorylation and phosphorylation processes involved in signalling of immune cell receptors. Additionally, Zn finger structures are essential for the transcriptional regulation of genes involved in host defence (Wessels *et al.*, 2017). Zinc aids the production of retinoic acid and accumulation of Zn thereby, eliminating harmful bacteria by playing a catalytic role for proteases that break down proteins derived from pathogens and facilitating the conversion of cytotoxic superoxide anions produced during the body's defence mechanism (Gao *et al.*, 2018).

Therapeutic Zn supplementation has been found to have positive benefits in treating several illnesses, such as diarrhea and acute lower respiratory tract infections (**Read** *et al.*, **2019**). These infections are significant causes of illness and mortality in children below age 5 in low to middle income countries. Zinc supplementation in low to middle income countries has a preventive effect, reducing the occurrence of diarrhea, perhaps lowering the frequency of acute infections in lower respiratory tract and decreasing rates of child mortality (**Brown** *et al.*, **2009**; **Lassi** *et al.*, **2016**; **Lassi** *et al.*, **2020**). Zinc has a variety of important functions in metabolic health and it is essential for the production of insulin crystals, which is necessary for their release in to the bloodstream (**Dunn** *et al.*, **2005**). Insulin secretion triggers Zn to signal pancreatic  $\beta$ -cells, preventing excessive release of insulin (**Zhou** *et al.*, **2007**). Additionally, Zn-dependent modulation of phosphorylation enhances insulin receptor sensitivity (**Cruz** *et al.*, **2018**).

Albumin, the extracellular Zn carrier protein, facilitates the transport of free fatty acids present in lipoprotein lipase to the cells during lipolysis. When free fatty acids get attached to albumin, it causes the release of Zn (**Lu** *et al.*, **2012**), which is then absorbed by tissues postprandially (**Lowe** *et al.*, **1998**). Chronic exposure to high blood glucose levels might cause albumin to undergo glycation, which decreases its ability to transport Zn and free fatty acids (**Iqbal** *et al.*, **2018**; **Anguizola** *et al.*, **2013**). The metabolism of essential fatty acids, which plays a critical role in regulating inflammation, is also influenced by variations in Zn consumption (**Hernandez** *et al.*, **2020**). Zinc has an imperative role in responding to oxidative stress, primarily by acting as a catalyst for the activity of enzyme superoxide dismutase (**Wessels** *et al.*, **2017**). It also helps in signalling cellular response to oxidative stress in presence of

ROS when released from metallothionein (Ling *et al.*, 2016). Zinc also controls vascular tone, triggers vasorelaxation (Betrie *et al.*, 2021) and decreases the stiffness of blood clots (Xia *et al.*, 2021).



#### (Pullakhandam et al., 2021).

Subclinical Zn deficiency can lead to a disruption of metabolic function and inflammation, which in turn can contribute to the development of (NCDs) noncommunicable diseases. The recent meta-analyses have indicated that individuals who consume a higher amount of Zn in their diet have a 13% lower likelihood of developing type – 2 diabetes (**Fernandez-Cao** *et al.*, **2019**). Additionally, the use of low-dose Zn supplementation has been shown to significantly decrease the risk factors for type 2 diabetes and cardiovascular disease, such as blood glucose during high fasting, total cholesterol level, insulin resistance and low – density lipoprotein cholesterol levels (**Pompano** *et al.*, 2021).

Fertilizer	Formula	Zn content (%)		
Inorganic fertilizers				
Ammoniated Zn	Zn(NH <sub>3</sub> ) <sub>4</sub> SO <sub>4</sub>	10		
Zinc frits	Fritted glass	10-30		
Zinc sulphate heptahydrate	ZnSO <sub>4</sub> .7H <sub>2</sub> O	22		
Zinc nitrate	$Zn(NO_3)_2.3H_2O$	23		
Zinc oxysulphate	ZnO.ZnSO <sub>4</sub>	20-50		
Zinc sulphate monohydrate	ZnSO <sub>4</sub> .H <sub>2</sub> O	36		
Zinc carbonate	ZnCO <sub>3</sub>	50-56		
Zinc oxide	ZnO	50-80		
Zinc phosphate	$Zn_3(PO_4)_2$	50		
Zinc chloride	ZnCl <sub>2</sub>	50		
Basic zinc sulphate	ZnSO <sub>4</sub> .4Zn(OH) <sub>2</sub>	55		
	Organic fertilizers			
Zinc lignosulphonate	-	5-8		
Zinc polyflavonoid	-	5-10		
Sodium Zn HEDTA	NaZnHEDTA	6-10		
Disodium Zn EDTA	Na <sub>2</sub> ZnEDTA	8-14		
Sodium Zn EDTA	NaZnEDTA	9-13		

Table 2.2: Zn fertilizer sources and their Zn content.

#### 2.2.4 Zinc fertilizers

The Zn fertilizers have multipurpose usage in agriculture. Different forms of fertilizers and their Zn content is shown in table 2.2. Zinc oxide (ZnO) nanoparticles possess the qualities of being non-toxic, environmentally benign and well-suited for biological purposes and they have been employed as fungicides in the field of agriculture as well (Ahmed *et al.*, 2016; Supraja *et al.*, 2018). The efficacy of ZnO nanoparticles in addressing deficiency of Zn in plants has been well known (Munir *et al.*, 2018). The study conducted by Helmy *et al.*, (2020) evaluated the antifungal effects of ZnO nanoparticles when applied before and after harvesting on pomegranate, specifically against *Aspergillus niger* fungus. The researchers discovered that antifungal properties of Zn are a result of its ability to generate ROS

and induce morphological anomalies, such as the distortion of fungal hyphae which leads to the death of fungal cells (**Savi** *et al.*, **2013**). Nanoparticles exert an influence on the proteins in cell wall and traverse the cell membrane by virtue of their physical characteristics. This is facilitated by their capacity to generate ROS, ultimately leading to cellular malfunction (**Arciniegas-Grijalba** *et al.*, **2019**).

Zinc is essential nutrient required for an production of the protochlorophyllides and synthesis of chlorophylls (Faizan and Hayat, 2019). As per reports, nitrogen (N) metabolism is crucial for the chlorophyll synthesis and it enhance the absorption of N by plants indirectly (Dimkpa et al., 2019). The efficiency of photosynthesis is enhanced by the transfer of energy from Zn nanoparticles to chlorophyll, which leads to an increase in pigments involved in photosynthesis and absorption of light (Faizan and Hayat, 2019; Xu et al., 2018). The zinc nanoparticles enhance photosynthetic efficiency by accumulation of proline and bolstering the antioxidant defence mechanism (Faizan et al., 2018). On the other hand, the presence of Zn in cells, organs and tissues increases the activity of carbonic anhydrase, an enzyme that helps in the movement of carbon dioxide (CO<sub>2</sub>) and protons across the biological membranes. Consequently, a deficit of Zn leads to a decline in stomatal conductance, resulting in a lower CO<sub>2</sub> concentration in the intercellular spaces in leaves (Tavallali et al., 2009). The presence of ZnO-NPs in the tissue culture medium has been found to enhance the length of stem and biomass of pomegranates (El-Mahdy and Elazab, 2020). Zinc oxide-nanoparticles have shown involvement in cells of meristem tissues, metabolic pathways and their involvement in the synthesis of precursor of the auxin biosynthesis that plays an important role in determining the rate of plant growth (Faizan and Hayat, 2019).

#### 2.2.4.1 Methods of Zn fertilizer application

Various methodologies are being employed to determine the most suitable approach, form and dosage of Zn fertiliser for application to plants. The predominant methods for Zn fertilisation include soil and foliar treatment as well as seed priming. These approaches serve as effective alternatives which enables the plant to thrive in soils deficient in Zn by enhancing crop performance, growth and yield (**Rehman** *et al.*, **2020**). Zinc fertilisers are administered by several methods, including broadcasting and spraying over the soil, applying in bands, using foliar sprays, priming seeds, or root dipping of transplanting crops (**Rehman** *et al.*, **2020**; **Jalal** *et al.*, **2022**). Various forms of Zn fertilisers exist and each one is utilised according to

its efficacy for crops. There are several types of Zn fertilisers available, such as Zn oxide (ZnO), Zn oxy-sulphate, Zn sulphate (ZnSO<sub>4</sub>), Zn chloride (ZnCl<sub>2</sub>) and Zn-coated urea/superphosphate. Zinc sulphate and Zn oxides are the predominant Zn fertilisers utilised globally. Zinc oxide has had a recent surge in usage, mostly in the field of agriculture, especially in crop production (**Zou** *et al.*, **2012**; **Weisany** *et al.*, **2021**). Zinc fertilisation exhibits a significant impact on soils with low Zn concentration, resulting in increased concentration of Zn in grains compared to soils with high Zn fertility.

#### 2.2.5 Limitation in Zn uptake and transportation

Plants must engage in the processes of absorption, mobilisation and transportation of Zn from the soil to the grains so that they could accumulate Zn. These processes encompass several intricate physiological mechanisms occurring at different levels inside the plant (**Swamy et al., 2016**). If there is enough Zn in the soil, the genotypes should be genetically capable and physiologically efficient to effectively use it. Comprehensive knowledge of the physiological mechanisms underlying the absorption, translocation and distribution of Zn in plants, as well as its regulation and efficient accumulation in grains is crucial for the biofortification of wheat (**Olsen and Palmgren, 2014; Stomph et al., 2009**).

Typically, there are 3 primary obstacles or barriers that restrict the rate of effective Zn accumulation in grains:

1) Barriers from the soil to the roots.

2) Barriers from the roots to the shoots.

3) Barriers that prevent the Zn loading into grains.

The initial step in the mechanism of accumulating Zn into the grains is the absorption of Zn by the roots of the plant. Plant characteristics that influence the Zn absorption by roots include the structure of the roots, number of root hairs, development of crown roots, surface area of the roots, anatomical structures of the roots and the modification of the chemistry of rhizosphere through proton exudation. These protons can alter the pH of the soil, making Zn more soluble and allowing it to move more easily to the surface of the roots (**Rose** *et al.*, **2013**). The Zn availability for all crops is influenced by numerous soil variables like soil texture, pH of soil, content of organic matter, mineralogy, soil microbial population, total sulphur concentration and soluble bicarbonate (**White and Broadley**, **2011; Hacisalihoglu and Kochian**, **2003; Impa and Johnson-Beebout**, **2012**). Therefore, it is crucial to

employ both genetic approaches and agronomic management measures in order to optimize soil health conditions and boost the Zn absorption by plant roots, respectively.

The primary sources of Zn in the grains are direct root absorption, remobilization from vegetative tissues or it may be a combination of both processes (Impa et al., 2013a). Transpiration facilitates the continuous flow of Zn through xylem from the root to the grain, enabling the direct transportation to grains (Krishnan and Dayanandan, 2003). But Zn movement is limited by barriers that hinder the transfer of Zn from the roots to the shoot, as well as the internal allocation and reallocation of Zn with in and among vegetative and reproductive tissues. Consequently, this restriction results in a decreased grain accumulation (Jiang et al., **2008**). Root-to-shoot obstacles for transport of Zn include suberin present in casparian strips and cell wall, sequestration of Zn in vacuoles and cytoplasm and structural differences at the root-shoot junction (Yamaji et al., 2013; Yamaguchi et al., 2012). Zinc absorbed by the roots is transported to various plant parts through the phloem and xylem. There is significant variability in the distribution and redistribution of Zn across different tissues, organs and cells of both the shoot and root (Jiang et al., **2008**). Nevertheless, there are variations among different genotypes in the mechanism of Zn transportation through the xylem & phloem and distribution to different tissues (Jiang et al., 2008). This suggests that through selective breeding, it may be possible to enhance effectiveness of Zn uptake through roots, transportation of Zn from roots to shoots, and the internal distribution of Zn.

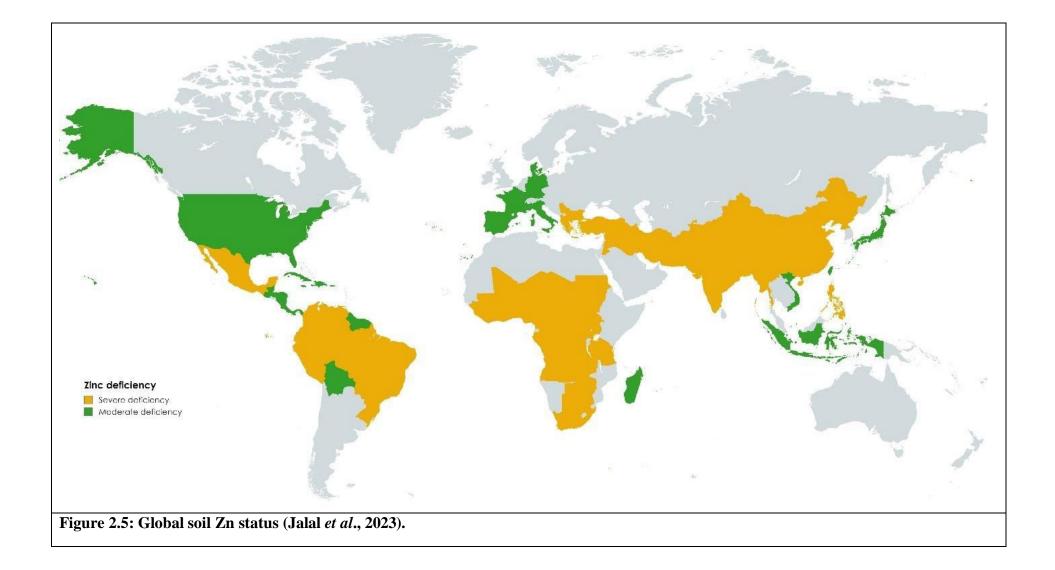
Effective Zn uptake into grains, particularly the endospermic cavity, is crucial for enhancing the Zn content (Waters and Sankaran, 2011). Despite the presence of large quantities of Zn inside the vegetative tissues of rice, the transfer from these tissues to the reproductive parts and ultimately to the grain is restricted. This limitation is caused by the selective movement of Zn through the phloem, which transport Zn from older tissue to newer tissue and eventually to grain (Impa *et al.*, 2013b; Wu *et al.*, 2010). The flag leaf is crucial for photosynthesis and production of grains, has shown minimal impact on grain Zn levels (Sperotto *et al.*, 2013). Wu *et al.*, (2010) revealed Zn transfer from the flag leaf to grain in large quantities. An essential characteristic of Zn efficient genotypes is the continuous delivery of Zn to various tissues through phloem remobilization and translocation to the grains (Yin *et al.*, 2016). Several studies have emphasized the significance of effective Zn

absorption and unobstructed Zn movement across various plant tissues, particularly during the grain filling phase (Ishimaru *et al.*, 2005; Sasaki *et al.*, 2014; Yamaji *et al.*, 2013; Chandel *et al.*, 2010).

#### 2.2.6 Approaches to improve bioavailability of Zn

Zinc fertiliser application through the soil and leaves is extensively researched and widely practiced in tropical regions to improve not only the growth and yield but nutritional status of crops. Efficacy of Zn fertilisation is more accurately determined by the timing of its application. Zinc fertiliser may be administered to the soil before sowing and by foliar spray to the plants at different stages of growth like vegetative, flowering and early grain-filling (Jalal et al., 2022; Zou et al., 2012). Applying Zn to the soil is a commonly used method to boost the concentration of grains Zn and improve crop production. The distribution of Zn applied to the soil is influenced by many transformation pathways, which are dependent on factors such as soil type, crop type, and environmental circumstances. Agricultural soils contain a range of Zn concentration that is determined by factors such as leaching, plant uptake, adsorption, extractability and interaction with the other minerals. Comprehending the efficacy of applying Zn to soil, plant and the environment along with its dispersion and movement in soil solution, is of utmost importance (Barreto et al., 2024; Maqbool and Beshir, 2019). Effectiveness of soil application of Zn is reduced because of its (Zn) limited mobility through soil, adsorption in soil, application of phosphorous fertilisers, and the presence of high levels of oxide and carbonate compounds in tropical agricultural soils. These factors negatively impact Zn nutrition (Malecki et al., 2016).

Applying nutrients directly to the leaves is another approach to tackle the existing difficulties in agricultural production systems. The administration of Zn to plant leaves known as foliar application, has gained significant interest in recent times. This method is favoured for its ability to be quickly absorbed by plants, being cost-effective and its little impact on soil health when addressing plant deficiency (**Dimkpa** *et al.*, 2022). Foliar Zn application during the vegetative phases of crops is absorbed through the leaves and rapidly employed in plant's metabolic activities.



The absorption rate of Zn might vary depending on the method of application to the leaf surface as it enters the leaves through stomata and is then transported to different parts of the plant by apoplastic and symplastic routes. Epidermal cells readily absorb Zn and deliver it to grain tissues through the vascular system. The application of a small amount of foliar Zn can enhance the ability of plants to withstand oxidative stress by protecting and preserving the cell's membrane (**Burman** *et al.*, **2013**; **Hong** *et al.*, **2021**). Furthermore, the process of treating selected crop's seeds with a solution containing Zn is employed to ensure consistent distribution of Zn, a crucial factor in promoting optimal crop growth in Zn deficient soil and challenging environmental circumstances. This procedure is sustainable and effectively accelerates the establishment of seeds and improves their quality, leading to enhanced plant growth.

Various research has documented that priming of seeds enhances their germination and emergence, as well as growth by influencing nutrient absorption, water use efficiency and enhancing protection against both abiotic and biotic stresses (Salam *et al.*, 2022; Rai-kalal and Jajoo, 2021). Immersing roots of crops ready to be transplanted in a solution of Zn has a significant influence on the root profile, stimulating the plant's growth, antioxidant system and photosynthetic efficiency (Faizan *et al.*, 2018). These strategies can enhance the absorption of Zn by the roots and its distribution from the roots to the grains, hence improving agronomic biofortification.

#### 2.3 Phytohormones

Phytohormones may be categorized into ten distinct groups according to their physiological roles. These groups include auxin, gibberellins, cytokinin, ethylene, abscisic acid, brassinosteroids (BR), oligolactones, jasmonic acid, salicylic acid and plant growth retardants. From a functional standpoint, phytohormones have the ability to influence several aspects of plant development such as germination, cell division, growth and differentiation, flowering, fruiting and sexual characteristics of plants (**Campos** *et al.*, 2023; **Zhang** *et al.*, 2023). They also control important aspects of plant's growth as well as development such as division, expansion and differentiation of cells (**Guo** *et al.*, 2024). The use of phytohormones can result in environmental residues, soil and water pollution as well as negative impacts on non-target vegetation. The remnants and discharges of phytohormones possess the capacity to affect the environment and human well-being (**Falchi** *et al.*, 2020).

## Table 2.3: The impact of phytohormones on wheat are as follows.

Growth	Phytohormones can directly influence the development and
and yield	productivity of wheat. They have the ability to enhance root
	development of wheat, facilitate nutrient absorption and optimize
	water utilization, fostering the growth of roots, stems and leaves.
	Meanwhile, phytohormones control the timing of flower initiation
	in wheat and also enhance the amount and weight of grains. The
	functional benefits have a significant positive influence on both the
	production and quality of wheat (Bakhoum et al., 2023).
Quality	Phytohormones exert a significant influence on the quality of wheat.
	They have the ability to control the morphology of wheat caryopsis,
	resulting in enhanced quality parameters such as protein content and
	lysine levels. By employing suitable methods, phytohormones can
	enhance the gluten strength, baking quality and flavour of wheat,
	therefore satisfying customers' demands for wheat-based goods
	(Bakhoum <i>et al.</i> , 2023).
Environ-	Phytohormones are crucial in the implementation of water-conserving
-mental	practices in wheat farming. Phytohormones have the potential to
conservation	improve the efficiency of water – use in wheat by minimising water
	evaporation, waste and greenhouse gas emissions. This has immense
	importance for wheat production in regions with limited water
	availability and phytohormones can efficiently conserve water
	resources and enhance wheat's capacity to thrive in dry situations (Al-
	Huqail <i>et al.</i> , 2023).
Stress	Phytohormones have the ability to augment the stress tolerance of
tolerance	wheat. Studies have demonstrated that phytohormones may
	effectively control the various mechanisms that leads to improved
	disease and insect resistance as well as increased tolerance to drought
	and salt alkalinity (Kholssi et al., 2021). Phytohormones can enhance
	the survival and yield stability of wheat. This helps to mitigate the
	negative impact of harsh conditions on wheat output (Gao et al.,
	<b>2023</b> ). Overall, phytohormones have the ability to enhance the growth
	of wheat roots, while also improving its resistance to diseases and
	posts increasing output and enhancing its shility to withstand stress
	pests, increasing output and enhancing its ability to withstand stress.

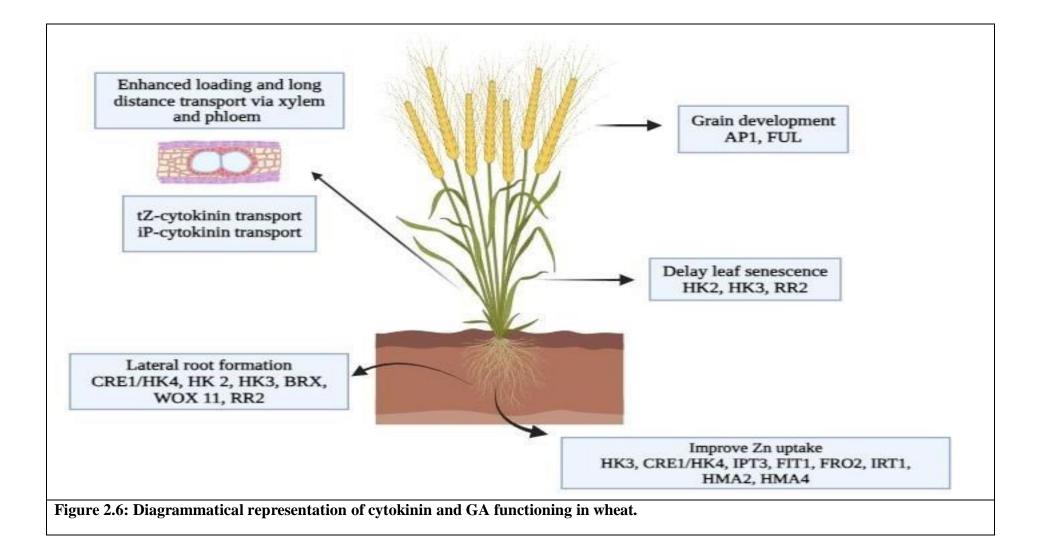
Microbial degradation offers several benefits in the treatment of phytohormone residues. It is a sustainable, secure and effective method to mitigate the influence of phytohormones on the natural environment and human health to some degree (Wang *et al.*, 2016). Hence, the utilization of various phytohormone can enhance agricultural productivity ensuring a good food supply while microbes play a crucial role in mitigating residual issues (Zhang *et al.*, 2022). Phytohormones are widely used in wheat production and primarily modes of application include seed treatment, soil application and foliar spray. Phytohormone have the potential to enhance crop's ability to withstand diseases and pests, as well as their capacity to adapt to challenging conditions and increase crop output (Zahid *et al.*, 2023). These can efficiently stimulate the growth and development of wheat as described in table 2.3. Figure 2.6 represents notable functions of cytokinin and GA in wheat.

#### 2.3.1 Gibberellin or gibberellic acid

Gibberellic acid (GA) is an essential phytohormone that plays several roles, such as it induces germination, promotes cell division, disrupts seed dormancy and increases leaf growth (Niharika *et al.*, 2021). Furthermore, GA is crucial in enhancing plant defence systems against stress by counteracting surplus ROS and enhancing the activities of antioxidant enzymes (Sabagh *et al.*, 2021). Research has shown that the external application of GA to wheat plants can reduce the detrimental effects of saline stress, enhance the uptake of nutrients and increase crop output. It also plays a crucial role in several developmental processes, including seed development, root growth, morphology of leaves, floral formation, pollination and fruit expansion.

#### 2.3.2 Cytokinin

Cytokinins are a prominent group of plant hormones that exert a significant influence on the cell cycle and impact the plant's growth as well as development (**Honig** *et al.*, **2018**). Cytokinins not only stimulate cell division and plant growth, but they also prevent plant ageing by inhibiting the breakdown of nucleic acids, chlorophyll, proteins and other substances in plants. Additionally, they redistribute essential amino acids, hormones, inorganic salts and other compounds to different parts of the plant (**Ullah** *et al.*, **2018**).



Recent studies have demonstrated that cytokinins have the ability to mitigate the harm inflicted on plants by various abiotic stimuli (**Keshishian** *et al.*, **2018; Mi** *et al.*, **2017; Prerostova** *et al.*, **2018**). However, the processes by which cytokinins alleviate stress vary depending on the specific type of stress encountered.

# 2.3.3 Role of phytohormones (Cytokinin and GA) on uptake and translocation of Zn

Cytokinin improves Zn accumulation in the grains. Application of cytokinin upregulates the expression of TaCKX gene that controls the cytokinin concentration. The TaCKX enhances translocation of assimilates during the grain development stages. This leads to increased grain weight, size and nutrient (Zn) content in the grains (**Zarea** *et al.*, **2023**). Cytokinin and GA regulate the activity of proteins like ZIP and HMA (heavy metal ATPase) inside the plant body. These proteins improve root Zn uptake to shoots and then from shoots to grains via different mechanisms (**Gao** *et al.*, **2019; Ren** *et al.*, **2023**).

The ZIP family plays a significant function in the transfer of Zn and the maintenance of metal balance in plants (**Wang** *et al.*, **2017**). Both the ZIP3 and ZIP1 present in Arabidopsis have a role in the Zn uptake from the soil into the roots (**Grotz and Guerinot, 2006**). The ZIP3 has a role in the process of Zn absorption and its distribution during the initial stages of reproductive growth in *Vitis vinifera* (**Gainza-Cortes** *et al.*, **2012**). Overexpression of ZmZIP5 led to higher levels of Fe and Zn in the shoots and roots of maize (*Zea mays* L.) (**Li** *et al.*, **2019**). However, there would be a decrease in the amount of these minerals in the seeds. The expression of OsZIP4 is increased in response to lower Zn levels and controls the Zn transport to the tillering bud in rice plant (**Mu** *et al.*, **2021; Ishimaru** *et al.*, **2005**).In barley (*Hordeum vulgare* L.), three specific proteins, HvZIP8, HvZIP5 and HvZIP3 function as Zn transporters. These proteins play a critical role in maintenance of balance of Zn<sup>2+</sup> ions inside the plant's cells (**Pedas** *et al.*, **2009; Zhang** *et al.*, **2024**).

Heavy-metal ATPase (HMA), also known as P1B-type ATPase, is a very significant transporter of heavy metal ions. It utilises ATP hydrolysis and metal ions to aid in the uptake and movement of heavy metal ions (such as  $Zn^{2+}$ ,  $Cu^{2+}$ ,  $Cd^{2+}$ ,  $Pb^{2+}$  and  $Co^{2+}$ ) across cell membranes (**Huang** *et al.*, **2022; Williams and Mills, 2005**). Currently, the HMA proteins have been studied and characterized in several plants, such as A. thaliana, maize (*Zea mays* L.), wheat (*Triticum aestivum* L.) and rice (*Oryza sativa* L.) at the genomic and molecular level (**Takahashi** *et al.*, **2012; Zhiguo** 

*et al.*, 2018; Williams and Mills, 2005; Zahra *et al.*, 2023). The functioning of HMA genes differs across many plant species, with each gene playing particular tasks. The OsHMA2, AtHMA2, and AtHMA4 play an imperative role in the movement of Zn<sup>2+</sup> ions and Cd<sup>2+</sup> ions from the xylem of the roots to the shoots inside the plant (Fan *et al.*, 2018; Satoh-Nagasawa *et al.*, 2012). Higher concentration of Zn was found in the roots than the shoots in AtHMA4 null mutant, indicating that AtHMA4 is important in xylem loading of Zn (Hussain *et al.*, 2004). The enhanced translocation efficiency of Zn from roots to shoots in *Arabidopsis halleri* is attributed to the increased expression of AhHMA4 in the roots (Verret *et al.*, 2004). In rice, OsHMA1 and OsHMA2 are involved in transportation of Zn during the processes of flowering and seed development (Suzuki *et al.*, 2012; Williams and Mills, 2005; Huang *et al.*, 2022; Khan *et al.*, 2024).

# CHAPTER – III

# **MATERIALS AND METHODS**

The research trial was performed at the agriculture farm, School of Agriculture, Lovely Professional University, Phagwara, Punjab during the rabi seasons of 2021-22 and 2022-23. Geographically, the farm is placed at 232 meters above mean sea level and latitude 31.25°N and a longitude 75°E. The meteorological data for season 2021-22 and 2022-23 is presented in figure 3.1 and figure 3.2. Jalandhar has a semiarid climate where both winters and summers are extreme. The climate of experimental site is characterized by sub-tropical and semi-arid type of climate with hot and dry summer from April to June, hot and humid from July to September and cold winter from November to January.

Soil character	Depth of soil (0-15 cm)	Rating	Instrument/Methods used
Mechanical Analys	sis		
Silt (%)	12.0		International pipette method ( <b>Piper</b> ,
Clay (%)	13.8		2019)
Sand (%)	74.2		
Textural class	Sandy loam		
Chemical analysis			
рН	7.4	Normal	Beckman's glass electrode pH meter in soil-water suspension of 1:2 ratio ( <b>Jackson, 2005</b> )
Electrical conductivity (ds/m) at 25° C	0.266	Normal	Solubridge conductivity meter in soil- water suspension of 1:2 ratio ( <b>Jackson</b> , <b>2005</b> )
Organic carbon (C) (%)	0.470	Medium	Rapid titration method by Walkley and Black ( <b>Piper, 2019</b> )
Available nitrogen (N) (kg/ha)	220	Low	Modified alkaline kMnO <sub>4</sub> method (Subbiah and Asija, 1956)
Available phosphorus (P) (kg/ha)	14.6	Medium	0.5 N NaHCO <sub>3</sub> extractable phosphorus by Olsen's Method ( <b>Olsen</b> <i>et al.</i> , <b>1954</b> )
Available potassium (K) (kg/ha)	140	Medium	1 N ammonium acetate extractable potassium (K) ( <b>Jackson, 2005</b> )
DTPA extractable Zn (mg/kg)	0.47	Low	DTPA extract (Microwave plasma atomic emission spectroscopy)

Table 3.1: Physical and chemical properties of the soil before trial.

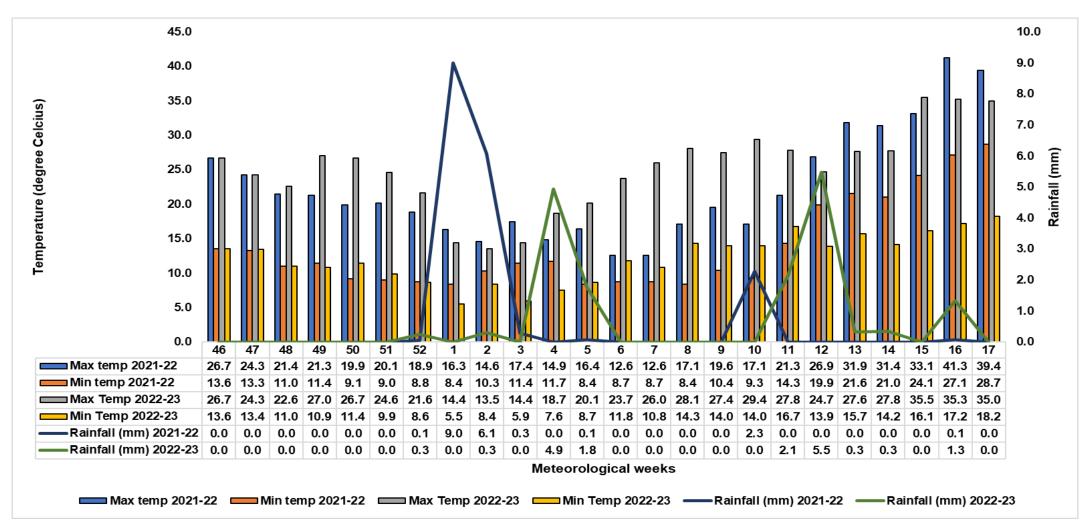


Figure 3.1: Weekly mean meteorological data recorded for season 2021-22 and 2022-23.

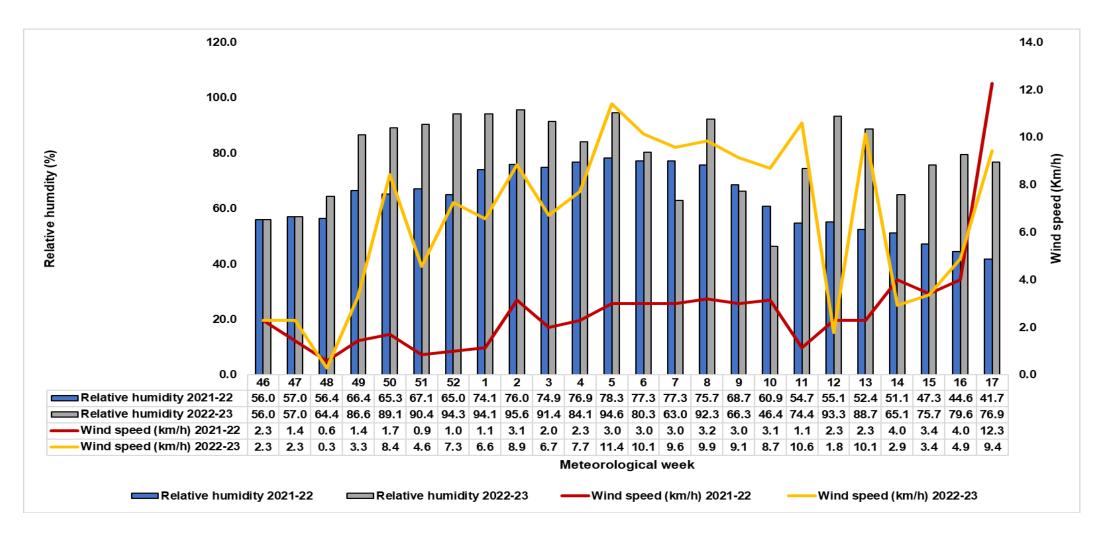


Figure 3.2: Weekly mean meteorological data recorded for season 2021-22 and 2022-23.

The maximum and minimum temperatures show considerable fluctuation during both summer and winter season. Maximum temperature goes above 47°C during summer, while temperature below 4°C accompanied by frosty spells is quite common during the months of December and January. The average annual rainfall is about 500-750 mm, most of which is received during the monsoon period from July to September. However, a few showers were received during winter season also. Winter season (December to March) with temperature ranges from 0°C to 15°C. The soil texture of experimental site was sandy loam, soil pH of 7.4 and electric conductivity (EC) of 0.266 dS/m. The readily oxidisable carbon (C) was 10.5 g and soil had 1.67 kg/ha of DTPA extractable zinc/kg soil, 220 kg of alkaline KMnO<sub>4</sub> hydrolysable N, Olsen P was 14.6 kg and ammonium acetate extractable K was 140 kg per ha.

#### **3.1 Experiment details**

The two wheat varieties, i.e., PBW 725 (timely sown variety) and PBW 752 (late sown variety) were used and their seeds were obtained from Punjab Agricultural University, Ludhiana, Punjab, India. Zinc (Zn) was applied via different modes, viz., no Zn application (Zn<sub>0</sub>), soil application of Zn @ 62.5 kg/ha (Zn<sub>62.5</sub>) and soil application of Zn @ 31.25 kg/hectare + foliar spray of 0.5% ZnSO<sub>4</sub>.7H<sub>2</sub>O (Zn<sub>31.25,F</sub>). Soil application of Zn (ZnSO<sub>4</sub>.7H<sub>2</sub>O) was done before sowing. Three (3) zinc foliar applications were done at 40, at 80 and at 120 days after sowing (DAS). The concentration of Zn solution which was applied to plant's foliage was 0.05 %. Concentrations of plant growth regulators (PGR) used for experiment were no hormone (H<sub>0</sub>), 10 mg/L GA (H<sub>1</sub>), 10 mg/L cytokinin (H<sub>2</sub>) and 5 mg/L GA + 5 mg/L cytokinin (H<sub>3</sub>). The source of gibberellic acid and cytokinin used in the experiment was GA3 and benzyl aminopurine (BAP), respectively. Hormones were applied foliarly and it was done instantaneously after the Zn foliar spray.

The design of experiment was split-split plot with each treatment having with 3 replications. The varieties were kept in main plot, the sub plots were assigned to Zn application methods and hormones were applied in the sub-sub plots. Two main plots ( $V_{725}$  and  $V_{752}$ ) were divided into three sub plots (Zn<sub>0</sub>, Zn<sub>62.5</sub> and Zn<sub>31.25,F</sub>). Each sub plot was further divided into 4 sub-sub plots (H<sub>0</sub>, H<sub>1</sub>, H<sub>2</sub> and H<sub>3</sub>).

#### 3.2 Layout of the experiment

Layout of experiment area is displayed in figure 3.3.

## 3.3 Agronomic practices

Following agronomic practices were followed for raising the wheat varieties during *rabi* season of 2021-22 and 2022-23.

 Table 3.2: Detail of the experiment

A. Main plot detail											
	Varieties										
V <sub>725</sub>	:	PBW 725									
V <sub>752</sub>	:	PBW 752									
B. Sub plot detail											
		Zinc levels (kg/ha)									
Zn <sub>0</sub>	:	No application of Zn									
Zn <sub>62.5</sub>	:	Soil application at 100% RDF (62.5 kg/hectare)									
Zn <sub>31.25,F</sub>	:	50% RDF (31.25 kg/hectare) soil application +									
		foliar spray of $ZnSO_4$ (0.5%) solution									
C. Sub – Sub plot	detail										
		Growth hormone Levels (ppm)									
Ho	:	No application of hormone									
$H_1$	:	10 mg/L foliar spray of GA									
H <sub>2</sub>	:	10 mg/L foliar spray of cytokinin									
H <sub>3</sub>	:	5 mg/L foliar spray of GA + 5 mg/L foliar spray of cytokinin									
*RDF of Zn	:	Recommended dose of fertilizer (62.5 kg									
		ZnSO <sub>4</sub> .7H <sub>2</sub> O/ha)									
Treatment	:	$2 \times 3 \times 4 = 24$									
Replications	:	3									
Total no. of plots	:	$24 \times 3 = 72$									
Design	:	Split – split plot design (SPD)									
Gross plot size	:	$3.4 \times 2.4 \ m^2$									
Net plot size	:	$3 \times 2 m^2$									
Seed rate	:	100 kg/ha									
Row-row Spacing	:	20 cm									
Total area	:	770 m <sup>2</sup>									
Sowing time	:	29 Nov, 2021 and 2022 for V725 (timely sown variety)									
		10 Dec, 2021 and 2022 for $V_{752}$ (late sown variety)									

Treatment
$V_{725} Zn_0 H_0$
$V_{725} Zn_0 H_1$
$V_{725} Zn_0 H_2$
$V_{725} Zn_0 H_3$
V725 Zn62.5 H0
V725 Zn62.5 H1
V725 Zn62.5 H2
V725 Zn62.5 H3
V725 Zn31.25,F H0
V <sub>725</sub> Zn <sub>31.25,F</sub> H <sub>1</sub>
V <sub>725</sub> Zn <sub>31.25,F</sub> H <sub>2</sub>
V <sub>725</sub> Zn <sub>31.25,F</sub> H <sub>3</sub>
$V_{752} Zn_0 H_0$
$V_{752} Zn_0 H_1$
V752 Zn0 H2
V <sub>752</sub> Zn <sub>0</sub> H <sub>3</sub>
V752 Zn62.5 H0
V752 Zn62.5 H1
V752 Zn62.5 H2
V752 Zn62.5 H3
V752 Zn31.25,F H0
V752 Zn31.25,F H1
V752 Zn31.25,F H2
V752 Zn31.25,F H3

**Table 3.3: Treatment details** 

#### **3.3.1** Field preparation

*Rauni* (Pre-sowing irrigation) was done on November 6 during 2021-22 and 2022-23 after removing the weeds manually from the field. Field was prepared on November 27 with two ploughings followed by planking for sowing of early sown wheat variety PBW 725. While, for late sown variety PBW 752, *rauni* was given on 20 November, followed by field preparation on 8 December.

# **3.3.3** Fertilizer application

Fertilizers were applied according to the respective treatment. The phosphorus fertilizer used was diammonium phosphate (DAP). Nitrogen which was added through application of DAP was deducted from the total N required and rest was supplied through urea (46% N). It was then applied in three (3) splits i.e.,  $1/3^{rd}$  at sowing,  $1/3^{rd}$  at  $1^{st}$  irrigation and leftover  $1/3^{rd}$  at the time of  $2^{nd}$  irrigation. Potassium requirement of crop was fulfilled by application of muriate of potash (MOP) at the recommended dose of 50 kg/ha.

S.N.	e 3.4: Calendar of operation Operation		1-22	2022-23		
0.14.	Operation	<b>V</b> 725	V <sub>752</sub>	V <sub>725</sub>	<b>V</b> 752	
1	Preparation of field layout	27/11/21	8/12/21	27/11/22	8/12/22	
2	Application of basal dose of N, P and K fertilizers	27/11/21	8/12/21	27/11/22	8/12/22	
3	Sowing	29/11/21	10/12/21	29/11/22	10/12/22	
4	Pre-emergence herbicide application (Pendimethalin)	30/11/21	11/12/21	30/11/22	11/12/22	
5	Hand weeding	17/12/21	10/1/22	17/12/22	5/1/23	
6	Post-emergence herbicide application (sulfosulfuron + metsulfuron)	-	-	30/12/22	12/1/23	
7	Insecticide application (thiamethoxam)	13/3/22	13/3/22	13/3/23	13/3/23	
8	Irrigation (1 <sup>st</sup> ) (2 <sup>nd</sup> ) (3 <sup>rd</sup> ) (4 <sup>th</sup> )	20/12/1 Rainfall Rainfall 15/3/22	Rainfall Rainfall 15/3/22 -	20/12/22 20/1/23 21/2/23 Rainfall	7/1/23 21/2/23 Rainfall -	
9	Zn foliar spray (1 <sup>st</sup> ) (2 <sup>nd</sup> ) (3 <sup>rd</sup> )	10/1/22 21/2/22 30/3/22	19/1/22 27/2//22 7/4/22	10/1/23 20/2/23 30/3/23	19/1/22 27/2//22 7/4/22	
10	Hormone foliar spray (1 <sup>st</sup> ) (2 <sup>nd</sup> ) (3 <sup>rd)</sup>	11/1/22 22/2/22 31/3/22	20/1/22 28/2//22 8/4/22	11/1/22 22/2/22 31/3/22	20/1/22 28/2//22 8/4/22	
11	Harvesting	18/4/22	18/4/22	18/4/23	18/4/23	
12	Threshing	20/4/22	20/4/22	20/4/23	20/4/23	

 Table 3.4: Calendar of operation

### 3.3.4 Weed control

Stomp 30 EC (pendimethalin) was used as a pre-emergence herbicide within 2 DAS. Manual weeding was done to remove the weeds at 35 DAS. The weeds in wheat crop rows were eradicated by the use of khurpa. A spray of Total/ Markpower 75 WG (sulfosulfuron + metsulfuron) was also done to control *Phalaris minor*.

#### **3.3.5** Insecticide application

One spray of Actara/ Taiyo (thiamethoxam) 25 WG @ 50 g/ha was done in March, to reduce aphid population.

# **3.3.6 Irrigations**

Irrigations were applied to all the plots according to the recommendations of Punjab Agricultural University, Ludhiana, Punjab, India. The first irrigation was applied at 22 DAS, i.e., at CRI (crown root initiation) stage. Subsequent irrigations were done as per crop requirement in accordance with rainfall received.

#### 3.3.7 Harvesting and threshing

Crop was manually harvested by using sickle on maturity (18<sup>th</sup> April) during both the seasons. Bundles were weighed and threshing was done after sun drying for four days. Produce of all the plots were separately threshed by labour and weight of the grains were recorded after cleaning. The weight of straw was separately taken by deducting weight of the grains from weight of bundle.

#### **3.4 Observations recorded**

#### 3.4.1 Plant height

Height of plant was measured using metre scale at 30, at 60, at 90 and at 120 DAS and at harvest. Average of five plants was taken by using metre scale and expressed in centimetre (cm).

# 3.4.2 Tiller count

Total number of tillers were counted at 30, at 60, at 90 and at 120 DAS. Counting of total quantity of tillers was done from 25 cm row length of 4 different rows of each plot and then changed to count of total tillers/metre<sup>2</sup>.

# 3.4.3 Leaf area index (LAI)

Leaves were harvested from randomly selected plants from 25 cm row length of each plot at 30, at 60, at 90 and at 120 DAS. Then, the leaf area meter was used to measure the leaf area. Thereafter, LAI was estimated by using the below mentioned formula where the ground area  $(1m^2)$  has been converted into cm<sup>2</sup>.

LAYOUT											
	REPLIC	ATION 3			REPLIC	ATION 2			REPLIC	ATION 1	
	V	725			V	725			V	725	
7 n	$H_{l}$	H <sub>3</sub>		Zn	H <sub>2</sub>	$\mathbf{H}_{0}$		Zn <sub>0</sub>	$\mathbf{H}_{2}$	${ m H}_3$	
Zn <sub>31.25,F</sub>	H <sub>2</sub>	${ m H}_0$		Zn <sub>62.5</sub>	H <sub>1</sub>	H <sub>3</sub>		ZII0	$\mathbf{H}_{1}$	$H_0$	
Zn <sub>0</sub>	$\mathbf{H}_{0}$	H <sub>2</sub>		7n	$\mathbf{H}_{0}$	H <sub>1</sub>		7n	$\mathbf{H}_{0}$	$H_2$	
<b>Z</b> 11 <sub>0</sub>	${ m H}_3$	$\mathbf{H}_{1}$		Zn <sub>31.25,F</sub>	H <sub>3</sub>	H <sub>2</sub>		Zn <sub>62.5</sub>	$\mathbf{H}_{1}$	H <sub>3</sub>	
Zn <sub>62.5</sub>	$H_2$	$\mathbf{H}_{0}$		Zn <sub>0</sub>	$H_1$	$\mathbf{H}_{0}$		Zn <sub>31.25,F</sub>	$H_3$	$\mathbf{H}_{0}$	
21162.5	H <sub>1</sub>	H <sub>3</sub>		<b>ZH</b> <sub>0</sub>	H <sub>2</sub>	$H_3$			$\mathbf{H}_{1}$	H <sub>2</sub>	
	V	752	Irrigation channel		V <sub>752</sub>		Irrigation channel		V	752	43 m
7n	$\mathbf{H}_{0}$	H <sub>2</sub>		Zn <sub>62.5</sub>	H <sub>3</sub>	H <sub>2</sub>		Zn <sub>0</sub>	$H_2$	$\mathbf{H}_{0}$	
Zn <sub>31.25,F</sub>	$H_1$	H <sub>3</sub>		2162.5	H <sub>1</sub>	$\mathbf{H}_{0}$		Z110	${ m H}_3$	$\mathbf{H}_{1}$	
Zn <sub>62.5</sub>	$H_3$	$H_2$		Zn <sub>o</sub>	$\mathbf{H}_{0}$	$H_3$		7n	$\mathbf{H}_{1}$	H <sub>3</sub>	
21162.5	$\mathbf{H}_{0}$	H <sub>1</sub>		<b>Σ</b> Π <sub>0</sub>	H <sub>2</sub>	$\mathbf{H}_{1}$		Zn <sub>31.25,F</sub>	$H_2$	$\mathbf{H}_{0}$	
Zn <sub>0</sub>	H <sub>2</sub>	$\mathbf{H}_{0}$		7n	$H_1$	$\mathbf{H}_{0}$		7n	H <sub>3</sub>	H <sub>2</sub>	
<b>ZH</b> <sub>0</sub>	$H_1$	H <sub>3</sub>		Zn <sub>31.25,F</sub>	$H_2$	${ m H}_3$		Zn <sub>62.5</sub>	$\mathbf{H}_{0}$	$H_1$	
					17	m			5		

Figure 3.3: Experimental Layout.

Leaf area index (LAI) = 
$$\frac{\text{Leaf area of plant } (cm)^2}{\text{Ground area of plant } (cm)^2}$$

# 3.4.4 Dry matter accumulation

The shoot portion of plants was taken from 25 cm row length at 30, at 60, at 90 and at 120 DAS. The plant samples were firstly dried in sun for 2 days. Thereafter, they were oven dried for 72 hours at a temperature of 65°C to evaporate water content. The weight of dried plants was recorded and accumulation of dry matter was expressed in grams per metre square.

#### 3.4.5 Effective tillers

The counting of effective tillers was done at crop maturity. Tillers having the ear were counted from 25 cm row length of 4 different rows of each plot. They were later expressed in effective tillers/metre<sup>2</sup>.

#### 3.4.6 Spike length

From each plot, 5 ears were chosen randomly and their length was measured using metre scale from base of first spikelet to tip of last spikelet and then the mean length was calculated.

# 3.4.7 Number of grains/spike

From each plot, 5 ears were selected randomly which were threshed manually. After counting of grains, the mean number of grains/spike were recorded.

#### 3.4.8 Thousand (1000) grain weight (grams)

From each plot, thousand (1000) grains were collected and their weight (in grams) were recorded and expressed as 1000-grain weight.

#### 3.4.9 Grain yield

From each plot, bundles of wheat varieties were separately threshed and grains were gathered in different bags. After cleaning, weighing of grains was done and the grain yield was noted.

# 3.4.10 Straw yield

Bundles of plants from each plot were sun-dried in open space for four days after harvesting. Later on, the bundles were threshed. After threshing, the grain weight was deducted from total weight of bundle and weight of remaining plant parts was considered as straw yield.

#### 3.4.11 Harvest index (HI)

Harvest index was estimated of each plot by using the formula mentioned below.

Harvest index = {Grain yield / Biological yield}

#### 3.5 Quality parameters

# 3.5.1 Zinc content in different plant parts

The Zn content of both the wheat varieties was calculated by following the method of **Isaac and Kerber (1971)**. Double distilled water was used to wash the samples to remove contaminations.

# Procedure

Wheat sample (0.1g) of different plant parts was taken in powder form and kept in conical flask of 100 ml. Ten ml (10 ml) mixture of tri-acid (HClO<sub>4</sub>:H<sub>2</sub>SO<sub>4</sub>: HNO<sub>3</sub> in 1:4:10) was added into it. The samples were then placed on hot plate for digestion at 100°C. Twenty ml (20 ml) of 0.1 N HCl was added after the digestion. By adding double distilled water, final volume of sample was made 50 ml. The samples were then filtered by using filter paper of Whatman No. 41. One ml (1 ml) of filtrate was taken for the determination of Zn by using atomic absorption spectrometer.

# 3.5.2 Protein content in grains (Bradford Method)

The grain protein content in wheat was estimated by following the method of **Bradford (1976)**. The grains were washed by using double distilled water to remove contamination and then grinded to form powder.

#### **Extraction buffer**

Extraction buffer was prepared from NaCl (50mM), EDTA (5mM) and NaH<sub>2</sub>PO<sub>4</sub> (25mM). The solutions of above-mentioned reagents (100 ml each) were separately prepared and mixed in a conical flask of 1000 ml. The pH of the buffer was maintained at 7.2. One gram (1g) of powdered grain sample was taken. Powder was mixed with cold extraction buffer (5 ml). Mixture was then centrifuged at 10,000 rpm at  $4^{\circ}$ C for 20 minutes.

#### **Bradford dye**

One hundred grams of Coomassie-Brilliant blue G-250 was mixed with 50 ml of 95% ethanol. Hundred ml (100 ml) of ortho-phosphoric acid (85 %) was added to the above solution and the final volume was made 1000 ml by the addition of double distilled water. After filtration, the dye was stored at 4°C in dark coloured bottle (amber bottle).

#### **Standard solution**

Stock solution having the strength of 1 mg/ml was prepared by adding 10 mg of bovine serum albumin (BSA) in 10 ml of double distilled water.

#### Standard curve preparation

A set of 5 test tubes was prepared having 0.2, 0.4, 0.6, 0.8, and 1.0 ml of BSA standard solution and addition of distilled water was done to make final volume 1 ml. Five ml (5 ml) of Bradford dye was added into each tube and mixed by using vortex mixture. After 10-30 minutes, absorbance was recorded at 595nm by using spectrophotometer. Blank was the test tube containing 1 ml of distilled water. The standard curve was prepared by plotting the absorbance against BSA concentration.

# Procedure for unknown sample

One ml aliquot of unknown sample was taken in a test tube and 5 ml of bradford dye was added into it. Proper mixing was done by using vortex mixture. After 10-30 minutes, the absorbance was recorded at 595nm by using spectrophotometer. The absorbance recorded was used to calculate the final protein content.

# 3.5.3 Phytic Acid

Phytic acid in wheat grains was estimated by using following a method described by **Harland and Oberleas (1977)**.

# **Reagents:**

#### Sodium phytate

A series of standard solutions were prepared containing 5-40 mg/L phytic acid.

#### FeCl<sub>3</sub>.6H<sub>2</sub>O (0.03%)

Ferric chloride [FeCl<sub>3</sub>.6H<sub>2</sub>O] (0.015g) was mixed in distilled water and final volume was made upto 50 ml.

# Sulfosalicylic acid (0.3%)

Sulfosalicylic acid (0.15 g) was mixed in distilled water and final volume was made up to 50 ml.

#### Hydrochloric acid (HCl) (0.65 N)

Hydrochloric acid (31.55 ml) diluted by distilled water and final volume was made upto 500 ml using distilled water.

#### **Preparation of standard curve**

A range of standard solutions were prepared by dissolving 50 to 200 mg of phytic acid in 1000 ml of distilled water. Distilled water was used as a blank. Three ml (3 ml) of above prepared standard solution was pipetted into centrifuge tubes of 20 ml capacity. One ml (1 ml) of Wade reagent (0.03% FeCl<sub>3</sub>.6H<sub>2</sub>O and 0.3% sulfosalicylic acid) was added to each tube. The tubes were then centrifuged at 4000 rpm for duration of 10 mins. The absorbance was recorded at 500 nm by using a spectrophotometer. Absorbance of standard solution was subtracted from absorbance of blank. A standard curve was plotted by using the values of difference in absorbance against the concentration of standard solution.

#### Procedure for unknown sample

To determine the phytic acid content, 1 g of powdered wheat grains were kept in centrifuge tubes and extraction of phytic acid was done by using 2.4% HCl. One ml (1 ml) of Wade reagent was added in to it. The tubes were centrifuged for 10 mins at 4000 rpm. The absorbance was recorded at 500 nm by using spectrophotometer. The recorded absorbance was used to calculate the phytic acid content of unknown sample.

# 3.6 Soil Zn content

The soil Zn content was estimated using the DTPA extraction method by Lindsay and Norvell (1978).

#### Diethylenetriamine penta-acetic acid (DTPA) extractant

The extractant was prepared by dissolving 1.97g of DTPA in distilled water. Then, triethanolamine (14.92g) and calcium chloride dihydrate (1.47g) was added to it. The pH of the extract was adjusted to 7.3. It was later diluted to 1 litre by using distilled water.

#### Procedure

Ten grams (10g) of air-dried soil (< 2mm) was taken in centrifuge tube and 20 ml of the DTPA extractant was added to it. The mixture was agitated for 2 hours on a mechanical shaker. The suspension was then filtered using filter paper (Whatman No. 42). Atomic absorption spectrophotometer (AAS) was used to analyse Zn from the filtrates.

#### **3.7 Economics**

Economics of experiments of both the years was estimated using prevailing input prices and yield of the crop. By multiplying the wheat grain yield with its minimum support price, gross return was calculated. The crop's cost of cultivation was estimated by using the inputs used. Additional cost involved and returns obtained under different treatment combination was worked out based on the market rates of all applied inputs throughout experiment on per hectare basis. The benefit :cost (B:C) ratio is computed by using following formula: -

$$B: C \ ratio = \frac{Gross \ return}{Total \ cost}$$

### **3.8 Statistical Analysis**

The data of all the observations was statistically analyzed by analysis of variance (ANOVA) using RStudio. Least significant difference (LSD) test was used for multiple comparisons at  $p\leq 0.05$ .

Source of variation	Formula	Degree of freedom
Replications	(r – 1)	2
Main plot (A)	(a – 1)	1
Error (A)	(r – 1) (a – 1)	2
Sub Plot (B)	(b – 1)	2
A * B	(a – 1) (b – 1)	2
Error (B)	a (r – 1) (b – 1)	8
Sub-sub plot (C)	(c – 1)	3
A * C	(a – 1) (c – 1)	3
B * C	(b – 1) (c – 1)	6
A * B * C	(a – 1) (b – 1) (c – 1)	6
Error (C)	ab (r – 1) (c – 1)	36
Total	(rab – 1)	71

# **CHAPTER IV**

### 4.1 Periodic plant height

The impact of different treatments on periodic plant height is shown in table 4.1. The results recorded for different wheat varieties showed significant increase in height of plant at all phases of growth in 2021-22 season. Maximum plant height was recorded at harvest for  $V_{725}$  (101.0 cm) followed by  $V_{752}$  (81.8 cm). As regard to Zn application methods, a significant effect on plant height was recorded at all phases of growth apart from at 30 DAS. Maximum height of plant was recorded at harvest under  $Zn_{31.25,F}$  (92.9 cm) that was statistically at par with  $Zn_{62.5}$  (91.8 cm) and significantly higher than  $Zn_0$  (89.4 cm). Among the phytohormones, all levels significantly enhanced plant height at all growth phases apart from at 30 DAS. The level  $H_1$  showed maximum plant height (92.8 cm) at harvest followed by  $H_3$  (91.8 cm),  $H_2$  (90.8 cm) and  $H_0$  (90.0 cm). Overall, the treatment combination  $V_{725} + Zn_{31.25,F} + H_1$  was most efficient in enhancing the plant height and resulted in maximum (104.0 cm) at harvest.

During season 2022-23, there was a significant varietal difference pertaining to the height of plant throughout all phases of growth. The plant height was maximum at harvest for V<sub>725</sub> (104.9 cm) followed by V<sub>752</sub> (88.7 cm). Pertaining to different Zn application methods, plant height was significantly affected at all phases of growth apart from at 30 DAS. Maximum height of plant was recorded under Zn<sub>31.25,F</sub> (99.3 cm) at harvest followed by Zn<sub>62.5</sub> (96.5 cm) and Zn<sub>0</sub> (94.6 cm). The application of various plant hormones significantly enhanced height of plant at all phases of growth apart from at 30 DAS. Maximum height of plant was recorded at harvest under H<sub>1</sub> (99.1 cm) which was significantly higher than H<sub>3</sub> (97.5 cm), H<sub>2</sub> (95.9 cm) and H<sub>0</sub> (94.7 cm). Overall, the treatment combination V<sub>725</sub> + Zn<sub>31.25,F</sub> + H<sub>1</sub> was most efficient in enhancing the plant height and maximum (110.2 cm) was recorded at harvest. The interaction effect among Zn and hormones at harvest on plant height during 2022-23 season is presented in table 4.1a. Maximum plant height was recorded under Zn<sub>31.25,F</sub>+H<sub>1</sub> (101.8 cm) and minimum was recorded for Zn<sub>0</sub>+H<sub>0</sub> (90.6 cm).

Plant height is an important parameter of wheat crop that affects its growth and development over a period and increases with the advancement of age. The varietal difference was evident with respect to plant height. Height of wheat plant might also be enhanced due to the improved enzymatic activity by Zn application. Zinc is also involved in activation of tryptophan synthetase that upregulated the tryptophan synthesis.

Plant height (cm)												
	2021- 22	2022- 23										
Treatments		DAS		DAS		DAS		0 DAS	i an marent	arvest		
Variety												
V725 (PBW 725)	15.6	16.5	41.4	42.4	51.8	64.2	83.7	92.9	101.0	104.9		
V752 (PBW 752)	10.2	11.2	35.8	37.8	45.9	56.1	72.0	76.3	81.8	88.7		
SEm (±)	0.1	0.3	0.3	0.4	0.5	0.8	0.1	0.6	0.6	1.1		
CD at p ≤ 0.05	0.8	2.1	1.7	2.6	3.1	4.7	0.8	3.4	3.3	6.4		
Zinc application modes												
Zn₀ (No application)	12.3	13.5	37.0	37.2	46.4	57.2	75.9	81.6	89.4	94.6		
Zn <sub>62.5</sub> (Soil)	13.3	14.2	38.9	40.9	49.2	60.9	78.4	85.7	91.8	96.5		
Zn31.25,F (Soil+Foliar)	13.0	13.9	39.9	41.2	50.8	62.4	79.3	86.5	92.9	99.3		
Zn SEm (±)	0.4	0.5	0.4	0.4	0.6	0.7	0.2	0.6	0.5	1.1		
Zn (CD at p ≤ 0.05)	NS	NS	1.4	1.3	1.8	2.1	0.6	2.0	1.5	3.6		
V x Zn SEm (±)	0.6	0.6	0.6	0.6	0.8	0.9	0.3	0.9	0.6	1.6		
V x Zn (CD at p≤ 0.05)	NS											
<b>F</b> =7	8 =		Н	ormone :	applicati	on						
H <sub>0</sub> (No	12.9	13.8	37.8	38.4	47.0	58.3	76.2	82.8	90.0	94.7		
application) H1 (10 mg/L GA)	12.9	14.0	39.3	41.2	50.4	62.1	79.5	86.6	92.8	99.1		
H <sub>2</sub> (10 mg/L	12.7	13.8	38.2	38.9	48.5	58.5	77.1	83.1	90.8	95.9		
Cyt) H3 (5 mg/L GA +5 mg/L Cyt)	13.1	14.1	39.0	40.9	49.4	61.8	78.6	85.8	91.8	97.5		
H SEm (±)	0.2	0.3	0.3	0.3	0.4	0.4	0.5	0.4	0.5	0.4		
H (CD at p ≤ 0.05)	NS	NS	1.0	0.8	1.2	1.0	1.5	1.2	1.4	1.0		
V x H SEm (±)	0.3	0.4	0.5	0.4	0.6	0.5	0.8	0.6	0.7	1.6		
V x H (CD at p ≤ 0.05)	NS											
Zn x H SEm (±)	0.4	0.5	0.6	0.5	0.7	0.6	0.9	0.7	0.9	0.6		
Zn x H (CD at p ≤ 0.05)	NS	1.8										
V x Zn x H SEm (±)	0.5	0.7	0.8	0.7	1.0	0.9	1.3	1.0	1.2	0.9		
V x Zn x H (CD at p≤ 0.05)	NS											

# Table 4.1: Effect of various treatments on periodic plant height (cm) of wheat during

Being a precursor of auxin, enhanced levels of tryptophan increase auxin biosynthesis which positively affect the meristem tissue involved in increasing the height. The results were supported by research outcomes of **Dhaliwal** *et al.* (2023), **Kaur** *et al.* (2023) and **Singh** *et al.* (2023). Increased cell division of the internodal area of the stem leads to increased height by external application of cytokinin and GA. Comparable results were stated by **Alharby** *et al.* (2021) and **Arif** *et al.* (2019). **Table 4.1a: Interaction effect of Zn and hormones on plant height (cm) of wheat for** 2022-23 at harvest.

	H <sub>0</sub> (No application)	H <sub>1</sub> (10 mg/L GA)	H <sub>2</sub> (10 mg/L Cyt)	H <sub>3</sub> (5 mg/L GA + 5 mg/L Cyt)	Mean				
Zn <sub>0</sub> (No application)	90.6	97.9	94.5	95.2	94.6				
Zn <sub>62.5</sub> (Soil)	95.8	97.8	95.0	97.4	96.5				
Zn <sub>31.25,F</sub> (Soil + Foliar)	97.3	101.8	98.2	99.9	99.3				
Mean	94.7	99.1	95.9	97.5					
Zn x H (CD at $p \le 0.05$ ) = 1.8									

# 4.2 Periodic number of tillers per metre square

The impact of different treatments on periodic tiller count of wheat is shown in table 4.2. Recorded results showed significant increase in tiller number due to different varieties in 2021-22 season. Maximum number of tillers were recorded in  $V_{725}$  (370.7) at 120 DAS which was significantly greater than  $V_{752}$  (313.3). The Zn application methods had a significant impact on the number of tillers at all growth phases. Mode Zn<sub>31.25,F</sub> (356.7) resulted in maximum number of tillers at 120 DAS which was statistically superior to Zn<sub>62.5</sub> (351.1) and Zn<sub>0</sub> (318.3). Among the phytohormones, all concentrations increased number of tillers significantly at all phases of growth except at 30 DAS. The level H<sub>2</sub> (358.3) had highest number of tillers at 120 DAS followed by H<sub>3</sub> (343.9), H<sub>1</sub> (334.8) and H<sub>0</sub> (330.9). Overall, the combination  $V_{725}$  + Zn<sub>31.25,F</sub> + H<sub>2</sub> was notable in improving the number of tillers and resulted in maximum (402.3) at 120 DAS.

In 2022-23 season, influence of varieties on number of tillers was observed to be statistically significant. The highest number of tillers were recorded for  $V_{725}$ (396.4) at 120 DAS which was significantly greater than  $V_{752}$  (377.4). The Zn application methods had a significant influence on number of tillers at all growth phases. Mode Zn<sub>31.25,F</sub> resulted in highest number of tillers (402.8) at 120 DAS which was statistically superior to Zn<sub>62.5</sub> (391.7) and Zn<sub>0</sub> (366.1).

Tillers per metre square											
	2021- 22	2022- 23	2021- 22	2022- 23	2021- 22	2022- 23	2021-	2022- 23			
Treatments		DAS		DAS		DAS	22 At 12	DAS			
Variety											
V725 (PBW 725)	205.5	238.1	465.3	504.4	404.3	434.4	370.7	396.4			
V752 (PBW 752)	174.8	226.9	421.9	465.4	351.1	403.3	313.3	377.4			
SEm (±)	1.2	3.7	4.3	4.4	3.6	3.5	1.8	3.1			
CD at p ≤ 0.05	7.5	NS	26.2	26.8	22.1	21.5	10.5	18.7			
Zinc application modes											
Zn <sub>0</sub> (No application)	180.6	216.3	416.8	463.3	351.2	396.6	318.3	366.1			
Zn62.5 (Soil)	199.2	246.0	450.7	493.4	385.1	424.2	351.1	391.7			
Zn31.25,F (Soil+Foliar)	190.5	235.1	463.3	498.0	396.8	436.1	356.7	402.8			
Zn SEm (±)	1.9	2.3	5.2	40	5.8	3.4	5.3	3.0			
Zn (CD at $p \le 0.05$ )	6.0	7.5	17.0	13.2	19.0	11.0	17.2	9.7			
V x Zn SEm (±)	2.6	3.3	7.4	5.7	8.2	4.8	7.5	4.2			
V x Zn (CD at p≤ 0.05)	NS										
,		Ho	ormone ap	plication			l				
H <sub>0</sub> (No application)	189.7	231.9	433.8	469.6	367.5	404.6	330.9	373.5			
H1 (10 mg/L GA)	189.2	232.2	436.5	474.7	370.8	411.0	334.8	379.4			
H <sub>2</sub> (10 mg/L Cyt)	190.3	233.6	460.0	500.1	394.3	432.3	358.3	399.3			
H <sub>3</sub> (5 mg/L GA +5 mg/L Cyt)	191.1	232.2	444.1	495.2	378.1	428.0	343.9	395.4			
H SEm (±)	2.0	3.4	4.1	2.7	4.1	2.1	3.3	1.9			
H (CD at $p \leq 0.05$ )	NS	NS	11.8	7.6	11.8	6.2	9.4	5.4			
V x H SEm (±)	2.8	4.8	5.8	3.8	5.8	3.0	4.6	2.7			
V x H (CD at p ≤ 0.05)	NS										
Zn x H SEm (±)	3.4	5.9	7.1	4.6	7.1	3.7	5.7	3.3			
Zn x H (CD at p ≤ 0.05)	NS										
V x Zn x H SEm (±)	4.8	8.3	10.1	6.5	10.1	5.3	8.0	4.6			
V x Zn x H (CD at p≤ 0.05)	NS										

Table 4.2: Effect of various treatments on number of tillers per metre square of wheatduring 2021-22 and 2022-23.

All the hormone concentrations increased count of tillers significantly at all phases of growth apart from at 30 DAS. Highest count of tillers were recorded at 120 DAS under H<sub>2</sub> (399.3) followed by H<sub>3</sub> (395.4), H<sub>1</sub> (379.4) and H<sub>0</sub> (373.5). The treatment combination  $V_{725}$  + Zn<sub>31.25,F</sub> + H<sub>2</sub> was notable in improving the number of tillers and maximum (423.2) were recorded at 120 DAS.

Tillering is a key growth stage in wheat that help plants to compensate for low plant populations or to benefit from favourable growth circumstances. The emergence of tillers is closely synchronised with the leaf formation on main stem and the number of tillers produced are determined by variety and growth circumstances (**Singh** *et al.*, **2021**). Application of Zn on the leaves may have enhanced the enzyme activity, chlorophyll, metabolite synthesis and nitrogen metabolism that allowed the plant to bear a greater number of tillers (**Arif** *et al.*, **2017**). Also, the exogeneous application of cytokinin enhanced chlorophyll synthesis which increased photosynthesis and sucrose production. Besides, increased the tiller bud growth resulted in higher number of tillers. The results inclined with the results of **Yang** *et al.* (**2021**).

# 4.3 Periodic leaf area index (LAI)

The influence of various treatments on periodic LAI is shown in table 4.3. During 2021-22 season, a comparison between wheat varieties showed significant results at 30, 60, 90 and 120 DAS. The highest LAI (4.14) was recorded at 90 DAS in V<sub>725</sub> followed by V<sub>752</sub> (3.87). Regarding the influence of Zn supply regimes, the effect was significant at 60, 90 and 120 DAS. Mode Zn<sub>31.25,F</sub> was statistically similar with Zn<sub>62.5</sub> at 60 DAS. Maximum LAI was recorded at 90 DAS under Zn<sub>31.25,F</sub> (4.19) which was followed by Zn<sub>62.5</sub> (4.0) and Zn<sub>0</sub> (3.83). Amongst the plant hormones, all levels significantly increased the LAI at 30, at 60, at 90 and also at 120 DAS. Highest LAI was recorded at 90 DAS under H<sub>2</sub> (4.15) followed by H<sub>3</sub> (4.04), H<sub>1</sub> (3.97) and H<sub>0</sub> (3.85). The levels H<sub>2</sub> and H<sub>3</sub> were statistically at par with each other at 90 DAS. In general, V<sub>725</sub> + Zn<sub>31.25,F</sub> + H<sub>2</sub> treatment combination was the most capable in improving the LAI and resulted in maximum (4.46) at 90 DAS.

During season 2022-23, a comparison of wheat varieties displayed significant impact on LAI at 30, at 60, at 90 and at 120 DAS. Maximum LAI was recorded at 90 DAS in V<sub>725</sub> (5.29) which was significantly greater than V<sub>752</sub> (4.71). Regarding the influence of Zn application methods, the effect was significant at 60, 90 and 120 DAS. Mode Zn<sub>31.25,F</sub> was statistically similar with mode Zn<sub>62.5</sub> at 60 DAS. Highest LAI was observed at 90 DAS under Zn<sub>31.25,F</sub> (5.23) which was significantly higher than Zn<sub>62.5</sub> (4.93) and Zn<sub>0</sub> (4.84). All levels of plant hormones significantly affected the LAI at 60, 90 and 120 DAS. The level H<sub>2</sub> was statistically similar with mode H<sub>3</sub> at 60 DAS. Highest LAI was recorded under H<sub>2</sub> (5.15) at 90 DAS followed by H<sub>3</sub> (4.99), H<sub>1</sub> (4.96) and H<sub>0</sub> (4.88). The levels H<sub>1</sub> and H<sub>3</sub> were statistically similar with each other at 90 DAS. In general,  $V_{725} + Zn_{31.25,F} + H_2$  treatment combination was the most effective in enhancing the LAI (5.73) at 90 DAS. The interaction effect among Zn and hormones on LAI at 90 DAS is presented in table 4.3a where highest LAI was recorded in Zn<sub>31.25,F</sub>+H<sub>2</sub> (5.44) and lowest LAI was recorded in Zn<sub>0</sub>+H<sub>0</sub> (4.74).

Leaf area index (LAI) is another important growth parameter of wheat that determines the potential of plants to trap solar radiation suitable for photosynthesis. The difference in leaf area was due to the variation in genetic makeup of the two varieties and the degree to which they are affected by the environment (Alzaayid and Aloush, 2021). Zinc application improves stomatal conductance, chlorophyll and other photosynthetic pigment content resulting in improved leaf area. Also, Zn is involved in regulation of auxin biosynthesis, which enhances root development and results in better nutrient and water intake, improving plant growth attributes (Ilyas *et al.*, 2020). Similarly, cytokinin application increased chlorophyll biosynthesis and stomatal conductance. In addition, cytokinin maintained leaf expansion and delayed leaf senescence contributed to enhanced leaf area. Nagar *et al.* (2015) and Zaheer *et al.* (2019) supported results of the present study.

#### 4.4 Periodic dry matter accumulation (g/m<sup>2</sup>)

Influence of various treatments on periodic dry matter accumulation is shown in table 4.4. The influence of wheat varieties on dry matter was found statistically significant at all growth stages in 2021-22 season. The highest dry matter was recorded in  $V_{725}$  (832.7 g/m<sup>2</sup>) which was significantly greater than  $V_{752}$  (725.2 g/m<sup>2</sup>) at 120 DAS. A significant result was recorded in context to the different Zn application methods at 60, 90 and 120 DAS. Modes Zn<sub>31.25,F</sub> and Zn<sub>62.5</sub> showed statistical parity at 60 DAS. The dry matter accumulation at 120 DAS was significantly higher under Zn<sub>31.25,F</sub> (819.2 g/m<sup>2</sup>) than Zn<sub>62.5</sub> (773.4 g/m<sup>2</sup>) and Zn<sub>0</sub> (744.3 g/m<sup>2</sup>). Comparing the different levels of phytohormones, all had a significant effect at 90 and 120 DAS. Level H<sub>1</sub> was statistically similar with H<sub>3</sub> at 90 DAS. Maximum dry matter resulted at harvest under H<sub>1</sub> (800.6 g/m<sup>2</sup>) followed by H<sub>3</sub> (790.9 g/m<sup>2</sup>), H<sub>2</sub> (789.0 g/m<sup>2</sup>) and H<sub>0</sub> (735.3 g/m<sup>2</sup>). Level H<sub>1</sub> was statistically at par with H<sub>2</sub> at harvest. Overall, the treatment combination  $V_{725}$  +  $Zn_{31.25,F}$  +  $H_3$  was recorded the most effective with maximum dry matter (903.3 g/m²) at harvest.

Table 4.3: Effect of various treatments on leaf area index (LAI) of wheat during 2021-22
and 2022-23.

	Leaf area index (LAI)											
	2021-22	2022-23	2021-22	2022-23	2021-22	2022-23	2021-22	2022-23				
Treatments	At 30	) DAS	At 60	DAS	At 90 DAS		At 12	0 DAS				
	Variety											
V725 (PBW 725)	0.51	0.56	2.14	3.15	4.14	5.29	3.21	4.15				
V752 (PBW 752)	0.39	0.43	1.96	2.74	3.87	4.71	3.00	3.90				
SEm (±)	0.01	0.01	0.01	0.03	0.04	0.01	0.02	0.04				
CD at $p \leq 0.05$	0.05	0.09	0.04	0.18	0.26	0.04	0.14	0.22				
Zinc application modes												
Zno (No application)	0.43	0.45	1.98	2.82	3.83	4.84	2.93	3.96				
Zn <sub>62.5</sub> (Soil)	0.46	0.52	2.06	2.98	4.00	4.93	3.15	4.00				
Zn <sub>31.25,F</sub> (Soil+Foliar)	0.44	0.50	2.12	3.03	4.19	5.23	3.25	4.13				
Zn SEm (±)	0.02	0.01	0.03	0.04	0.05	0.01	0.03	0.02				
Zn (CD at p ≤ 0.05)	NS	0.03	0.08	0.13	0.16	0.03	0.11	0.06				
V x Zn SEm (±)	0.03	0.04	0.04	0.05	0.07	0.02	0.05	0.06				
V x Zn (CD at p≤ 0.05)	NS	NS	NS	NS	NS	NS	NS	NS				
			Hormon	e applicatio	on							
H₀ (No application)	0.44	0.49	1.98	2.82	3.85	4.88	2.98	3.97				
H <sub>1</sub> (10 mg/L GA)	0.44	0.49	2.03	2.92	3.97	4.96	3.09	4.01				
H <sub>2</sub> (10 mg/L Cyt)	0.45	0.50	2.13	3.05	4.15	5.15	3.21	4.10				
H <sub>3</sub> (5 mg/L GA +5 mg/L Cyt)	0.45	0.50	2.08	2.98	4.04	4.99	3.15	4.02				
H SEm (±)	0.01	0.01	0.03	0.03	0.04	0.01	0.03	0.02				
H (CD at p ≤ 0.05)	NS	NS	0.07	0.09	0.12	0.04	0.07	0.06				
V x H SEm (±)	0.01	0.01	0.04	0.04	0.06	0.02	0.04	0.03				
V x H (CD at p ≤ 0.05)	NS	NS	NS	NS	NS	NS	NS	NS				
Zn x H SEm (±)	0.02	0.02	0.04	0.05	0.07	0.03	0.04	0.04				
$Zn \ge H (CD at p \le 0.05)$	NS	NS	NS	NS	NS	0.07	NS	NS				
$\frac{\mathbf{V} \mathbf{x} \mathbf{Z} \mathbf{n} \mathbf{x} \mathbf{H}}{\mathbf{SEm} (\pm)}$	0.03	0.02	0.06	0.08	0.10	0.04	0.06	0.06				
V x Zn x H (CD at p≤ 0.05)	NS	NS	NS	NS	NS	NS	NS	NS				

	H <sub>0</sub> (No application)	H1 (10 mg/L GA)	H <sub>2</sub> (10 mg/L Cyt)	H <sub>3</sub> (5 mg/L GA + 5 mg/L Cyt)	Mean				
Zn <sub>0</sub> (No application)	4.74	4.84	4.94	4.83	4.84				
Zn <sub>62.5</sub> (Soil)	4.82	4.89	5.07	4.93	4.93				
Zn <sub>31.25.F</sub> (Soil + Foliar)	5.08	5.17	5.44	5.22	5.23				
Mean	4.88	4.96	5.15	4.99					
$Zn \ x \ H \ (CD \ at \ p \le 0.05) = 0.07$									

Table 4.3a: Interaction effect of Zn and hormones on leaf area index (LAI) of wheat for 2022-23 at 90 DAS.

For season 2022-23, comparison of two wheat varieties revealed that there was significant varietal difference on dry matter accumulation at all phases of growth. The highest dry matter was recorded at 120 DAS in V<sub>725</sub> (1067.3 g/m<sup>2</sup>) was statistically superior to V<sub>752</sub> (916.6 g/m<sup>2</sup>). Concerning the methods of Zn application, all affected dry matter significantly at 60, at 90 and at 120 DAS. The mean dry matter under Zn<sub>31.25,F</sub> (1043.6 g/m<sup>2</sup>) at 120 DAS was significantly higher than Zn<sub>62.5</sub> (989.3 g/m<sup>2</sup>) and Zn<sub>0</sub> (942.9 g/m<sup>2</sup>). The different levels of phytohormones affected the dry matter significantly at all phases of growth apart from at 30 DAS. Level H<sub>1</sub> was statistically similar with H<sub>3</sub> at 60 and at 90 DAS. Levels H<sub>2</sub> and H<sub>3</sub> showed statistical parity at 120 DAS. Highest accumulation of dry matter was recorded at 120 DAS under H<sub>1</sub> (1026.8 g/m<sup>2</sup>) which was followed by H<sub>3</sub> (1003.9 g/m<sup>2</sup>), H<sub>2</sub> (996.6 g/m<sup>2</sup>) and H<sub>0</sub> (940.6 g/m<sup>2</sup>). Overall, V<sub>725</sub> + Zn<sub>31.25,F</sub> + H<sub>1</sub> treatment resulted in maximum dry matter (1162.0 g/m<sup>2</sup>) at 120 DAS.

Dry matter accumulation is a vital parameter of growth that expresses the plant's metabolic efficiency. Dry matter continuously increased over the crop life cycle. Higher leaf area, tiller per m<sup>2</sup> and plant height collectively may be the reason of the improved dry matter, as increased leaf area resulted in improved interception and utilisation of solar energy, resulted in more photosynthesis and eventually more accumulation dry matter. The timely sown wheat variety had more time to accumulate dry matter than the late sown wheat variety. It may have caused the higher dry matter and yield in timely sown wheat variety (**Arduini** *et al.*, **2006**). The soil+foliar method increased the availability of Zn in plant system which led to improved metabolism and functioning of the plant by enhancing growth of roots, shoots and leaves, thus resulted in more accumulation of dry matter (**Suganya** *et al.*, **2020**). The hormone application upregulates the metabolic processes in the plant and the positive effect

was observed in height of plant, number of tillers and leaf area culminating to more accumulation of dry matter (Mathpal *et al.*, 2023; Zaheer *et al.*, 2022).

		Dry n	natter accu	mulation	(g/m <sup>2</sup> )			
	2021- 22	2022-23	2021- 22	2022- 23	2021- 22	2022- 23	2021- 22	2022-23
Treatments	At 3	) DAS	At 60 DAS		At 90 DAS		At 120 DAS	
			Var	iety				
V725 (PBW 725)	43.1	50.8	259.2	340.4	523.6	684.0	832.7	1067.3
V752 (PBW 752)	36.9	49.8	185.5	285.0	405.6	534.6	725.2	916.8
SEm (±)	0.5	0.6	2.7	3.8	5.0	15.7	4.8	6.8
CD at p ≤ 0.05	2.8	NS	16.3	23.0	30.4	95.3	29.5	41.4
		Z	inc applic	ation mode	es			
Zno (No application)	38.2	44.7	203.4	292.2	441.8	579.7	744.3	942.9
Zn62.5 (Soil)	39.9	50.8	226.3	316.2	463.3	607.6	773.4	989.3
Zn <sub>31.25,F</sub> (Soil+Foliar)	42.0	55.5	237.3	329.7	488.6	640.6	819.2	1043.6
Zn SEm (±)	1.2	1.3	5.8	3.4	6.4	8.4	11.0	10.0
Zn (CD at p ≤ 0.05)	NS	4.1	19.1	11.0	21.0	27.3	36.0	32.7
V x Zn SEm (±)	1.8	1.8	8.3	4.8	9.1	11.9	15.6	14.2
V x Zn (CD at p≤ 0.05)	NS	NS	NS	NS	NS	NS	NS	NS
		]	Hormone :	application	l			
H₀ (No application)	39.9	50.2	206.6	296.9	441.0	578.6	735.3	940.6
H1 (10 mg/L GA)	40.2	50.3	234.6	323.2	480.8	630.4	800.6	1026.8
H <sub>2</sub> (10 mg/L Cyt)	40.1	50.5	222.2	311.5	466.6	611.9	789.0	996.6
H <sub>3</sub> (5 mg/L GA +5 mg/L Cyt)	39.9	50.3	226.0	319.4	470.0	616.3	791.0	1003.9
H SEm (±)	1.4	1.3	7.9	2.4	8.1	10.5	11.0	10.4
H (CD at p ≤ 0.05)	NS	NS	NS	6.9	23.2	30.1	31.6	29.8
V x H SEm (±)	1.9	1.9	11.2	3.4	11.4	14.8	15.6	14.7
V x H (CD at p ≤ 0.05)	NS	NS	NS	NS	NS	NS	NS	NS
Zn x H SEm (±)	2.3	2.3	13.7	4.2	14.0	18.2	19.1	18.0
Zn x H (CD at p ≤ 0.05)	NS	NS	NS	NS	NS	NS	NS	NS
V x Zn x H SEm (±)	3.3	3.3	19.4	5.9	20.0	25.7	27.0	25.5
V x Zn x H (CD at p≤ 0.05)	NS	NS	NS	NS	NS	NS	NS	NS

# Table 4.4: Effect of various treatments on dry matter accumulation $(g/m^2)$ of wheat during 2021-22 and 2022-23.

#### 4.5 Number of effective tillers per metre square at harvest

The influence of various treatments on number of effective tillers at harvest is shown in table 4.5. The recorded results for both the wheat varieties showed significant difference regarding the count of effective tillers in the season 2021-22. The highest effective tillers count was recorded in  $V_{725}$  (321.9) that was significantly greater than to  $V_{752}$  (270.3). Among the Zn application methods, all significantly influenced the count of effective tillers. The highest count of effective tillers resulted under  $Zn_{31.25,F}$  (309.3) followed by  $Zn_{62.5}$  (306.6) and  $Zn_0$  (272.5). The methods  $Zn_{62.5}$  and  $Zn_{31.25,F}$  showed statistical parity to each other. A significant effect of plant hormones was recorded on count of effective tillers. The count of effective tiller were maximum under  $H_2$  (312.2) followed by  $H_3$  (298.4),  $H_1$  (288.7) and  $H_0$  (285.1). The levels  $H_3$  &  $H_1$  and  $H_0$  &  $H_1$  were statistically similar to each other. The treatment combination  $V_{725} + Zn_{31.25,F} + H_2$  was most influential and had the highest count of effective tillers (351.6).

For 2022-23 season, a significant variation in number of effective tillers was recorded between varieties. Maximum effective tillers count was recorded in V<sub>725</sub> (340.3) which was significantly higher than V<sub>752</sub> (328.3). Pertaining to the methods of Zn application, the effective tillers count was significantly affected by the different methods. The method Zn<sub>31.25,F</sub> (352.2) had maximum effective tillers count which was significantly greater than to Zn<sub>62.5</sub> (336.7) and Zn<sub>0</sub> (314.0). All the hormone concentrations significantly influenced the number of effective tillers. Highest number of effective tillers were recorded under H<sub>2</sub> (354.5) followed by H<sub>3</sub> (341.1), H<sub>1</sub> (326.5) and H<sub>0</sub> (315.0). The interaction effect of Zn and hormones on count of effective tillers is presented in table 4.5a. Maximum effective tiller count was recorded under Zn<sub>31.25,F</sub>+H<sub>2</sub> (375.5) and minimum was recorded under Zn<sub>0</sub>+H<sub>0</sub> (299.4). The interaction effect of variety, Zn and hormones on effective tillers during 2022-23 season is presented in table 4.5b. The treatment combination V<sub>725</sub>+ Zn<sub>31.25,F</sub> + H<sub>2</sub> resulted in maximum number of effective tillers (378.3).

The effective tillers indicate the crop's performance. Therefore, higher number of effective tillers signifies higher crop performance (**Kaur** *et al.*, **2017**). The differential capacity of different varieties to accumulate and transport assimilates from source to sink showed an effect on count of effective tillers (**Madhu** *et al.*, **2018**). Application of Zn enhances the enzymatic and metabolic activity, thus enhanced the count of effective tillers (**Arif** *et al.*, **2017**). The exogeneous cytokinin application

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promotes the tiller bud growth and improves assimilate translocation so increased count of effective tillers. The results inclined with the results of **Yang** *et al.* (2021).

Table 4.5: Effect of various treatments on yield contributing attributes of wheat during
2021-22 and 2022-23.

20		Y	ïeld contril	outing attri	ibutes		10 C	
	2021- 22	2022-23	2021-22	2022-23	2021-22	2022-23	2021-22	2022-23
Treatments	Eff	ective rs/m²	Spike ler	igth (cm)	Number of grains/spike		1000-grain weigh (g)	
			v	ariety				8/
V725 (PBW 725)	321.9	340.3	10.3	11.5	45.3	52.6	42.6	43.7
V752 (PBW 752)	270.3	328.3	9.3	10.5	39.2	47.1	41.2	42.0
SEm (±)	1.1	1.1	0.1	0.1	0.9	0.3	0.2	0.3
CD at $p \le 0.05$	6.6	6.7	0.7	0.8	5.6	1.8	1.1	1.5
			Zinc appl	ication mo	des			
Zn <sub>0</sub> (No application)	272.5	314.0	9.5	10.6	40.0	47.0	40.8	41.2
Zn62.5 (Soil)	306.6	336.7	9.8	11.0	41.8	49.4	41.7	42.6
Zn31.25,F (Soil+Foliar)	309.3	352.2	10.1	11.3	45.0	53.1	43.1	44.7
Zn SEm (±)	4.1	0.5	0.1	0.1	1.1	0.7	0.2	0.2
Zn (CD at p ≤ 0.05)	13.5	1.7	0.4	0.4	3.6	2.4	0.8	0.6
V x Zn SEm (±)	5.9	0.7	0.2	0.2	1.6	1.1	0.3	0.3
V x Zn (CD at p≤ 0.05)	NS	NS	NS	NS	NS	NS	NS	NS
			Hormon	e applicatio	)n			
H₀ (No application)	285.1	315.0	9.5	10.6	39.0	47.2	40.6	41.2
H1 (10 mg/L GA)	288.7	326.5	10.0	11.2	42.9	49.3	41.6	41.8
H <sub>2</sub> (10 mg/L Cyt)	312.2	354.5	9.8	11.0	44.3	52.1	43.1	44.7
H <sub>3</sub> (5 mg/L GA +5 mg/L Cyt)	298.4	341.1	9.9	11.1	42.9	50.6	42.9	43.6
H SEm (±)	3.5	1.1	0.1	0.1	1.0	0.9	0.3	0.1
H (CD at p ≤ 0.05)	10.1	3.1	0.4	0.4	2.8	2.5	0.9	0.3
V x H SEm (±)	5.0	1.6	0.2	0.2	1.4	1.2	0.5	0.2
V x H (CD at p ≤ 0.05)	NS	NS	NS	NS	NS	NS	NS	NS
Zn x H SEm (±)	6.1	1.9	0.2	0.2	1.7	1.5	0.6	0.2
Zn x H (CD at p ≤ 0.05)	NS	5.4	NS	NS	NS	NS	NS	0.6
V x Zn x H SEm (±)	8.7	2.7	0.3	0.3	2.4	2.1	0.8	0.3
V x Zn x H (CD at p≤ 0.05)	NS	7.7	NS	NS	NS	NS	NS	NS

	H <sub>0</sub> (No application)	H <sub>1</sub> (10 mg/L GA)	H <sub>2</sub> (10 mg/L Cyt)	H <sub>3</sub> (5 mg/L GA + 5 mg/L Cyt)	Mean		
Zn <sub>0</sub> (No application)	299.4	308.7	330.2	317.7	314.0		
Zn <sub>62.5</sub> (Soil)	316.8	328.2	357.9	343.8	336.7		
Zn31.25,F (Soil + Foliar)	328.7	342.7	375.5	361.7	352.2		
Mean	315.0	326.5	354.5	341.1			
Zn x H (CD at p≤ 0.05) = 5.4							

Table 4.5a: Interaction effect of Zn and hormones on number of effective tillers permetre square of wheat for 2022-23.

Table 4.5b: Interaction effect of variety, Zn and hormones on number of effective tillers per metre square of wheat for 2022-23.

V725 (PBW 725)					V <sub>752</sub> (PBW 752)			
	Zn <sub>0</sub> (No application)	Zn <sub>62.5</sub> (Soil)	Zn <sub>31.25,F</sub> (Soil + Foliar)	Mean	Zn <sub>0</sub> (No application)	Zn <sub>62.5</sub> (Soil)	Zn <sub>31.25,F</sub> (Soil + Foliar)	Mean
H <sub>0</sub> (No application)	302.5	323.7	337.6	321.3	296.3	310.0	320.1	308.8
H1 (10 mg/L GA)	314.0	334.0	352.0	333.3	303.3	322.3	333.3	319.7
H <sub>2</sub> (10 mg/L cyt)	339.4	365.4	378.3	361.0	321.0	350.4	372.7	348.0
H <sub>3</sub> (5 mg/L GA+ 5 mg/L cyt)	324.0	347.5	365.0	345.5	311.3	340.2	358.4	336.6
Mean	320.0	342.6	358.2		308.0	330.7	346.1	
V x Zn x H (CD at p≤0.05) = 7.7								

Table 4.5c: Interaction effect of Zn and hormones on thousand grain weight (g) of wheat
for 2022-23.

	H <sub>0</sub> (No application)	H1 (10 mg/L GA)	H <sub>2</sub> (10 mg/L Cyt)	H <sub>3</sub> (5 mg/L GA + 5 mg/L Cyt)	Mean			
Zn <sub>0</sub> (No application)	40.0	40.4	42.7	41.6	41.2			
Zn <sub>62.5</sub> (Soil)	40.8	41.7	44.6	43.5	42.6			
Zn <sub>31.25,F</sub> (Soil + Foliar)	42.8	43.4	46.8	45.9	44.7			
Mean	41.2	41.8	44.7	43.7				
	Zn x H (CD at p≤ 0.05) = 0.6							

#### 4.6 Spike length (cm)

Data regarding the effect of various treatments on spike length is displayed in table 4.5. The varieties showed significant difference in the spike length during season 2021-22. The maximum length of spike was recorded in  $V_{725}$  (10.3 cm) that was significantly higher than  $V_{752}$  (9.3 cm). The different Zn application methods significantly impacted the spike length. Mode Zn<sub>31.25,F</sub> had the longest spike (10.1 cm) that was followed by Zn<sub>62.5</sub> (9.8 cm) and Zn<sub>0</sub> (9.5 cm). The methods Zn<sub>62.5</sub> & Zn<sub>62.5</sub> and Zn<sub>62.5</sub> & Zn<sub>0</sub> were statistically similar to each other. A significant impact was recorded on spike length by the different plant hormone levels. The longest spike resulted under H<sub>1</sub> (10.0 cm) that was followed by H<sub>3</sub> (9.9 cm), H<sub>1</sub> (9.8 cm) and H<sub>0</sub> (9.5 cm). The levels H<sub>1</sub>, H<sub>2</sub> and H<sub>3</sub> were statistically similar. The treatment  $V_{725}$  + Zn<sub>31.25,F</sub> + H<sub>1</sub> was most effective in increasing the spike length and resulted in longest spike (11.0 cm).

During season 2022-23, the spike length of varieties varied significantly. The maximum length of spike was recorded in V<sub>725</sub> (11.5 cm) that was significantly longer than V<sub>752</sub> (10.5 cm). The various Zn application methods significantly influenced the spike length. The longest spike was recorded under Zn<sub>31.25,F</sub> (11.3 cm) that was significantly higher than Zn<sub>62.5</sub> (11.0 cm) and Zn<sub>0</sub> (10.6 cm). Regarding the various levels of plant hormone, a significant effect was recorded on spike length. The longest spike was recorded under H<sub>1</sub> (11.2 cm) followed by H<sub>3</sub> (11.1 cm), H<sub>2</sub> (11.0 cm) and H<sub>0</sub> (10.6 cm). The level H<sub>1</sub> was statistically similar to H<sub>3</sub> and H<sub>2</sub>. Overall, the combination V<sub>725</sub> + Zn<sub>31.25,F</sub> + H<sub>1</sub> was most effective in increasing the spike length and resulted the longest spike (12.3 cm).

Spike length is a key component of wheat yield. An increase in the spike length promotes the number of spikelets/spike, increasing grains/spike and grain yield. The difference in spike length of the varieties was due to the inherent capacity of different genotypes (Madhu *et al.*, 2018; Ullah *et al.*, 2021). Zinc participates in various metabolic processes which leads to enhancement of green pigments, biosynthesis of flavonoids and photosynthetic efficiency leading to increased spike length (Sattar *et al.*, 2022). Cytokinin accumulates in the meristem of inflorescence thus, regulates the development of spike by increasing flowering. Raza *et al.* (2020) supported the results and reported increase in spike length by the application of cytokinin at different stages of crop.

#### 4.7 Number of grains/spike

Influence of various treatments on number of grains/spike is displayed in table 4.5. During 2021-22 season, the varieties differ significantly in the count of grains/spike. Higher grains/spike were recorded in  $V_{725}$  (45.3) followed by  $V_{752}$  (39.2). A significant impact was recorded pertaining to the different Zn supply regimes. Mode Zn<sub>31.25,F</sub> had the maximum number of grains (45.0) followed by Zn<sub>62.5</sub> (41.8) and Zn<sub>0</sub> (40.0). Modes Zn<sub>31.25,F</sub> & Zn<sub>62.5</sub> and Zn<sub>62.5</sub> & Zn<sub>0</sub> were statistically at par with each other. Pertaining to the impact of various phytohormone levels, the number of grains/spike differed significantly. Maximum grain count was recorded under H<sub>2</sub> (44.3) followed by H<sub>3</sub> (42.9), H<sub>1</sub> (42.9) and H<sub>0</sub> (39.0). The levels H<sub>2</sub>, H<sub>3</sub> and H<sub>1</sub> were statistically similar. Overall, the treatment combination of  $V_{725}$  + Zn<sub>31.25,F</sub> + H<sub>2</sub> was most efficient and had the maximum number of grains (50.0) per spike.

During 2022-23 season, the varieties showed significant difference for the grain count per spike. The highest number was recorded in V<sub>725</sub> (52.6) followed by V<sub>752</sub> (47.1). A significant effect was recorded with respect to various methods of Zn application. The maximum number of grains per spike resulted under Zn<sub>31.25,F</sub> (53.1) which was followed by Zn<sub>62.5</sub> (49.4) and Zn<sub>0</sub> (47.0). There was a significant influence of various phytohormone levels on number of grains. Level H<sub>2</sub> (52.2) had maximum number of grains followed by H<sub>3</sub> (50.6), H<sub>1</sub> (49.3) and H<sub>0</sub> (47.2). Overall, the treatment combination of V<sub>725</sub> + Zn<sub>31.25,F</sub> + H<sub>2</sub> had highest number of grains/spike (58.3).

The count of grains/spike is a pivotal yield attribute and increase in the grains/spike directly affects the grain yield. The variation among count of grains/spike for two varieties may be ascribed to the genetic make-up and flag leaf area of both the varieties. The results were supported by **Alzaayid and Aloush (2021)**. Zinc (Zn) is an important part of different enzymes and metabolic processes. It helps in biosynthesis of chlorophyll, development the pollen tubes, pollen viability, utilization of starch which enhanced the number of grains per spike (**Arif** *et al.*, **2017**). Cell division in endosperm and boosted sink capacity by cytokinin resulted in increased assimilate accumulation culminating to higher number of grains/spike. The findings of the study were supported by **Raza** *et al.* (**2020**).

#### **4.8 Thousand grain weight (g)**

Effect of different treatment on thousand grain weight is shown in table 4.5. The difference between varieties was found statistically significant during 2021-22 season. Variety  $V_{725}$  (42.6 g) had significantly higher grain weight than variety  $V_{752}$  (41.2 g). A significant impact was recorded pertaining to the Zn application methods on grain weight. The method Zn<sub>31.25,F</sub> (43.1 g) resulted in significantly heavier grains than Zn<sub>62.5</sub> (41.7 g) and Zn<sub>0</sub> (40.8 g). With regard to different levels of hormones, a significant effect was observed. The maximum grain weight was recorded under H<sub>2</sub> (43.1 g) followed by H<sub>3</sub> (42.3 g), H<sub>1</sub> (41.6 g) and H<sub>0</sub> (40.6 g). The levels H<sub>2</sub> & H<sub>3</sub> and H<sub>1</sub> & H<sub>3</sub> were statistically at par with each other. Overall,  $V_{725} + Zn_{31.25,F} + H_2$  treatment was most efficient and resulted in the highest grain weight (45.3 g).

During 2022-23 season, the varietal difference was found statistically significant regarding the grain weight. Heavier grains were recorded in V<sub>725</sub> (43.7 g) than V<sub>752</sub> (42.0 g). Pertaining to different methods of Zn application, a significant effect was recorded. Mode Zn<sub>31.25,F</sub> (44.7) resulted in significantly heavier grains followed by Zn<sub>62.5</sub> (42.6 g) and Zn<sub>0</sub> (41.2 g). There was a significant influence of various levels of phytohormone on the grain weight. The heaviest grains were recorded under H<sub>2</sub> (44.7 g) which was significantly greater than H<sub>3</sub> (43.7 g), H<sub>1</sub> (41.8 g) and H<sub>0</sub> (41.2 g). Overall, the V<sub>725</sub> + Zn<sub>31.25,F</sub> + H<sub>2</sub> treatment combination resulted in heaviest grains (47.6 g). The interaction effect of Zn and hormones on grain weight during the 2022-23 season is presented in table 4.5c. The heaviest grains were recorded under Zn<sub>31.25,F</sub>+H<sub>2</sub> (46.8 g) while the lightest grains were recorded under Zn<sub>0</sub>+H<sub>0</sub> (40.0 g).

Grain weight along with the number of grains are two important yield contributing attributes of crops. Heavier weight of grains is linked with better quality of grains. Hence, it is vital to increase grain weight not only to increase yield but also to produce high quality wheat. Varietal difference for grain weight is due to differential capacity of the various genotypes to translocate assimilate from source to sink (**Madhu** *et al.*, **2018** and **Ullah** *et al.*, **2021**). Zinc fertilization through soil and foliar spray led to improved Zn content in plant which increase dry matter production and its translocation to grains, enhancing the grain size and weight. Findings of this study were corroborated by **Ghasal** *et al.* (**2017b**). Cytokinin increased assimilate translocation and enhanced the division of endospermic cells which further increased the sink strength and resulted in heavier grains in wheat (**Raza** *et al.*, **2020**).

#### 4.9 Grain yield (q/ha)

Influence of various treatments on grain yield is displayed in table 4.6. During 2021-22 season, a significant difference was recorded between the wheat varieties. Higher grain yield was recorded in  $V_{725}$  (45.5 q/ha) which was significantly greater than  $V_{752}$  (41.4 q/ha). Grain yield was significantly influenced by the Zn application methods. The maximum yield was recorded under Zn<sub>31.25,F</sub> (45.6 q/ha) that was followed by Zn<sub>62.5</sub> (43.5 q/ha) and Zn<sub>0</sub> (41.3 q/ha). The methods Zn<sub>31.25,F</sub> was statistically at par with Zn<sub>62.5</sub>. The different levels phytohormone had a significant impact on grain yield. Highest wheat grain yield was recorded under H<sub>2</sub> (45.1 q/ha) that was followed by H<sub>3</sub> (44.1 q/ha), H<sub>1</sub> (43.2 q/ha) and H<sub>0</sub> (41.4 q/ha). The levels H<sub>2</sub> & H<sub>3</sub> and H<sub>1</sub> & H<sub>3</sub> were statistically at par with each other. In general, treatment combination  $V_{725}$  + Zn<sub>31.25,F</sub> + H<sub>2</sub> was the most effective in increasing grain yield (49.3 q/ha).

During 2022-23 season, impact of varieties on grain yield was found statistically significant. Maximum grain yield was recorded for V<sub>725</sub> (52.9 q/ha) which was significantly greater than V<sub>752</sub> (46.3 q/ha). Regarding the impact of Zn on wheat, all methods had a significant influence on grain yield. Highest grain yield was recorded under Zn<sub>31.25,F</sub> (51.3 q/ha) which was significantly greater than Zn<sub>62.5</sub> (49.7 q/ha) and Zn<sub>0</sub> (47.7 q/ha). For different levels of phytohormone, all had a significant impact on grain yield. Maximum grain yield was recorded under H<sub>2</sub> (51.6 q/ha) that was followed by H<sub>3</sub> (50.2 q/ha), H<sub>1</sub> (48.8 q/ha) and H<sub>0</sub> (47.7 q/ha). The interaction effect of variety and Zn on grain yield is presented in table 4.6a. Maximum grain yield was recorded for V<sub>725</sub>+Zn<sub>31.25,F</sub> (54.9 q/ha). The interaction effect of Zn and hormones on grain yield is presented in table 4.6b with Zn<sub>31.25,F</sub>+H<sub>2</sub> having maximum grain yield (54.0 q/ha). The interaction effect of variety, Zn and hormones on grain yield is presented in table 4.6c. The treatment combination V<sub>725</sub> + Zn<sub>31.25,F</sub> + H<sub>2</sub> was the most effective and resulted in maximum grain yield (57.6 q/ha).

Grain yield is the most commercially valued component of the crops. The difference in grain yield in two varieties was due to different dates of sowing wherein, the late sown variety had less duration of vegetative and reproductive phases (**Panigrahi** *et al.*, 2022). Enhanced wheat grain yield maybe due to more carbohydrate synthesis in plants. The metabolic processes related to carbohydrate synthesis influenced by Zn which resulted in increased photosynthesis, sugar conversions and seed development. So, an increase in Zn content and its efficient

assimilation led to bolder grains, which subsequently led to an increased grain yield. The results inclined with findings of **Ghasal** *et al.* (2017a) and **Kumar** *et al.* (2019). The cytokinin application increased the number of productive tillers and helped in transportation of assimilates to grains, increasing grain size, weight and yield. **Koprna** *et al.* (2020) and **Zaheer** *et al.* (2022) also supported the results of our study. During the 2021-22 season, heat stress was caused during the grain-filling and ripening stage due to increase in both maximum and minimum temperature by more than 3 degree Celsius. This resulted in the decreased grain yield. These results aligned with the findings of **Singh** *et al.*, (2022).

Table 4.6: Effect of various treatments on yield and harvest index of wheat during 2021-
22 and 2022-23.

	3	lield and ha	vest index					
	2021-22	2022-23	2021-22	2022-23	2021-22	2022-23		
Treatments	Grain yield (q/ha)		Straw yield (q/ha)		Harvest index (%)			
Variety								
V725 (PBW 725)	45.5	52.9	63.0	79.2	42.0	40.0		
V752 (PBW 752)	41.4	46.3	55.7	69.3	42.7	40.0		
SEm (±)	0.5	0.1	0.5	0.1	0.2	0.01		
CD at p ≤ 0.05	3.1	0.4	2.8	0.6	NS	NS		
	2	Zinc applicat	ion modes					
Zn <sub>0</sub> (No application)	41.3	47.7	56.1	71.6	42.4	40.0		
Zn62.5 (Soil)	43.5	49.7	59.4	74.4	42.3	40.1		
Zn31.25,F (Soil+Foliar)	45.6	51.3	62.5	76.7	42.2	40.1		
Zn SEm (±)	0.6	0.1	0.9	0.1	0.4	0.1		
Zn (CD at $p \le 0.05$ )	2.0	0.2	2.9	0.3	NS	NS		
V x Zn SEm (±)	0.9	0.1	1.3	0.1	0.6	0.1		
V x Zn (CD at p≤0.05)	NS	0.3	NS	0.4	NS	NS		
		Hormone ap	plication					
H <sub>0</sub> (No application)	41.4	47.7	56.3	71.5	42.4	40.1		
H1 (10 mg/L GA)	43.2	48.8	61.6	77.4	41.3	38.7		
H <sub>2</sub> (10 mg/L Cyt)	45.1	51.6	59.3	73.2	43.2	41.3		
H3 (5 mg/L GA +5 mg/L Cyt)	44.1	50.2	60.1	75.1	42.3	40.1		
H SEm (±)	0.6	0.1	0.8	0.2	0.4	004		
H (CD at $p \leq 0.05$ )	1.7	0.2	2.2	0.4	1.2	0.1		
V x H SEm (±)	0.9	0.1	1.1	0.2	0.6	0.1		
V x H (CD at p $\leq$ 0.05)	NS	NS	NS	NS	NS	0.2		
Zn x H SEm (±)	1.1	0.1	1.3	0.3	0.7	0.1		
Zn x H (CD at $p \le 0.05$ )	NS	0.4	NS	0.7	NS	0.2		
V x Zn x H SEm (±)	1.5	0.2	1.8	0.4	1.0	0.1		
V x Zn x H (CD at p≤ 0.05)	NS	0.5	NS	NS	NS	NS		

	Zn <sub>0</sub> (No application)	Zn <sub>62.5</sub> (Soil)	Zn <sub>31.25,F</sub> (Soil + Foliar)	Mean		
V725 (PBW 725)	50.5	53.3	54.9	52.9		
V752 (PBW 752)	44.9	46.1	47.7	46.3		
Mean	47.7	49.7	51.3			
V x Zn (CD at p ≤ 0.05) = 0.3						

Table 4.6a: Interaction effect of variety and Zn on grain yield (q/ha) of wheat for 2022-23.

Table 4.6b: Interaction effect of Zn and hormones on grain yield (q/ha) of wheat for 2022-23.

	H <sub>0</sub> (No application)	H1 (10 mg/L GA)	H <sub>2</sub> (10 mg/L Cyt)	H <sub>3</sub> (5 mg/L GA + 5 mg/L Cyt)	Mean		
Zn <sub>0</sub> (No application)	46.3	47.2	49.2	48.2	47.7		
Zn <sub>62.5</sub> (Soil)	47.9	49.0	51.6	50.4	49.7		
Zn <sub>31.25,F</sub> (Soil + Foliar)	49.1	50.2	54.0	52.0	51.3		
Mean	47.7	48.8	51.6	51.2			
Zn x H (CD at p ≤ 0.05) = 0.4							

Table 4.6c: Interaction effect of variety, Zn and hormones on grain yield (q/ha) of wheat	t
for 2022-23.	

V725 (PBW 725)					V752 (PBW 752)			
	Zn <sub>0</sub> (No application)	Zn <sub>62.5</sub> (Soil)	Zn <sub>31.25,F</sub> (Soil + Foliar)	Mean	Zn <sub>0</sub> (No application)	Zn <sub>62.5</sub> (Soil)	Zn <sub>31.25,F</sub> (Soil + Foliar)	Mean
H <sub>0</sub> (No application)	48.9	51.6	53.0	51.2	43.6	44.1	45.3	44.3
H1 (10 mg/L GA)	50.0	52.7	53.8	52.1	44.5	45.3	46.5	45.4
H <sub>2</sub> (10 mg/L cyt)	52.1	55.0	57.6	54.9	46.2	48.1	50.5	48.3
H <sub>3</sub> (5 mg/L GA+ 5 mg/L cyt)	51.1	53.8	55.2	53.4	45.3	47.0	48.7	47.0
Mean	50.5	53.3	54.9		44.9	46.1	47.7	
V x Zn x H (CD at p≤ 0.05) = 0.5								

#### 4.10 Straw yield (q/ha)

Influence of different treatments on the straw yield of the two wheat varieties is shown in table 4.6. During 2021-22 season, difference between two wheat varieties was found significant. The straw yield of  $V_{725}$  (63.0 q/ha) was significantly greater than  $V_{752}$  (55.7 q/ha). Pertaining to influence of Zn application methods on straw yield of wheat, all were significant in their effect. Maximum straw yield was recorded under Zn<sub>31.25,F</sub> (62.5 q/ha) that was followed by Zn<sub>62.5</sub> (59.4 q/ha) and Zn<sub>0</sub> (56.1 q/ha). Various phytohormone levels had a significant influence on enhancing the straw yield. Maximum was recorded under H<sub>1</sub> (61.6 q/ha) followed by H<sub>3</sub> (60.1 q/ha), H<sub>2</sub> (59.3 q/ha) and H<sub>0</sub> (56.3 q/ha). The levels H<sub>1</sub> & H<sub>3</sub> and H<sub>2</sub> & H<sub>3</sub> showed statistical parity with each other. Overall, the treatment  $V_{725}$  + Zn<sub>31.25,F</sub> + H<sub>1</sub> was the most influential resulted the highest straw yield (69.3 q/ha).

During 2022-23 season, a significant varietal difference was recorded with regard to straw yield. The variety  $V_{725}$  (79.2 q/ha) had significantly higher straw yield than  $V_{752}$  (69.3 q/ha). The influence of Zn application methods on straw yield was found statistically significant. Maximum straw yield resulted under Zn<sub>31.25,F</sub> (76.7 q/ha) that was followed by Zn<sub>62.5</sub> (74.4 q/ha) and Zn<sub>0</sub> (71.6 q/ha). Different levels of phytohormones significantly impacted the straw yield. The highest was recorded under H<sub>1</sub> (77.4 q/ha) that was followed by H<sub>3</sub> (75.1 q/ha), H<sub>2</sub> (73.2 q/ha) and H<sub>0</sub> (71.5 q/ha). Overall, the treatment  $V_{725} + Zn_{31.25,F} + H_1$  was the most effective, resulted in highest straw yield (86.2 q/ha). The interaction effect of variety and Zn on straw yield is presented in table 4.6d. Maximum straw yield was recorded for  $V_{725}+Zn_{31.25,F}$  (82.1 q/ha). The interaction effect of Zn and hormones on straw yield is presented in table 4.6e where the combination of Zn<sub>31.25,F</sub>+H<sub>1</sub> resulted in maximum straw yield of 80.8 q/ha.

Mixed approaches of Zn fertilisation, i.e. soil + foliar application, was found to be superior than single fertilisation methods. It might be attributed to increased auxin production, which played a key role in division of cells and internode elongation in plants (**Ghasal** *et al.*, **2017a**). Thus, combined Zn application (soil + foliar) might be more promising than other fertilisation approaches in enhancing plant development and dry matter at maturity which resulted in higher straw yield (**Imran & Rehim, 2017**). Application of GA led to increased plant height and dry matter which contributed to higher straw yield (**Mathpal** *et al.*, **2023**).

	Zn <sub>0</sub> (No application)	Zn <sub>62.5</sub> (Soil)	Zn <sub>31.25,F</sub> (Soil + Foliar)	Mean	
V725 (PBW 725)	75.8	79.8	82.1	79.2	
V752 (PBW 752)	67.5	69.1	71.4	69.3	
Mean	71.6	74.4	76.7		
V x Zn (CD at $p \le 0.05$ ) = 0.4					

Table 4.6d: Interaction effect of variety and Zn on straw yield (q/ha) of wheat for 2022-23.

Table 4.6e: Interaction effect of Zn and hormones on straw yield (q/ha) of wheat for 2022-23.

	H <sub>0</sub> (No application)	H1 (10 mg/L GA)	H <sub>2</sub> (10 mg/L Cyt)	H <sub>3</sub> (5 mg/L GA + 5 mg/L Cyt)	Mean
Zn <sub>0</sub> (No application)	69.4	73.8	71.1	72.1	71.6
Zn62.5 (Soil)	71.6	77.4	73.3	75.3	74.4
Zn <sub>31.25,F</sub> (Soil + Foliar)	73.4	80.8	75.1	77.7	76.7
Mean	71.5	77.4	73.2	75.1	
Zn x H (CD at p≤ 0.05) = 0.7					

#### 4.11 Harvest index (%)

Data regarding the impact of different treatments on harvest index (HI) is presented in table 4.6. In 2021-22 season, effect of both variety and Zn application methods on HI was statistically non-significant. The maximum HI was recorded in  $V_{752}$  (42.7 %) and minimum in  $V_{725}$  (42.0 %). Among the Zn applications methods, maximum HI was recorded under Zn<sub>0</sub> (42.4 %) followed by Zn<sub>62.5</sub> (42.3 %) and Zn<sub>31.25,F</sub> (42.2 %). A significant effect was recorded for the various levels of plant hormone. The level H<sub>2</sub> (43.2 %) had maximum harvest index followed by H<sub>0</sub> (42.4 %), H<sub>3</sub> (42.3 %) and H<sub>1</sub> (41.3 %). The levels H<sub>2</sub>, H<sub>0</sub> and H<sub>3</sub>; H<sub>0</sub> and H<sub>3</sub>; H<sub>1</sub> and H<sub>3</sub> were statistically at par of each other. In general, the treatment combination  $V_{752}$  + Zn<sub>0</sub> + H<sub>2</sub> was the most effective and resulted in highest harvest index (43.8 %).

Similarly, in 2022-23 season, a non-significant effect was recorded for the varieties and Zn application methods. The higher HI was recorded in  $V_{725}$  (40.0 %) followed by  $V_{752}$  (40.0 %). For the various Zn supply regimes,  $Zn_{31.25,F}$  (40.1 %) resulted in higher HI followed by  $Zn_{62.5}$  (40.1 %) and  $Zn_0$  (40.0 %). The HI was significantly affected by the different levels of plant hormone. Maximum HI was recorded under H<sub>2</sub> (41.3 %) followed by H<sub>3</sub> (40.1 %), H<sub>0</sub> (40.1 %) and H<sub>1</sub> (38.7 %). The level H<sub>0</sub> and H<sub>3</sub> were statistically at par with each other. Overall,  $V_{752}$  +

 $Zn_{31.25,F}$  + H<sub>2</sub> treatment combination was the most influential and resulted in highest harvest index (42.0 %). The interaction effect of Zn and hormones on harvest index is presented in table 4.6f, where  $Zn_{31.25,F}$ +H<sub>2</sub> had highest harvest index (41.9 %). The interaction effect of variety and hormones on harvest index is presented in table 4.6g. Maximum harvest index is recorded for  $V_{752}$ +H<sub>2</sub> (41.5 %).

The harvest index (HI) of a crop determines its efficiency and capacity to convert total dry matter into commercial output. It is the proportion of grains in relation to the overall dry matter of crop. Greater the value of harvest index, more the physiological efficiency of dry matter conversion and vice versa. Soil + foliar method of Zn application increased wheat's yield attributes such as count of effective tillers and grains/spike (**Ghasal** *et al.*, **2017a**). This was further enhanced by foliar spray of PGR which improved grain weight and size (**Mathpal** *et al.*, **2023**). The phytohormones via different mechanisms upregulated grain development. So, the increased effective tillers, grain weight and number of grains enhanced the grain yield that led to higher harvest index.

Table 4.6f: Interaction effect of Zn and hormones on harvest index (HI) of wheat for2022-23.

	H <sub>0</sub> (No application)	H <sub>1</sub> (10 mg/L GA)	H <sub>2</sub> (10 mg/L Cyt)	H <sub>3</sub> (5 mg/L GA + 5 mg/L Cyt)	Mean
Zn <sub>0</sub> (No application)	40.0	39.0	40.9	40.0	40.0
Zn <sub>62.5</sub> (Soil)	40.1	38.7	41.3	40.1	40.1
Zn <sub>31.25,F</sub> (Soil + Foliar)	40.1	38.3	41.9	40.1	40.1
Mean	40.1	38.7	41.3	40.1	
Zn x H (CD at p≤ 0.05) = 0.2					

Table 4.6g: Interaction effect of variety and hormones on harvest index (HI) of wheatfor 2022-23.

	H <sub>0</sub> (No application)	H1 (10 mg/L GA)	H <sub>2</sub> (10 mg/L Cyt)	H <sub>3</sub> (5 mg/L GA + 5 mg/L Cyt)	Mean
V725 (PBW 725)	40.1	38.8	41.2	40.1	40.0
V752 (PBW 752)	40.0	38.6	41.5	40.1	40.0
Mean	40.1	38.7	41.3	40.1	
V x H (CD at $p \le 0.05$ ) = 0.2					

#### 4.12 Grain protein content (%)

Table 4.7 illustrates the influence of different treatments on the grain protein content. During 2021-22 season, there was a significant varietal difference in the grain protein content. The results revealed that the protein content of  $V_{752}$  (12.0 %) was significantly greater than  $V_{725}$  (10.3 %). Regarding the influence of Zn application methods on wheat, all were statistically significant in their effect. The highest grain protein content was recorded under  $Zn_{31.25,F}$  (11.7 %) followed by  $Zn_{62.5}$  (11.1 %) and  $Zn_0$  (10.6 %). A significant impact on grain protein content was recorded on comparing the various phytohormones. Maximum grain protein content was recorded under H<sub>2</sub> (11.5 %) followed by H<sub>3</sub> (11.2 %), H<sub>1</sub> (11.0 %) and H<sub>0</sub> (10.9 %). The levels H<sub>2</sub> and H<sub>3</sub>; H<sub>0</sub>, H<sub>1</sub> and H<sub>3</sub> were statistically similar to each other. Overall, the treatment combination  $V_{752} + Zn_{31.25,F} + H_2$  was the most efficient and resulted in maximum (13.1

%) grain protein content.

During 2022-23 season, protein content in grains was significantly impacted by varieties. Higher grain protein content was recorded in V<sub>752</sub> (12.1 %) followed by V<sub>725</sub> (11.0 %). The impact of Zn application methods was found statistically significant on the grain protein content and maximum was recorded under Zn<sub>31.25,F</sub> (12.1 %) which was significantly greater than Zn<sub>62.5</sub> (11.5 %) and Zn<sub>0</sub> (11.0 %). The method Zn<sub>62.5</sub> was statistically at par with Zn<sub>0</sub>. On comparing the levels of phytohormone, all had a significant impact on grain protein content. Highest grain protein content was recorded under H<sub>2</sub> (11.9 %) followed by H<sub>3</sub> (11.6 %), H<sub>1</sub> (11.4 %) and H<sub>0</sub> (11.3 %). The levels H<sub>2</sub> and H<sub>3</sub>; H<sub>0</sub>, H<sub>1</sub> and H<sub>3</sub> were statistically similar. In general, the maximum grain protein content (13.2 %) was recorded for the V<sub>752</sub> + Zn<sub>31.25,F</sub> + H<sub>2</sub> treatment.

The protein content of grain is a crucial quality parameter of wheat. The protein in wheat is crucial for its use in the baking industry. Wheat with a higher protein content is regarded as highly valuable and suitable for human consumption, whereas wheat with a lower protein content is often offered on the stock-feed market (**Hare, 2017**). The increased grain protein content of wheat variety PBW 752 might be attributed to its genetic superiority (**Singh** *et al.*, **2021**; **Singh** *et al.*, **2023**). Zinc application by both soil and foliar methods resulted in an improved Zn availability to wheat crop. This increase in Zn availability enhanced the activities of nitrate reductase and glutamine synthetase, which had a favourable impact on the grain

protein content (**Tao** *et al.*, **2018**). The application of cytokinin leads to an acceleration in the rates of protein synthesis (**Tepfer and Fosket, 1978**). The observed phenomenon was ascribed to the interactions between cytokinin and tRNA, which enhanced ribosome affinity for aminoacyl-tRNAs, hence improving codon recognition in the polyribosome complex (**Karunadasa** *et al.*, **2020**).

Grain protein content (%)							
Treatments         2021-22         2022-23							
Variety							
V725 (PBW 725)	10.3	11.0					
V752 (PBW 752)	12.0	12.1					
SEm (±)	0.2	0.1					
CD at p ≤ 0.05	1.5	0.4					
	Zinc application modes						
Zn <sub>0</sub> (No application)	10.6	11.0					
Zn62.5 (Soil)	11.1	11.5					
Zn31.25,F (Soil+Foliar)	11.7	12.1					
Zn SEm (±)	0.1	0.2					
Zn (CD at p ≤ 0.05)	0.5	0.6					
V x Zn SEm (±)	0.2	0.3					
V x Zn (CD at p≤0.05)	NS	NS					
	Hormone application						
H₀ (No application)	10.9	11.3					
H1 (10 mg/L GA)	11.0	11.4					
H <sub>2</sub> (10 mg/L Cyt)	11.5	11.9					
H <sub>3</sub> (5 mg/L GA +5 mg/L Cyt)	11.2	11.6					
H SEm (±)	0.1	0.1					
H (CD at p $\leq$ 0.05)	0.4	0.4					
V x H SEm (±)	0.2	0.2					
V x H (CD at p $\leq$ 0.05)	NS	NS					
Zn x H SEm (±)	0.2	0.3					
Zn x H (CD at $p \le 0.05$ )	NS	NS					
V x Zn x H SEm (±)	0.3	0.4					
V x Zn x H (CD at p≤0.05)	NS	NS					

Table 4.7: Effect of various treatments on grain protein content (%) of wheat during2021-22 and 2022-23.

#### 4.13 Grain phytic acid (%)

The impact of different treatments on the grain phytic acid content is illustrated in table 4.8. In 2021-22 season, the comparison between wheat varieties revealed a non-significant effect on the grain phytic acid content. The variety  $V_{752}$  (1.05 %) had more phytic acid than  $V_{725}$  (1.03 %). Pertaining to the impact of different Zn application methods on grain phytic acid content, a significant effect was recorded. Highest concentration of phytic acid in the grain was recorded under Zn<sub>0</sub> (1.12 %) followed by Zn<sub>62.5</sub> (1.02 %) and Zn<sub>31.25,F</sub> (0.97 %). Mode Zn<sub>62.5</sub> was statistically at par with Zn<sub>31.25,F</sub>. The grain phytic acid content was significantly influenced by various plant hormone levels. Maximum grain phytic acid was recorded under H<sub>0</sub> (1.09 %) which was significantly greater than H<sub>1</sub> (1.05 %), H<sub>3</sub> (1.02 %) and H<sub>2</sub> (0.98 %). The levels H<sub>0</sub> and H<sub>1</sub>; H<sub>1</sub> and H<sub>3</sub>; H<sub>2</sub> and H<sub>3</sub> were statistically similar to each other. In general, the treatment combination  $V_{725} + Zn_{31.25,F} + H_2$  was the most effective and resulted in maximum grain phytic acid (0.91 %).

During 2022-23 season, there was a non-significant varietal difference pertaining to grain phytic acid. Maximum was recorded in V<sub>752</sub> (0.98 %) and minimum in V<sub>725</sub> (0.94 %). A significant result was recorded under different methods of Zn application on the grain phytic acid. The method Zn<sub>0</sub> (1.13 %) had significantly higher grain phytic acid content than Zn<sub>62.5</sub> (0.93 %) and Zn<sub>31.25,F</sub> (0.82 %). The method Zn<sub>31.25,F</sub> was statistically at par with Zn<sub>62.5</sub>. Various plant hormones significantly affected the grain phytic acid content and the highest was recorded under H<sub>0</sub> (1.07 %) followed by H<sub>1</sub> (0.99 %), H<sub>3</sub> (0.92 %) and H<sub>2</sub> (0.86 %). The levels H<sub>0</sub> and H<sub>1</sub>; H<sub>1</sub> and H<sub>3</sub>; H<sub>2</sub> and H<sub>3</sub> were statistically similar to each other. Overall, the treatment combination V<sub>725</sub> + Zn<sub>31.25,F</sub> + H<sub>2</sub> was the most effective resulting in highest grain phytic acid (0.70 %).

Phytic acid is a naturally occurring chemical compound present in cereal grains. It acts as the primary reservoir of phosphorus in seeds (**Zarea** *et al.*, **2023**). It is an anti-nutritional compound and has the ability to bind with Zn that leads to deficiency of this mineral (**Kumar** *et al.*, **2017**). Both the varieties did not differ in phytic acid content significantly which indicates that phytic acid was a genetic character of the wheat crop. In present investigation, altered Zn application timing also had a favourable impact on the absorption of Zn. This might be attributed to either a modification in the reaction time or a reduced binding of Zn by the soil P. **Wang** *et al.* (**2015**) found a strong and positive correlation between increasing Zn

levels and enhanced Zn absorption. The increased absorption of Zn had a crucial role in decreasing the content of phytic acid and enhancing the Zn fortification in plants (**Bibi** *et al.*, **2020**). The phytohormones significantly enhanced the Zn content (**Mathpal** *et al.*, **2023**) and potentially contributed in the reduction of phytic acid content of the wheat grains.

Table 4.8: Effect of different treatments on grain phytic acid content (%) of wheat
during 2021-22 and 2022-23.

Grain phytic acid content (%)					
Treatments	2022-23				
	Variety				
V725 (PBW 725)	1.03	0.94			
V752 (PBW 752)	1.05	0.98			
SEm (±)	0.04	0.09			
CD at p ≤ 0.05	NS	NS			
	Zinc application modes				
Zn <sub>0</sub> (No application)	1.12	1.13			
Zn62.5 (Soil)	1.02	0.93			
Zn31.25,F (Soil+Foliar)	0.97	0.82			
Zn SEm (±)	0.02	0.05			
Zn (CD at p ≤ 0.05)	0.08	0.16			
V x Zn SEm (±)	0.03	0.07			
V x Zn (CD at p≤0.05)	NS	NS			
	Hormone application	• -			
H <sub>0</sub> (No application)	1.09	1.07			
H1 (10 mg/L GA)	1.05	0.99			
H <sub>2</sub> (10 mg/L Cyt)	0.98	0.86			
H3 (5 mg/L GA +5 mg/L Cyt)	1.02	0.92			
H SEm (±)	0.02	0.04			
H (CD at p ≤ 0.05)	0.05	0.11			
V x H SEm (±)	0.03	0.05			
V x H (CD at p ≤ 0.05)	NS	NS			
Zn x H SEm (±)	0.03	0.07			
Zn x H (CD at p ≤ 0.05)	NS	NS			
V x Zn x H SEm (±)	0.05	0.09			
V x Zn x H (CD at p≤0.05)	NS	NS			

## 4.14 Grain Zn content

Impact of various treatments on the Zn content in grains of two wheat varieties is presented in table 4.9. In 2021-22 season, comparison of wheat varieties revealed a significant impact on grain Zn content. The variety  $V_{725}$  (32.8 mg/kg) was significantly superior to variety  $V_{752}$  (26.9 mg/kg). Regarding the influence of Zn application methods, all exhibited a significant effect. The maximum grain Zn was recorded under Zn<sub>31.25,F</sub> (33.1 mg/kg) followed by Zn<sub>62.5</sub> (30.0 mg/kg) and Zn<sub>0</sub> (26.3 mg/kg). Influence of different phytohormone levels was found significant on grain Zn content. The highest grain Zn resulted under H<sub>2</sub> (31.3 mg/kg) followed by H<sub>3</sub> (30.1 mg/kg), H<sub>1</sub> (29.5 mg/kg) and H<sub>0</sub> (28.3 mg/kg). The levels H<sub>1</sub> and H<sub>3</sub> showed statistical parity with each other. Overall, treatment combination  $V_{725} + Zn_{31.25,F} + H_2$  resulted in maximum grain Zn content (37.9 mg/kg).

During 2022-23 season, the grain Zn of both the varieties differed significantly, where  $V_{725}$  (40.7 mg/kg) had significantly higher Zn content in grain than  $V_{752}$  (35.8 mg/kg). The influence of Zn application methods on Zn content of wheat grains was found statistically significant. Among all, the maximum grain Zn content resulted under Zn<sub>31.25,F</sub> (40.2 mg/kg) which was followed by Zn<sub>62.5</sub> (37.6 mg/kg) and Zn<sub>0</sub> (36.9 mg/kg). Comparing the different phytohormones, all levels had a significant impact on the grain Zn concentration. Maximum grain Zn was recorded under H<sub>2</sub> (39.6 mg/kg) followed by H<sub>3</sub> (38.2 mg/kg), H<sub>1</sub> (38.0 mg/kg) and H<sub>0</sub> (37.2 mg/kg). The levels H<sub>1</sub> and H<sub>3</sub> were statistically at par with each other. Overall, treatment combination  $V_{725} + Zn_{31.25,F} + H_2$  had the highest impact and resulted in maximum grain Zn content (44.5 mg/kg). The interaction effect of Zn and hormones on grain Zn content during the 2022-23 season is presented in table 4.9a. Maximum grain Zn content was recorded under Zn<sub>31.25,F</sub>+H<sub>2</sub> (42.1 mg/kg) and minimum under Zn0+H0 (36.1 mg/kg).

The significant difference in Zn concentrations between both the varieties may be attributed to the differential capacity of these varieties to absorb and use nutrients from the soil. **Shekari** *et al.* (2015) revealed notable variations in micronutrient concentration among different varieties. Zinc application through soil + foliar method enhanced its content in the plant due to efficient uptake through leaves in addition to the Zn accumulated through roots. Leaves exhibited a higher capacity to absorb and transport Zn to reproductive organs, resulting in greater accumulation as compared to when only applied through the soil. Similar results were also reported by **Ghasal** *et al.*  (2017b) and Mathpal *et al.* (2015). Exogenous application of cytokinin upregulates the expression of CKX (cytokinin oxidase/dehydrogenase) genes which controls the concentration of cytokinin in wheat and increase Zn translocation to grains (Zarea *et al.*, 2023). Furthermore, these hormones regulate the activity of transporter proteins like ZIP that are associated with uptake of Zn by roots and its transport through shoots. Therefore, enhancing the concentration of grain Zn in wheat (Gao *et al.*, 2019; Ren *et al.*, 2023). Mathpal *et al.* (2022) found that applying cytokinin through foliar spray resulted in increased Zn content. Similarly, Wang *et al.* (2017) reported an increase in iron levels in rice seedlings when treated with paclobutrazol, a GA biosynthesis inhibitor (Mathpal *et al.*, 2023). During the 2021-22 season, heat stress was caused during the grain-filling and ripening stage to the crop because both maximum and minimum temperatures increased by over 3 degree Celsius. This resulted in the decreased grain Zn content. The results were supported by the findings of Panigrahi *et al.*, (2022).

## 4.15 Stem Zn content

Table 4.9 illustrates the impact of various treatments on the Zn concentration in the stems. During 2021-22 season, comparison of wheat varieties revealed a significant effect on stem Zn content. The stem Zn content in V<sub>725</sub> (23.7 mg/kg) was significantly higher than V<sub>752</sub> (18.9 mg/kg). The Zn application methods displayed a significant impact on the stem Zn content. Highest concentration of stem Zn resulted under Zn<sub>31.25,F</sub> (23.4 mg/kg) followed by Zn<sub>62.5</sub> (21.1 mg/kg) and Zn<sub>0</sub> (19.4 mg/kg). The different levels of phytohormones had a significant impact on stem Zn content. The highest Zn content in wheat stem was recorded under H<sub>1</sub> (22.5 mg/kg) followed by H<sub>3</sub> (21.5 mg/kg), H<sub>2</sub> (21.0 mg/kg) and H<sub>0</sub> (20.2 mg/kg). The levels H<sub>2</sub> and H<sub>3</sub> showed statistical parity with each other. The treatment combination V<sub>725</sub> + Zn<sub>31.25,F</sub> + H<sub>1</sub> was the most efficient and resulted in maximum Zn content in stem (27.1 mg/kg).

During 2022-23 season, the wheat varieties significantly differed from each other with regard to stem Zn content. The Zn content in  $V_{725}$  (26.8 mg/kg) was significantly superior to  $V_{752}$  (22.5 mg/kg). Various Zn application methods significantly affected the Zn content in stem. Maximum stem Zn content resulted under Zn<sub>31.25,F</sub> (26.2 mg/kg) followed by Zn<sub>62.5</sub> (25.4 mg/kg) and Zn<sub>0</sub> (22.3 mg/kg). Application of phytohormones had a significant influence on the stem Zn content and the highest was recorded under H<sub>1</sub> (25.6 mg/kg) followed by H<sub>3</sub> (24.9 mg/kg), H<sub>2</sub>

(24.5 mg/kg) and H<sub>0</sub> (23.5 mg/kg). The level H<sub>2</sub> was statistically at par with H<sub>3</sub>. The treatment combination  $V_{725} + Zn_{31,25,F} + H_1$  was the most efficient and resulted in highest stem Zn content (29.9 mg/kg).

Table 4.9: Effect of various treatments on Zn content (mg/kg) in different parts of wheat during 2021-22 and 2022-23.

		Zn content	(mg/kg)				
	2021-22	2022-23	2021-22	2022-23	2021-22	2022-23	
Treatments	Grain Zn content (mg/kg)			Stem Zn content (mg/kg)		Leaf Zn content (mg/kg)	
		Varie	80 B.	5 5/		, 0/	
V725 (PBW 725)	32.8	40.7	23.7	26.8	9.9	10.5	
V752 (PBW 752)	26.9	35.8	18.9	22.5	7.8	8.7	
SEm (±)	0.01	0.1	0.2	0.2	0.2	0.1	
CD at $p \le 0.05$	0.1	0.4	1.3	0.9	1.4	0.3	
		Zinc applicat	ion modes				
Zno (No application)	26.3	36.9	19.4	22.3	7.9	8.9	
Zn62.5 (Soil)	30.0	37.6	21.1	25.4	8.8	9.8	
Zn31.25,F (Soil+Foliar)	33.1	40.2	23.4	26.2	9.9	10.2	
Zn SEm (±)	0.1	0.1	0.3	0.1	0.2	0.1	
Zn (CD at p ≤ 0.05)	0.4	0.3	0.8	0.4	0.5	0.3	
V x Zn SEm (±)	0.2	0.1	0.4	0.2	0.2	0.1	
V x Zn (CD at p≤ 0.05)	NS	NS	NS	NS	NS	NS	
		Hormone ap	plication				
H <sub>0</sub> (No application)	28.3	37.2	20.2	23.5	8.0	9.1	
H1 (10 mg/L GA)	29.5	38.0	22.5	25.6	9.6	10.0	
H <sub>2</sub> (10 mg/L Cyt)	31.3	39.6	21.0	24.5	8.7	9.6	
H3 (5 mg/L GA +5 mg/L Cyt)	30.1	38.2	21.5	24.9	9.1	9.7	
H SEm (±)	0.2	0.1	0.4	0.2	0.1	0.1	
H (CD at p ≤ 0.05)	0.6	0.4	1.0	0.5	0.4	0.2	
V x H SEm (±)	0.3	0.2	0.5	0.3	0.3	0.1	
V x H (CD at $p \le 0.05$ )	NS	NS	NS	NS	NS	NS	
Zn x H SEm (±)	0.4	0.2	0.6	0.3	0.3	0.1	
Zn x H (CD at p ≤ 0.05)	NS	0.6	NS	NS	NS	NS	
V x Zn x H SEm (±)	0.6	0.3	0.8	0.4	0.5	0.2	
V x Zn x H (CD at p≤ 0.05)	NS	NS	NS	NS	NS	NS	

	H <sub>0</sub> (No application)	H1 (10 mg/L GA)	H <sub>2</sub> (10 mg/L Cyt)	H <sub>3</sub> (5 mg/L GA + 5 mg/L Cyt)	Mean
Zn <sub>0</sub> (No application)	36.1	36.9	37.8	36.8	36.9
Zn <sub>62.5</sub> (Soil)	36.7	37.3	38.9	37.7	37.6
Zn <sub>31.25,F</sub> (Soil + Foliar)	38.9	39.7	42.1	40.2	40.2
Mean	37.2	38.0	39.6	38.2	
Zn x H (CD at p≤ 0.05) = 0.6					

Table 4.9a: Interaction effect of Zn and hormones on grain Zn content (mg/kg) of wheat for 2022-23.

 $2\pi \times \pi (0) = 0.00 = 0.00$ 

The differential capacity of nutrient absorption from the soil might be the reason of variation in stem Zn content between the two varieties (**Shekari** *et al.*, **2015**). Soil + foliar method of Zn application enhanced concentration of Zn in the plant due to efficient uptake as absorption through leaves minimise the losses associated with absorption via roots. Similar results were also reported by **Ghasal** *et al.* (2017b) and **Mathpal** *et al.* (2015). The results can be attributed to the influence of GA, which regulates the transporter proteins that facilitate the absorption and transportation of Zn. In addition to its involvement in regulating the expression of transporter proteins, GA also mitigate heavy metal toxicity (**Ren** *et al.*, 2023). The results of our study aligned with the findings of **Kamboj and Mathpal** (2019), which reported an increase in Zn levels in rice when 20 mg/L GA was applied.

# 4.16 Leaf Zn content

Table 4.9 presents the influence of different treatments on the Zn content of leaves. During 2021-22 season, a significant result was recorded on comparing the wheat varieties. The leaf Zn content in  $V_{725}$  (9.9 mg/kg) was significantly higher than  $V_{752}$  (7.8 mg/kg). Pertaining to different Zn application methods, all significantly impacted the Zn content in leaves of wheat. Maximum content of leaf Zn was recorded under Zn<sub>31.25,F</sub> (9.9 mg/kg) followed by Zn<sub>62.5</sub> (8.8 mg/kg) and Zn<sub>0</sub> (7.9 mg/kg). The various levels of plant hormones had a significant influence on the leaf Zn content. The highest leaf Zn content resulted under H<sub>1</sub> (9.6 mg/kg) followed by H<sub>3</sub> (9.1 mg/kg), H<sub>2</sub> (8.7 mg/kg) and H<sub>0</sub> (8.0 mg/kg). The levels H<sub>2</sub> and H<sub>3</sub> were statistically at par with each other. The treatment combination  $V_{725} + Zn_{31.25,F} + H_1$  was most effective with highest Zn content of leaves (12.1 mg/kg).

During 2021-22 season, the leaf Zn content was significantly affected by the varieties. Maximum leaf Zn was recorded in  $V_{725}$  (10.5 mg/kg) followed by  $V_{752}$  (8.7 mg/kg). Concerning the impact of different Zn application methods on leaf Zn content, a significant result was recorded. The maximum content was recorded under Zn<sub>31.25,F</sub> (10.2 mg/kg) followed by Zn<sub>62.5</sub> (9.8 mg/kg) and Zn<sub>0</sub> (8.8 mg/kg). The various plant hormone levels significantly affected the leaf Zn content. Highest Zn content in leaves resulted under H<sub>1</sub> (10.0 mg/kg) followed by H<sub>3</sub> (9.7 mg/kg), H<sub>2</sub> (9.6 mg/kg) and H<sub>0</sub> (9.1 mg/kg). The levels H<sub>2</sub> and H<sub>3</sub> were statistically at par with each other. The V<sub>725</sub> + Zn<sub>31.25,F</sub> + H<sub>1</sub> treatment combination was the most effective, resulting in the highest leaf Zn content (11.7 mg/kg).

The substantial variation in Zn contents between both the varieties might be linked to the varied capacities of absorption and utilisation of nutrients from the soil. **Shekari** *et al.* (2015) discovered significant variability in micronutrient content across various varieties. The application of Zn by soil+foliar method boosted its concentration in the plant owing to excellent absorption via leaves in addition to Zn accumulated through the plant roots. Leaves had a stronger potential to absorb Zn leading to its greater accumulation. Similar results were also reported by **Ghasal** *et al.* (2017b) and **Mathpal** *et al.* (2015). The transporter proteins that are involved in absorption and transport of Zn inside the plant system are being regulated by GA. Also, GA has been shown to reduce heavy metal toxicity, which confirm its role in nutrient remobilization (**Ren** *et al.*, 2023). Our findings correspond with those of **Kamboj and Mathpal** (2019), which reported an increased Zn content when GA was applied foliarly.

# 4.17 Soil Zn content

The impact of various treatments on DTPA extractable soil Zn content is presented in table 4.10. Soil analysis after 2021-22 season revealed that the two varieties showed non-significant impact on soil Zn content. The value of soil Zn content in plots allocated for V<sub>725</sub> was 0.49 mg/kg which was same to soil Zn content recorded in plots for V<sub>752</sub>. Regarding Zn application methods, the affect was found to be statistically significant. The soil Zn content in plots for Zn<sub>62.5</sub> (0.52 mg/kg) was significantly higher than Zn<sub>31.25,F</sub> (0.50 mg/kg) and Zn<sub>0</sub> (0.44 mg/kg). The methods Zn<sub>2</sub> and Zn<sub>1</sub> were statistically at par with each other. In context to different hormone levels, the result was found non-significant. The value recorded for soil Zn from plots under H<sub>2</sub>, H<sub>3</sub>, H<sub>1</sub> and H<sub>0</sub> was 0.49 mg/kg. Overall, treatment  $V_{725} + Zn_{31.25,F} + H_2$  and  $V_{752}$ 

+ Zn<sub>62.5</sub> + H<sub>2</sub> resulted in the maximum Zn content (0.52 mg/kg) in soil.

Soil zinc content (mg/kg)					
Treatments	Soil after 2021-22 season	Soil after 2022-23 season			
	Variety				
V725 (PBW 725)	0.49	0.50			
V <sub>752</sub> (PBW 752)	0.48	0.50			
SEm (±)	0.01	0.003			
CD at p ≤ 0.05	NS	NS			
	Zinc application modes				
Zn₀ (No application)	0.44	0.42			
Zn62.5 (Soil)	0.52	0.56			
Zn31.25,F (Soil+Foliar)	0.50	0.52			
Zn SEm (±)	0.01	0.002			
Zn (CD at p ≤ 0.05)	0.02	0.01			
V x Zn SEm (±)	0.01	0.002			
V x Zn (CD at p≤0.05)	NS	NS			
	Hormone application				
H₀ (No application)	0.49	0.50			
H1 (10 mg/L GA)	0.49	0.50			
H2 (10 mg/L Cyt)	0.49	0.50			
H <sub>3</sub> (5 mg/L GA +5 mg/L Cyt)	0.49	0.50			
H SEm (±)	0.001	0.001			
H (CD at p ≤ 0.05)	NS	NS			
V x H SEm (±)	0.001	0.002			
V x H (CD at $p \le 0.05$ )	NS	NS			
Zn x H SEm (±)	0.001	0.002			
Zn x H (CD at p ≤ 0.05)	NS	NS			
V x Zn x H SEm (±)	0.002	0.003			
V x Zn x H (CD at p≤0.05)	NS	NS			

# Table 4.10: Effect of different treatments on soil Zn content (mg/kg) of wheat during 2021-22 and 2022-23.

Similarly, the soil Zn content after 2022-23 season, the influence of varieties was statistically non-significant. Soil Zn content in plots allocated for V<sub>725</sub> was 0.50 mg/kg which was same to soil Zn content recorded in plots for V<sub>752</sub>. The different Zn application methods significantly influenced soil Zn content. The highest soil Zn content was recorded from plots under Zn<sub>62.5</sub> (0.56 mg/kg) which was significantly higher than Zn<sub>31.25,F</sub> (0.52 mg/kg) and Zn<sub>0</sub> (0.42 mg/kg). The various plant hormones had non-significant impact on the soil Zn content. The value of soil Zn content resulted from plots under H<sub>2</sub>, H<sub>3</sub>, H<sub>1</sub> and H<sub>0</sub> was 0.50 mg/kg. Overall, treatment V<sub>725</sub> + Zn<sub>62.5</sub> + H<sub>3</sub> and V<sub>752</sub> + Zn<sub>62.5</sub> + H<sub>3</sub> resulted in the highest Zn content (0.56 mg/kg) in soil. The reduction in soil Zn was recorded under no application of Zn, however, an increase in soil Zn content was found under soil application. The results of this study were endorsed by **Keram** *et al.* (2012), Dhaliwal *et al.* (2019) and Amjadian *et al.* (2021).

Benefit-cost ratio					
Treatments	2021-22	2022-23			
	Variety				
V <sub>725</sub> (PBW 725)	2.0	2.6			
V752 (PBW 752)	1.9	2.3			
	Zinc application modes				
Zn <sub>0</sub> (Control)	2.0	2.3			
Zn <sub>62.5</sub> (Soil)	1.8	2.4			
Zn <sub>31.25,F</sub> (Soil + Foliar)	2.0	2.5			
	Hormone application				
H <sub>0</sub> (Control)	1.9	2.3			
H <sub>1</sub> (10 mg/L GA)	1.9	2.4			
H <sub>2</sub> (10 mg/L Cyt)	2.0	2.5			
H <sub>3</sub> (5 mg/L GA+5 mg/L Cyt)	2.0	2.5			

Table 4.11: Effect of different treatments on benefit-cost ratio of wheat during 2021-22and 2022-23.

## 4.18 Benefit-cost ratio (B:C ratio)

The benefit-cost ratio expresses the extent of benefit or profit earned by applying a particular treatment over its cost. It was calculated for the different treatment combinations in both the seasons and presented in table 4.11. During 2021-22 season, the benefit-cost ratio in  $V_{725}$  (2.0) followed by  $V_{752}$  (1.9). Pertaining to different Zn application methods, Zn<sub>31.25,F</sub> and Zn<sub>0</sub> had B:C ratio of 2.0 followed by

Zn<sub>62.5</sub> (1.8). Among the various levels of plant hormones, the highest B:C ratio was recorded in H<sub>2</sub> and H<sub>3</sub> (2.0) followed by H<sub>0</sub> and H<sub>1</sub> (1.9). The treatment combination  $V_{725} + Zn_{31.25,F} + H_2$  was most effective with B:C ratio (2.2). During 2022-23 season, the benefit-cost ratio in  $V_{725}$  (2.6) followed by  $V_{752}$  (2.3). Pertaining to different Zn application methods, Zn<sub>31.25,F</sub> had highest B:C ratio (2.5) followed by Zn<sub>62.5</sub> (2.4) and Zn<sub>0</sub> (2.3). Among the various levels of plant hormones, the maximum B:C ratio was recorded in H<sub>2</sub> and H<sub>3</sub> (2.5) which was followed by H<sub>1</sub> (2.4) and H<sub>0</sub> (2.3). The treatment combination  $V_{725} + Zn_{31.25,F} + H_2$  was most effective with maximum B:C ratio (2.8).

# CHAPTER – V

# SUMMARY AND CONCLUSIONS

The field experiment entitled "**Response of various zinc application methods and concentrations of phytohormones on yield and grain enrichment of zinc in early and late sown wheat** (*Triticum aestivum* L.) varieties" was conducted at agriculture farm, School of Agriculture, Lovely Professional University, Phagwara, Punjab during the rabi season of 2021-22 and 2022-23. The first objective was to study the effect of various methods of zinc application on zinc uptake, yield and quality of early and late sown wheat varieties. The second objective was to evaluate the impact of various concentrations of phytohormones on zinc uptake, transport and quality of early and late sown wheat varieties. The third objective was to study the comparative economics of various methods of zinc application and concentrations of phytohormones on early and late sown wheat varieties. While the last objective was to evaluate the effect of various methods of zinc application on nutrient status of the soil.

The low concentration of zinc (Zn) in cereal crops is primarily caused by Zndeficient soils and poor transportation from the source to sink. The study examined the impact of Zn, cytokinin and gibberellic acid (GA) on wheat growth, yield and quality. The design of experiment was split-split plot replicated thrice for each treatment. The main plots consisted of two varieties, namely V<sub>725</sub> (PBW 725) and V<sub>752</sub> (PBW 752) that were split into three sub plots: Zn<sub>0</sub> (no zinc application), Zn<sub>62.5</sub> (62.5 kg/ha of zinc sulphate applied to the soil) and Zn<sub>31.25,F</sub> (31.25 kg/ha of zinc sulphate applied to the soil together with a foliar spray of 0.5% zinc sulphate solution). The subplots were further split into four sub-sub plots, namely H<sub>0</sub> (no hormone application), H<sub>1</sub> (10 mg/L GA), H<sub>2</sub> (10 mg/L cytokinin) and H<sub>3</sub> (5 mg/L GA plus 5 mg/L cytokinin).

A brief summary of experiment findings are given below.

- 1. Height of plants was measured at 30, at 60, at 90, at 120 DAS and at harvest. All the treatments significantly affected the plant height at all growth stages. The maximum plant height was recorded at harvest and it was 104.0 cm and 110.2 cm during 2021-22 and 2022-23 seasons, respectively under treatment  $V_{725}$ +Zn<sub>31.25,F</sub>+H<sub>1</sub>. The minimum plant height was recorded under treatment  $V_{752}$ +Zn<sub>0</sub>+H<sub>0</sub> and it was 78.9 cm and 83.6 cm during 2021-22 and 2022-23 seasons, respectively.
- 2. The maximum number (402.3 and 423.2) of tillers were recorded at 120 DAS

during 2021-22 and 2022-23 seasons, respectively. They were maximum under the treatment  $V_{725}$ + $Zn_{31.25,F}$ + $H_2$  while the lowest number (277.0 and 340.5) of tillers were recorded under the treatment  $V_{752}$ + $Zn_0$ + $H_0$  during 2021-22 and 2022-23 seasons, respectively.

- The leaf area index (LAI) was maximum at 90 days after sowing (DAS). During the 2021-22 season, maximum LAI was 4.5 under the treatment V<sub>725</sub>+Zn<sub>31.25,F</sub>+H<sub>2</sub>. Similarly, maximum LAI was 5.7 under same treatment during 2022-23 season. The minimum LAI (3.5 and 4.5) was recorded under treatment V<sub>752</sub>+Zn<sub>0</sub>+H<sub>0</sub> during both 2021-22 and 2022-23 seasons, respectively.
- 4. The highest accumulation of dry matter was recorded at 120 DAS. Maximum dry matter was 903.3 g/m<sup>2</sup> during 2021-22 season and 1162.0 g/m<sup>2</sup> during 2022-23 season under treatment V<sub>725</sub>+Zn<sub>31.25,F</sub>+H<sub>1</sub> while, the minimum dry matter accumulation was 653.0 g/m<sup>2</sup> and 821.5 g/m<sup>2</sup> under treatment V<sub>752</sub>+Zn<sub>0</sub>+H<sub>0</sub> during 2021-22 and 2022-23 seasons, respectively.
- The highest number (351.62 and 378.33) of effective tillers were recorded under treatment V<sub>725</sub>+Zn<sub>31.25,F</sub>+H<sub>2</sub> during 2021-22 and 2022-23 seasons, respectively. The lowest number (233.04 and 296.33) of effective tillers were recorded under treatment V<sub>752</sub>+Zn<sub>0</sub>+H<sub>0</sub> during 2021-22 and 2022-23 seasons, respectively.
- 6. The length of spike was significantly influenced by different treatments applied in the experiment. The maximum spike length during 2021-22 season was 11.0 cm under treatment V<sub>725</sub>+Zn<sub>31.25,F</sub>+H<sub>1</sub> while maximum spike length during 2022-23 season was 12.3 cm under treatment V<sub>725</sub>+Zn<sub>31.25,F</sub>+H<sub>2</sub>. The minimum spike length (8.8 cm and 9.9 cm) was recorded under V<sub>752</sub>+Zn<sub>0</sub>+H<sub>0</sub> during 2021-22 and 2022-23 seasons, respectively.
- 7. The number of grains/spike directly correlates to the grain yield. The maximum number of grains per spike were recorded under V<sub>725</sub>+Zn<sub>31.25,F</sub>+H<sub>2</sub> in both crop seasons. During the 2021-22 season, the number of grains were 50.0 and during 2022-23 season, there were 58.3 grains. The lowest number of grains per spike (34.3 and 41.9) were recoded under V<sub>752</sub>+Zn<sub>0</sub>+H<sub>0</sub> treatment during 2021-22 and 2022-23 seasons, respectively.
- 8. The 1000-grain weight indicates heaviness and thickness of the grains. The heaviest grains were recorded under V<sub>725</sub>+Zn<sub>31.25,F</sub>+H<sub>2</sub> treatment. The weight recorded during 2021-22 season was 45.3 grams and during 2022-23 season was 47.6 grams. The lightest grains were recorded under V<sub>752</sub>+Zn<sub>0</sub>+H<sub>0</sub> treatment during both the seasons. The weight recorded was 38.7 grams and 39.2 grams

during 2021-22 and 2022-23 seasons, respectively.

- 9. Different treatments significantly affected the grain yield during both the seasons. The highest grain yield (49.3 q/ha and 57.6 q/ha) was recorded under V<sub>725</sub>+Zn<sub>31.25,F</sub>+H<sub>2</sub> during 2021-22 and 2022-23 seasons, respectively. While the minimum grain yield (37.3 q/ha and 43.6 q/ha) was recorded under V<sub>752</sub>+Zn<sub>0</sub>+H<sub>0</sub> treatment during 2021-22 and 2022-23 seasons, respectively.
- 10. The highest straw yield (69.3 q/ha and 86.2 q/ha) was recorded under V<sub>725</sub>+Zn<sub>31.25,F</sub>+H<sub>1</sub> during 2021-22 and 2022-23 seasons, respectively. The minimum straw yield (51.7 q/ha and 65.4 q/ha) was recorded under V<sub>752</sub>+Zn<sub>0</sub>+H<sub>0</sub> treatment during 2021-22 and 2022-23 seasons, respectively.
- 11. Maximum harvest index was recorded 43.8 % under treatment  $V_{752}+Zn_{31.25,F}+H_2$ during 2021-22 season and 42.0 % under same treatment during 2022-23 season. The minimum harvest index was recorded under  $V_{725}+Zn_{31.25,F}+H_1$  (40.2 %) during 2021-22 season and under  $V_{752}+Zn_{31.25,F}+H_1$  (39.0 %) during 2022-23 season.
- 12. The highest grain protein content (13.1 %) was recorded under  $V_{752}+Zn_{31.25,F}+H_2$ treatment during 2021-22 season and it was 13.2 % under the same treatment during 2022-23 season. The lowest grain protein content (9.5 % and 10.2 %) was recorded under  $V_{725}+Zn_0+H_0$  for both the seasons, respectively.
- 13. The lowest grain phytic acid content was recorded under V<sub>725</sub>+Zn<sub>31.25,F</sub>+H<sub>2</sub> (0.91 %) during 2021-22 and it was 0.70 % under the same treatment during 2022-23. The highest grain phytic acid content was recorded under treatment V<sub>752</sub>+Zn<sub>0</sub>+H<sub>0</sub>. The content was 1.19 % during 2021-22 season and 1.27 % during 2022-23 season.
- 14. The highest grain Zn content (37.9 mg/kg) was recorded under the  $V_{725}+Zn_{31.25,F}+H_2$  treatment during 2021-22 and 44.5 mg/kg during 2022-23 season. The minimum grain Zn content (22.4 mg/kg and 33.8 mg/kg) was recorded under the treatment  $V_{752}+Zn_0+H_0$  during 2021-22 and 2022-23 seasons, respectively.
- 15. The highest Zn content in stem was recorded under the V<sub>725</sub>+Zn<sub>31.25,F</sub>+H<sub>1</sub> treatment. The stem Zn content was 27.1 mg/kg during 2021-22 season and 29.9 mg/kg during 2022-23 season. The minimum stem Zn content (16.0 mg/kg and 19.5 mg/kg) was recorded under V<sub>752</sub>+Zn<sub>0</sub>+H<sub>0</sub> treatment during 2021-22 and 2022-23 seasons, respectively.

- 16. Similarly, the leaf Zn content was maximum under  $V_{725}+Zn_{31.25,F}+H_1$  treatment. The leaf Zn content was 12.1 mg/kg during 2021-22 season and 11.7 mg/kg during 2022-23 season. The minimum leaf Zn content (6.4 mg/kg and 7.7 mg/kg) was recorded under  $V_{752}+Zn_0+H_0$  treatment during 2021-22 and 2022-23 seasons, respectively.
- 17. The DTPA extractable soil Zn was mainly affected by the Zn application modes, however in context to the effect of different treatment used in the experiment it was highest under  $V_{725}$ +Zn<sub>62.5</sub>+H<sub>2</sub> (0.52 mg/kg) after 2021-22 season and under  $V_{725}$ +Zn<sub>62.5</sub>+H<sub>3</sub> (0.56 mg/kg) after 2022-23 season. The lowest soil Zn content was recorded under the treatment  $V_{752}$ +Zn<sub>0</sub>+H<sub>0</sub>.
- 18. The benefit-cost ratio was calculated and it was maximum under treatment  $V_{725}+Zn_{31.25,F}+H_2$ . During the 2021-22 season, the benefit-cost ratio was 2.4 while in season 2022-23, it was 2.7. The minimum benefit cost ratio was recorded under  $V_{752}+Zn_{62.5}+H_0$  during season 2021-22 while during 2022-23 season  $V_{752}+Zn_{62.5}+H_1$  resulted minimum.

Availability of Zn to the wheat crop decreases over time due to its fixation in soil. There are certain internal barriers which hinder its transport from root to shoots. This lead to stunted growth, yield reduction and poor quality of grains. This can overcome by foliar application of Zn on the foliage as leaves have higher efficiency for nutrient absorption. Furthermore, the source to sink translocation of Zn face multiple barriers such as low sink capacity and fixation by phytate inside the plant. These problems can be minimized by using phytohormones such as cytokinin and GA which improves translocation of Zn. These hormones positively affect proteins that are involved in Zn transport. The combined use of Zn and phytohormones enhanced the yield and quality of the crop and had higher benefit-cost ratio than the conventional practices justifying the additional expenditure of money for their use. The best results were recorded under the  $V_{725}$ +Zn<sub>31.25,F</sub>+H<sub>2</sub> treatment. Further studies are required for better understanding regarding the use of phytohormones in crop improvement under different climatic conditions.

- Abdel-Aal, E. M., Sosulski, F. W., Youssef, M. M., & Shehata, A. A. Y. (1993). Selected nutritional, physical and sensory characteristics of pan and flat breads prepared from composite flours containing fababean. *Plant Foods for Human Nutrition*, 44, 227-239.
- Abdel-Aal, E. S. M., Sosulski, F. W., & Hucl, P. (1998). Origins, characteristics, and potentials of ancient wheats. *Cereal Foods World*, *43*, 708–715.
- Achard, P., Cheng, H., De Grauwe, L., Decat, J., Schoutteten, H., Moritz, T., ... & Harberd, N. P. (2006). Integration of plant responses to environmentally activated phytohormonal signals. *Science*, 311(5757), 91-94.
- Achard, P., Renou, J. P., Berthomé, R., Harberd, N. P., & Genschik, P. (2008). Plant DELLAs restrain growth and promote survival of adversity by reducing the levels of reactive oxygen species. *Current biology*, 18(9), 656-660.
- Adams, M. L., Lombi, E., Zhao, F. J., & McGrath, S. P. (2002). Evidence of low selenium concentrations in UK bread-making wheat grain. *Journal of the Science of Food and Agriculture*, 82(10), 1160-1165.
- Adil, M., Bashir, S., Bashir, S., Aslam, Z., Ahmad, N., Younas, T., ... & Elshikh, M. S. (2022). Zinc oxide nanoparticles improved chlorophyll contents, physical parameters, and wheat yield under salt stress. *Frontiers in plant science*, 13, 932861.
- Ahmed, S., Ahmad, M., Swami, B. L., & Ikram, S. (2016). A review on plants extract mediated synthesis of silver nanoparticles for antimicrobial applications: a green expertise. *Journal of advanced research*, 7(1), 17-28.
- Aiqing, Z., Zhang, L., Ning, P., Chen, Q., Wang, B., Zhang, F., Yang, X. & Zhang, Y. (2022). Zinc in cereal grains: Concentration, distribution, speciation, bioavailability, and barriers to transport from roots to grains in wheat. *Critical reviews in food science and nutrition*, 62(28), 7917-7928.
- Ajiboye, B., Cakmak, I., Paterson, D., De Jonge, M. D., Howard, D. L., Stacey, S. P.,
  ... & McLaughlin, M. J. (2015). X-ray fluorescence microscopy of zinc localization in wheat grains biofortified through foliar zinc applications at different growth stages under field conditions. *Plant and soil*, 392, 357-370.

- Alharby, H. F., Rizwan, M., Iftikhar, A., Hussaini, K. M., ur Rehman, M. Z., Bamagoos, A. A., ... & Ali, S. (2021). Effect of gibberellic acid and titanium dioxide nanoparticles on growth, antioxidant defense system and mineral nutrient uptake in wheat. *Ecotoxicology and environmental safety*, 221, 112436.
- Al-Huqail, A. A., Alshehri, D., Nawaz, R., Irshad, M. A., Iftikhar, A., Hussaini, K. M., ... & Abeed, A. H. (2023). The effect of gibberellic acid on wheat growth and nutrient uptake under combined stress of cerium, zinc and titanium dioxide nanoparticles. *Chemosphere*, 336, 139199.
- Alloway, B. J. (2008). Zinc in soils and crop nutrition. 2nd ed.; International Zinc Association: Brussels, Belgium; International Fertilizer Industry Association: Paris, France.
- Alloway, B. J. (2009). Soil factors associated with zinc deficiency in crops and humans. *Environmental geochemistry and health*, *31*(5), 537-548.
- Alzaayid, D. T. J., & Aloush, R. H. (2021, November). Effect of cytokinin levels on some varieties of wheat on yield, growth and yield components. In *IOP Conference Series: Earth and environmental science*, 910, 1, 012090.
- Amjadian, E., Zeinodini, A., & Doğan, H. (2021). Effect of fertilizer management systems on growth and balance of nutrients in wheat cultivation. *Central* asian journal of plant science innovation, 1(2), 56-69.
- Anguizola, J., Matsuda, R., Barnaby, O. S., Hoy, K. S., Wa, C., DeBolt, E., ... & Hage, D. S. (2013). Glycation of human serum albumin. *Clinica chimica* acta, 425, 64-76.
- Anonymous, (2023). <u>https://www.statista.com/statistics/237912/global-top-wheat-producing-countries/.</u>

Anonymous,(2024).<u>https://agriwelfare.gov.in/en/Agricultural\_Statistics\_at\_a\_Glance</u>.

Arciniegas-Grijalba, P. A., Patino-Portela, M. C., Mosquera-Sánchez, L. P., Sierra, B. G., Muñoz-Florez, J. E., Erazo-Castillo, L. A., & Rodríguez-Páez, J. E. (2019). ZnO-based nanofungicides: Synthesis, characterization and their effect on the coffee fungi *Mycena citricolor* and Colletotrichum sp. *Materials science and engineering: C*, 98, 808-825.

- Arduini, I., Masoni, A., Ercoli, L., & Mariotti, M. (2006). Grain yield, and dry matter and nitrogen accumulation and remobilization in durum wheat as affected by variety and seeding rate. *European journal of agronomy*, 25(4), 309-318.
- Arif, M., Dashora, L. N., Choudhary, J., Kadam, S. S., & Mohsin, M. (2019). Effect of nitrogen and zinc management on growth, yield and economics of bread wheat (*Triticum aestivum*) varieties. *Indian journal of agricultural sciences*, 89(10), 1664-8.
- Arif, M., Tasneem, M., Bashir, F., Yaseen, G., & Anwar, A. (2017). Evaluation of different levels of potassium and zinc fertilizer on the growth and yield of wheat. *International journal of biosensors & bioelectronics*, 3(2), 1-5.
- Arshad, M. A. (2021). A review on wheat management, strategies, current problems and future perspectives. *Haya: the saudi journal of life science*, *6*, 14-18.
- Asadi, E., Ghehsareh, A. M., Moghadam, E. G., Hoodaji, M., & Zabihi, H. R. (2019). Improvement of pomegranate colorless arils using iron and zinc fertilization. *Journal of cleaner production*, 234, 392-399.
- Bakhoum, G. S., Tawfik, M. M., Kabesh, M. O., & Sadak, M. S. (2023). Potential role of algae extract as a natural stimulating for wheat production under reduced nitrogen fertilizer rates and water deficit. *Biocatalysis and* agricultural biotechnology, 51, 102794.
- Banaszak, M., Górna, I., & Przysławski, J. (2021). Zinc and the innovative zinc-α2glycoprotein adipokine play an important role in lipid metabolism: a critical review. *Nutrients*, 13(6), 2023.
- Barreto, M. S. C., Elzinga, E. J., Rouff, A. A., Siebecker, M. G., Sparks, D. L., & Alleoni, L. R. F. (2024). Zinc speciation in highly weathered tropical soils affected by large scale vegetable production. *Science of the total environment*, 916, 170223.
- Baublis, A. J., Lu, C., Clydesdale, F. M., & Decker, E. A. (2000). Potential of wheatbased breakfast cereals as a source of dietary antioxidants. *Journal of the American College of Nutrition*, 19(3), 308-311.
- Belcar, J., Buczek, J., Kapusta, I., & Gorzelany, J. (2022). Quality and pro-healthy properties of belgian witbier-style beers relative to the cultivar of winter wheat and raw materials used. *Foods*, 11(8), 1150.

- Bernhardt, M. L., Kong, B. Y., Kim, A. M., O'Halloran, T. V., & Woodruff, T. K. (2012). A zinc-dependent mechanism regulates meiotic progression in mammalian oocytes. *Biology of reproduction*, 86(4), 114-1.
- Betrie, A. H., Brock, J. A., Harraz, O. F., Bush, A. I., He, G. W., Nelson, M. T., ... & Ayton, S. (2021). Zinc drives vasorelaxation by acting in sensory nerves, endothelium and smooth muscle. *Nature communications*, 12(1), 3296.
- Bharti, K., Pandey, N., Shankhdhar, D., Srivastava, P., & Shankhdhar, S. (2013). Evaluation of some promising wheat genotypes (*Triticum aestivum* L.) at different zinc regimes for crop production. *Cereal research communications*, 41(4), 539-549.
- Bibi, F., Saleem, I. S., Ehsan, S., Jamil, S., Ullah, H., Mubashir, M., ... & Danish, S. (2020). Effect of various application rates of phosphorus combined with different zinc rates and time of zinc application on phytic acid concentration and zinc bioavailability in wheat. *Agriculture and natural resources*, 54(3), 265-272.
- Blechl, A., Lin, J., Nguyen, S., Chan, R., Anderson, O. D., & Dupont, F. M. (2007). Transgenic wheats with elevated levels of Dx5 and/or Dy10 high-molecularweight glutenin subunits yield doughs with increased mixing strength and tolerance. *Journal of Cereal Science*, 45(2), 172-183.
- Bradford, M. M. (1976). A rapid and sensitive method for the quantitation of microgram quantities of protein utilizing the principle of protein-dye binding. *Analytical biochemistry*, 72(1-2), 248-254.
- Brennan, R. F. (2001). Residual value of zinc fertiliser for production of wheat. *Australian Journal of Experimental Agriculture*, *41*(4), 541-547.
- Brown, K. H., Peerson, J. M., Baker, S. K., & Hess, S. Y. (2009). Preventive zinc supplementation among infants, preschoolers, and older prepubertal children. *Food and nutrition bulletin*, 30(1), 12-40.
- Brugière, N., Humbert, S., Rizzo, N., Bohn, J., & Habben, J. E. (2008). A member of the maize isopentenyl transferase gene family, *Zea mays* isopentenyl transferase 2 (ZmIPT2), encodes a cytokinin biosynthetic enzyme expressed during kernel development: Cytokinin biosynthesis in maize. *Plant molecular biology*, 67, 215-229.

- Burman, U., Saini, M., & Kumar, P. (2013). Effect of zinc oxide nanoparticles on growth and antioxidant system of chickpea seedlings. *Toxicological & environmental chemistry*, 95(4), 605-612.
- Cakmak, I. (2000). Tansley Review No. 111 Possible roles of zinc in protecting plant cells from damage by reactive oxygen species. *The New Phytologist*, *146*(2), 185-205.
- Cakmak, I. (2002). Plant nutrition research: Priorities to meet human needs for food in sustainable ways. *Plant and soil*, 247, 3-24.
- Cakmak, I. (2008). Enrichment of cereal grains with zinc: agronomic or genetic biofortification?. *Plant and soil*, 302, 1-17.
- Cakmak, I., & Marschner, H. (1988). Increase in membrane permeability and exudation in roots of zinc deficient plants. *Journal of Plant physiology*, 132(3), 356-361.
- Campos, E. V., do ES Pereira, A., Aleksieienko, I., Do Carmo, G. C., Gohari, G., Santaella, C., ... & Oliveira, H. C. (2023). Encapsulated plant growth regulators and associative microorganisms: Nature-based solutions to mitigate the effects of climate change on plants. *Plant science*, 111688.
- Carducci, B., Keats, E.C., & Bhutta, Z.A. (2021). Zinc supplementation for improving pregnancy and infant outcome. *Cochrane database of systematic reviews*, *3*(3).
- Castro-Camba, R., Sánchez, C., Vidal, N., & Vielba, J. M. (2022). Plant development and crop yield: The role of gibberellins. *Plants*, *11*(19), 2650.
- Chandel, G., Banerjee, S., See, S., Meena, R., Sharma, D. J., & Verulkar, S. B. (2010). Effects of different nitrogen fertilizer levels and native soil properties on rice grain Fe, Zn and protein contents. *Rice science*, 17(3), 213-227.
- Chater, J. M., & Garner, L. C. (2019). Foliar nutrient applications to 'Wonderful' pomegranate (*Punica granatum* L.). I. Effects on fruit mineral nutrient concentrations and internal quality. *Scientia horticulturae*, 244, 421-427.
- Colebrook, E. H., Thomas, S. G., Phillips, A. L., & Hedden, P. (2014). The role of gibberellin signalling in plant responses to abiotic stress. *Journal of experimental biology*, 217(1), 67-75.

- Cruz, K. J. C., de Oliveira, A. R. S., Morais, J. B. S., Severo, J. S., Mendes, P. M. V., de Sousa Melo, S. R., ... & Marreiro, D. D. N. (2018). Zinc and insulin resistance: biochemical and molecular aspects. *Biological trace element research*, 186, 407-412.
- Davarpanah, S., Tehranifar, A., Davarynejad, G., Abadía, J., & Khorasani, R. (2016). Effects of foliar applications of zinc and boron nano-fertilizers on pomegranate (*Punica granatum* cv. Ardestani) fruit yield and quality. *Scientia horticulturae*, 210, 57-64.
- Dhaliwal, S. S., Naresh, R. K., Walia, M. K., Gupta, R. K., Mandal, A., & Singh, R. (2019). Long-term effects of intensive rice–wheat and agroforestry-based cropping systems on build-up of nutrients and budgets in alluvial soils of Punjab, India. Archives of agronomy and soil science, 66(3), 330-342.
- Dhaliwal, S. S., Sharma, V., Shukla, A. K., Gupta, R. K., Verma, V., Kaur, M., ... & Singh, P. (2023). Residual effect of organic and inorganic fertilizers on growth, yield and nutrient uptake in wheat under a basmati rice–wheat cropping system in North-Western India. *Agriculture*, 13(3), 556.
- Dimkpa, C. O., Campos, M. G., Fugice, J., Glass, K., Ozcan, A., Huang, Z., ... & Santra, S. (2022). Synthesis and characterization of novel dual-capped Zn– urea nanofertilizers and application in nutrient delivery in wheat. *Environmental science: Advances*, 1(1), 47-58.
- Dimkpa, C. O., Singh, U., Bindraban, P. S., Elmer, W. H., Gardea-Torresdey, J. L., & White, J. C. (2019). Zinc oxide nanoparticles alleviate drought-induced alterations in sorghum performance, nutrient acquisition, and grain fortification. *Science of the total environment*, 688, 926-934.
- Dobrá, J., Černý, M., Štorchová, H., Dobrev, P., Skalák, J., Jedelský, P. L., ... & Vanková, R. (2015). The impact of heat stress targeting on the hormonal and transcriptomic response in Arabidopsis. *Plant science*, 231, 52-61.
- Drankhan, K., Carter, J., Madl, R., Klopfenstein, C., Padula, F., Lu, Y., ... & Takemoto, D. J. (2003). Antitumor activity of wheats with high orthophenolic content. *Nutrition and cancer*, *47*(2), 188-194.
- Dunn, M. F. (2005). Zinc–ligand interactions modulate assembly and stability of the insulin hexamer–a review. *Biometals*, *18*, 295-303.

- El-Mahdy, M. T., & Elazab, D. S. (2020). Impact of zinc oxide nanoparticles on pomegranate growth under in vitro conditions. *Russian journal of plant physiology*, 67, 162-167.
- Evenson, R. E., & Gollin, D. (2003). Assessing the impact of the Green Revolution, 1960 to 2000. *Science*, *300*(5620), 758-762.
- Faizan, M., & Hayat, S. (2019). Effect of foliar spray of ZnO-NPs on the physiological parameters and antioxidant systems of *Lycopersicon* esculentum. Polish journal of natural sciences, 34, 87-105.
- Faizan, M., Faraz, A., Yusuf, M., Khan, S. T., & Hayat, S. (2018). Zinc oxide nanoparticle-mediated changes in photosynthetic efficiency and antioxidant system of tomato plants. *Photosynthetica*, 56, 678-686.
- Falchi, R., Bonghi, C., Drincovich, M. F., Famiani, F., Lara, M. V., Walker, R. P., & Vizzotto, G. (2020). Sugar metabolism in stone fruit: Source-sink relationships and environmental and agronomical effects. *Frontiers in plant science*, 11, 573982.
- Fan, D., Ran, L., Hu, J., Ye, X., Xu, D., Li, J., ... & Luo, K. (2020). miR319a/TCP module and DELLA protein regulate trichome initiation synergistically and improve insect defenses in *Populus tomentosa*. *New phytologist*, 227(3), 867-883.
- Fan, W., Liu, C., Cao, B., Qin, M., Long, D., Xiang, Z., & Zhao, A. (2018). Genomewide identification and characterization of four gene families putatively involved in cadmium uptake, translocation and sequestration in mulberry. *Frontiers in plant science*, 9, 879.

FAOSTAT, (2024). https://www.fao.org/faostat/en/#data/QCL/visualize.

Fernandez-Cao, J. C., Warthon-Medina, M., H. Moran, V., Arija, V., Doepking, C., Serra-Majem, L., & Lowe, N. M. (2019). Zinc intake and status and risk of type 2 diabetes mellitus: a systematic review and metaanalysis. *Nutrients*, 11(5), 1027.

- Fraley, R. T. (2002). Improving the nutritional quality of plants. In Plant Biotechnology 2002 and Beyond: Proceedings of the 10th IAPTC&B Congress June 23–28, 2002 Orlando, Florida, USA (60-67).
- Francavilla, A., & Joye, I. J. (2020). Anthocyanins in whole grain cereals and their potential effect on health. *Nutrients*, *12*(10), 2922.
- Fukao, T., Xu, K., Ronald, P. C., & Bailey-Serres, J. (2006). A variable cluster of ethylene response factor–like genes regulates metabolic and developmental acclimation responses to submergence in rice. *The plant cell*, 18(8), 2021-2034.
- Gainza-Cortes, F., Pérez-Diaz, R., Perez-Castro, R., Tapia, J., Casaretto, J. A., Gonzalez, S., ... & Gonzalez, E. (2012). Characterization of a putative grapevine Zn transporter, VvZIP3, suggests its involvement in early reproductive development in *Vitis vinifera L. BMC plant biology*, 12, 1-13.
- Gao, H., Dai, W., Zhao, L., Min, J., & Wang, F. (2018). The role of zinc and zinc homeostasis in macrophage function. *Journal of immunology research*, 2018(1), 6872621.
- Gao, S., Xiao, Y., Xu, F., Gao, X., Cao, S., Zhang, F., ... & Chu, C. (2019). Cytokinin-dependent regulatory module underlies the maintenance of zinc nutrition in rice. *New phytologist*, 224(1), 202-215.
- Gao, Y., Tian, X., Wang, W., Xu, X., Su, Y., Yang, J., ... & Ma, J. (2023). Changes in concentrations and transcripts of plant hormones in wheat seedling roots in response to Fusarium crown rot. *The crop journal*, 11(5), 1441-1450.
- Garcia-Lopez, J. I., Nino-Medina, G., Olivares-Saenz, E., Lira-Saldivar, R. H., Barriga-Castro, E. D., Vazquez-Alvarado, R., ... & Zavala-Garcia, F. (2019).
  Foliar application of zinc oxide nanoparticles and zinc sulfate boosts the content of bioactive compounds in habanero peppers. *Plants*, 8(8), 254.
- Geremew, A., Carson, L., Woldesenbet, S., Carpenter, C., Peace, E., & Weerasooriya,
  A. (2021). Interactive effects of organic fertilizers and drought stress on growth and nutrient content of *Brassica juncea* at vegetative stage. *Sustainability*, *13*(24), 13948.

- Ghasal, P. C., Shivay, Y. S., Pooniya, V., Choudhary, M., & Verma, R. K. (2017b). Zinc accounting for different varieties of wheat (*Triticum aestivum*) under different source and methods of application. *Indian journal of agricultural sciences*, 87(9), 1111-6.
- Ghasal, P. C., Shivay, Y. S., Pooniya, V., Choudhary, M., & Verma, R. K. (2017a). Response of wheat genotypes to zinc fertilization for improving productivity and quality. *Archives of agronomy and soil science*, 63(11), 1597-1612.
- Giraldo, P., Ruiz, M., Ibba, M. I., Morris, C. F., Labuschagne, M. T., & Igrejas, G. (2020). Durum wheat storage protein composition and the role of LMW-GS in quality. Wheat quality for improving processing and human health, 73-108.
- Gogia, S., & Sachdev, H. S. (2012). Zinc supplementation for mental and motor development in children. *Cochrane database of systematic reviews*, 12.
- Grote, U., Fasse, A., Nguyen, T. T., & Erenstein, O. (2021). Food security and the dynamics of wheat and maize value chains in Africa and Asia. *Frontiers in Sustainable Food Systems*, 4, 617009.
- Grotz, N., & Guerinot, M. L. (2006). Molecular aspects of Cu, Fe and Zn homeostasis in plants. Biochimica et Biophysica Acta (BBA)-Molecular Cell Research, 1763(7), 595-608.
- Guan, P., Li, X., Zhuang, L., Wu, B., Huang, J., Zhao, J., ... & Zheng, X. (2022). Genetic dissection of lutein content in common wheat via association and linkage mapping. *Theoretical and Applied Genetics*, 135(9), 3127-3141.
- Guo, H., Wu, H., Sajid, A., & Li, Z. (2022). Whole grain cereals: The potential roles of functional components in human health. *Critical Reviews in Food Science* and Nutrition, 62(30), 8388-8402.
- Guo, Y., Wang, J., Liu, W., Liu, J., Wang, C., Wu, Q., & Wang, Z. (2024). Construction of magnetic hydroxyl group-enriched hyper cross-linked polymers with functional triazine as the core for efficient enrichment of plant growth regulators. *Food chemistry*, 433, 137309.

- Gupta, N., Ram, H., & Kumar, B. (2016). Mechanism of Zinc absorption in plants: uptake, transport, translocation and accumulation. *Reviews in environmental* science and biotechnology, 15, 89-109.
- Gupta, S., & Pandey, S. (2020). Enhanced salinity tolerance in the common bean (*Phaseolus vulgaris*) plants using twin ACC deaminase producing rhizobacterial inoculation. *Rhizosphere*, 16, 100241.
- Guzman, C., Ibba, M. I., Álvarez, J. B., Sissons, M., & Morris, C. (2022). Wheat quality. In Wheat improvement: Food security in a changing climate (pp. 177-193). Cham: Springer International Publishing.
- Hacisalihoglu, G., & Kochian, L. V. (2003). How do some plants tolerate low levels of soil zinc? Mechanisms of zinc efficiency in crop plants. *New phytologist*, 159(2), 341-350.
- Hamouda, H. A., Khalifa, R. K. M., El-Dahshouri, M. F., & Zahran, N. G. (2016). Yield, fruit quality and nutrients content of pomegranate leaves and fruit as influenced by iron, manganese and zinc foliar spray. *International journal of pharm tech research*, 9(3), 46-57.
- Hare, R. (2017). Durum wheat: Grain-quality characteristics and management of quality requirements. *Cereal grains*, 135-151. Woodhead Publishing.
- Harland, B., & Oberleas, D. (1977). Reprinted from Cereal Chemistry 54(4), 827-832.
  A modified method for phytate analysis using an ion-exchange procedure: application to textured vegetable proteins. *Selected technical publications*, (26), 400.
- Hasan, M. N., Khaliq, Q. A., Mia, M. D. A. B., Bari, M. N., & Islam, M. R. (2016).
  Chlorophyll Meter-Based Dynamic Nitrogen Management in Wheat (*Triticum aestivum* L.) Under Subtropical Environment. *Current agriculture research journal*, 4(1).
- Hasani, M., Zamani, Z., Savaghebi, G., & Fatahi, R. (2012). Effects of zinc and manganese as foliar spray on pomegranate yield, fruit quality and leaf minerals. *Journal of soil science and plant nutrition*, 12(3), 471-480.

- Hassan, M.U., Aamer, M., Chattha, M.U., Haiying, T., Shahzad, B., Barbanti, L., ... & Guoqin, H. (2020). The critical role of zinc in plants facing the drought stress. *Agriculture*, 10(9), 396.
- Hassan, Z. A., & AL-Shaheen, M. R. (2021, November). Vital Response of the Wheat to Gibberellic Acid "GA3" and Prolin Under Water Defect Conditions. In *IOP conference series: earth and environmental science* (Vol. 904, No. 1, p. 012072). IOP Publishing.
- Hattori, Y., Nagai, K., Furukawa, S., Song, X. J., Kawano, R., Sakakibara, H., ... & Ashikari, M. (2009). The ethylene response factors SNORKEL1 and SNORKEL2 allow rice to adapt to deep water. *Nature*, 460(7258), 1026-1030.
- Helmy, K. G., Partila, A. M., & Salah, M. (2020). Gamma radiation and polyvinyl pyrrolidone mediated synthesis of zinc oxide/zinc sulfide nanoparticles and evaluation of their antifungal effect on pre and post harvested orange and pomegranate fruits. *Biocatalysis and agricultural biotechnology*, 29, 101728.
- Hernandez, M. C., Rojas, P., Carrasco, F., Basfi-Fer, K., Valenzuela, R., Codoceo, J.,
  ... & Ruz, M. (2020). Fatty acid desaturation in red blood cell membranes of patients with type 2 diabetes is improved by zinc supplementation. *Journal of trace elements in medicine and biology*, 62, 126571.
- Hirose, N., Makita, N., Kojima, M., Kamada-Nobusada, T., & Sakakibara, H. (2007). Overexpression of a type-A response regulator alters rice morphology and cytokinin metabolism. *Plant and cell physiology*, 48(3), 523-539.
- Hong, J., Wang, C., Wagner, D. C., Gardea-Torresdey, J. L., He, F., & Rico, C. M. (2021). Foliar application of nanoparticles: mechanisms of absorption, transfer, and multiple impacts. *Environmental science: nano*, 8(5), 1196-1210.
- Honig, M., Plihalova, L., Husickova, A., Nisler, J., & Dolezal, K. (2018). Role of cytokinins in senescence, antioxidant defence and photosynthesis. *International journal of molecular sciences*, 19(12), 4045.

- Hotz, C., & Brown, K. H. (2004). Assessment of the risk of zinc deficiency in populations and options for its control. *Journal of food nutrition bulletin*, 2, 94–204.
- Huang, Q., Qiu, W., Yu, M., Li, S., Lu, Z., Zhu, Y., ... & Zhuo, R. (2022). Genomewide characterization of Sedum plumbizincicola HMA gene family provides functional implications in cadmium response. *Plants*, 11(2), 215.
- Hussain, D., Haydon, M. J., Wang, Y., Wong, E., Sherson, S. M., Young, J., ... & Cobbett, C. S. (2004). P-type ATPase heavy metal transporters with roles in essential zinc homeostasis in Arabidopsis. *The plant cell*, 16(5), 1327-1339.
- Hussain, F., Hadi, F., & Rongliang, Q. (2021). Effects of zinc oxide nanoparticles on antioxidants, chlorophyll contents, and proline in *Persicaria hydropiper* L. and its potential for Pb phytoremediation. *Environmental science and pollution research*, 28(26), 34697-34713.
- Illouz-Eliaz, N., Ramon, U., Shohat, H., Blum, S., Livne, S., Mendelson, D., & Weiss, D. (2019). Multiple gibberellin receptors contribute to phenotypic stability under changing environments. *The plant cell*, 31(7), 1506-1519.
- Ilyas, M., Khan, I., Chattha, M. U., Hassan, M. U., Zain, M., Farhad, W., ... & Adeel, M. (2020). Evaluating the effect of zinc application methods on growth and yield of wheat cultivars. *Journal of innovative sciences*, 6(2), 150-156.
- Impa, S. M., & Johnson-Beebout, S. E. (2012). Mitigating zinc deficiency and achieving high grain Zn in rice through integration of soil chemistry and plant physiology research. *Plant and soil*, 361, 3-41.
- Impa, S. M., Gramlich, A., Tandy, S., Schulin, R., Frossard, E., & Johnson-Beebout, S. E. (2013b). Internal Zn allocation influences Zn deficiency tolerance and grain Zn loading in rice (*Oryza sativa* L.). *Frontiers in plant science*, 4, 534.
- Impa, S. M., Morete, M. J., Ismail, A. M., Schulin, R., & Johnson-Beebout, S. E. (2013a). Zn uptake, translocation and grain Zn loading in rice (*Oryza sativa* L.) genotypes selected for Zn deficiency tolerance and high grain Zn. *Journal of experimental botany*, 64(10), 2739-2751.

- Imran, M., & Rehim, A. (2017). Zinc fertilization approaches for agronomic biofortification and estimated human bioavailability of zinc in maize grain. Archives of agronomy and soil science, 63(1), 106-116.
- Ioio, R. D., Linhares, F. S., Scacchi, E., Casamitjana-Martinez, E., Heidstra, R., Costantino, P., & Sabatini, S. (2007). Cytokinins determine Arabidopsis root-meristem size by controlling cell differentiation. *Current biology*, 17(8), 678-682.
- Ioio, R. D., Nakamura, K., Moubayidin, L., Perilli, S., Taniguchi, M., Morita, M. T., ... & Sabatini, S. (2008). A genetic framework for the control of cell division and differentiation in the root meristem. *Science*, 322(5906), 1380-1384.
- Iqbal, M. J., Shams, N., & Fatima, K. (2022). Nutritional quality of wheat. In *Wheat-Recent Advances*. IntechOpen.
- Iqbal, S., Qais, F. A., Alam, M. M., & Naseem, I. (2018). Effect of glycation on human serum albumin–zinc interaction: a biophysical study. *JBIC Journal of biological inorganic chemistry*, 23, 447-458.
- Isaac, R. A., & Kerber, J. D. (1971). Atomic absorption and flame photometry: Techniques and uses in soil, plant, and water analysis. *Instrumental methods* for analysis of soils and plant tissue, 17-37.
- Ishimaru, Y., Suzuki, M., Kobayashi, T., Takahashi, M., Nakanishi, H., Mori, S., & Nishizawa, N. K. (2005). OsZIP4, a novel zinc-regulated zinc transporter in rice. *Journal of Experimental Botany*, 56(422), 3207-3214.
- Jackson, M. L. (2005). Soil chemical analysis: advanced course: a manual of methods useful for instruction and research in soil chemistry, physical chemistry of soils, soil fertility, and soil genesis. UW-Madison Libraries parallel press.
- Jalal, A., Galindo, F. S., Freitas, L. A., da Silva Oliveira, C. E., de Lima, B. H., Pereira, Í. T., ... & Teixeira Filho, M. C. M. (2022). Yield, zinc efficiencies and biofortification of wheat with zinc sulfate application in soil and foliar nano zinc fertilisation. *Crop and pasture science*, 73,749-759.

- Jalal, A., Júnior, E. F., & Teixeira Filho, M. C. M. (2024). Interaction of Zinc Mineral Nutrition and Plant Growth-Promoting Bacteria in Tropical Agricultural Systems: A Review. *Plants*, 13(5), 571.
- Jalal, A., Oliveira, C. E. D. S., Bastos, A. D. C., Fernandes, G. C., De Lima, B. H., Furlani Junior, E., ... & Teixeira Filho, M. C. M. (2023). Nano zinc and plant growth-promoting bacteria improve biochemical and metabolic attributes of maize in tropical Cerrado. *Frontiers in plant science*, 13, 1046642.
- Jiang, W., Struik, P. C., Van Keulen, H., Zhao, M., Jin, L. N., & Stomph, T. J. (2008). Does increased zinc uptake enhance grain zinc mass concentration in rice?. Annals of applied biology, 153(1), 135-147.
- Joye, I. J. (2020). Dietary fibre from whole grains and their benefits on metabolic health. *Nutrients*, *12*(10), 3045.
- Kambe, T., Tsuji, T., Hashimoto, A., & Itsumura, N. (2015). The physiological, biochemical, and molecular roles of zinc transporters in zinc homeostasis and metabolism. *Physiological reviews*, 95(3), 749-84.
- Kamboj, S., & Mathpal, B. (2019). Improving rice grain quality by foliar application of plant growth regulators under various mode of Zn application. *Plant archives*, 19(2), 2181-2184.
- Karunadasa, S. S., Kurepa, J., Shull, T. E., & Smalle, J. A. (2020). Cytokinin-induced protein synthesis suppresses growth and osmotic stress tolerance. *New phytologist*, 227(1), 50-64.
- Kaur, H., Srivastava, S., Goyal, N., & Walia, S. (2024). Behavior of zinc in soils and recent advances on strategies for ameliorating zinc phytotoxicity. *Environmental and experimental botany*, 105676.
- Kaur, K., Mavi, G. S., Bhagat, I., Sharma, A., Srivastava, P., Kaur, H., & Sohu, V. S. (2023). Phenotypic evaluation of grain zinc enhanced wheat lines for agronomic and quality traits. *Indian journal of agricultural research*, 57(1), 30-34.
- Kaur, R., Jaidka, M., & Kingra, P. K. (2017). Correlation analysis of growth, yield and yield components of wheat (*Triticum aestivum*) under varying weed densities. *Indian journal of agricultural sciences*, 87(6), 746-53.

- Kawade, R. (2012). Zinc status and its association with the health of adolescents: a review of studies in India. *Global health action*, 5(1), 7353.
- Keram, K. S., Sharma, B. L., & Sawarkar, S. D. (2012). Impact of Zn application on yield, quality, nutrients uptake and soil fertility in a medium deep black soil (vertisol). *International journal of science, environment and technology*, 1(5), 563-571.
- Keshishian, E. A., Hallmark, H. T., Ramaraj, T., Plačková, L., Sundararajan, A., Schilkey, F., ... & Rashotte, A. M. (2018). Salt and oxidative stresses uniquely regulate tomato cytokinin levels and transcriptomic response. *Plant direct*, 2(7), e00071.
- Khalil, I. A., & Jan, A. (2002). Textbook of cropping technology. *National book found, Pakistan,* 204-224.
- Khan, N. M., Ali, A., Wan, Y., & Zhou, G. (2024). Genome-wide identification of heavy-metal ATPases genes in Areca catechu: investigating their functionality under heavy metal exposure. *BMC Plant Biology*, 24(1), 484.
- Khandal, H., Gupta, S. K., Dwivedi, V., Mandal, D., Sharma, N. K., Vishwakarma, N. K., ... & Chattopadhyay, D. (2020). Root-specific expression of chickpea cytokinin oxidase/dehydrogenase 6 leads to enhanced root growth, drought tolerance and yield without compromising nodulation. *Plant biotechnology journal*, 18(11), 2225-2240.
- Khatun, M. A., Hossain, M. M., Bari, M. A., Abdullahil, K. M., Parvez, M. S., Alam, M. F., & Kabir, A. H. (2018). Zinc deficiency tolerance in maize is associated with the up-regulation of Zn transporter genes and antioxidant activities. *Plant biology*, 20(4), 765-770.
- Kholssi, R., Marks, E. A., Miñón, J., Maté, A. P., Sacristán, G., Montero, O., ... & Rad, C. (2021). A consortium of cyanobacteria and plant growth promoting rhizobacteria for wheat growth improvement in a hydroponic system. *South African journal of botany*, 142, 247-258.
- Khorsandi, F., Yazdi, F. A., & Vazifehshenas, M. R. (2009). Foliar zinc fertilization improves marketable fruit yield and quality attributes of pomegranate. *International journal of agriculture and biology*, 11(6), 766-770.

- King, J. C. (2011). Zinc: an essential but elusive nutrient. *The American journal of clinical nutrition*, 94(2), 679S-684S.
- Kislev, M. E., Nadel, D., & Carmi, I. (1992). Epipalaeolithic (19,000 BP) cereal and fruit diet at Ohalo II, Sea of Galilee, Israel. *Review of palaeobotany and palynology*, 73(1-4), 161-166.
- Knijnenburg, J. T., Laohhasurayotin, K., Khemthong, P., & Kangwansupamonkon,W. (2019). Structure, dissolution, and plant uptake of ferrous/zinc phosphates. *Chemosphere*, 223, 310-318.
- Koprna, R., Humplík, J. F., Špíšek, Z., Bryksová, M., Zatloukal, M., Mik, V., ... & Doležal, K. (2020). Improvement of tillering and grain yield by application of cytokinin derivatives in wheat and barley. *Agronomy*, 11(1), 67.
- Krishnan, S., & Dayanandan, P. (2003). Structural and histochemical studies on grainfilling in the caryopsis of rice (*Oryza sativa* L.). *Journal of biosciences*, 28, 455-469.
- Kumar, A., Lal, M. K., Kar, S. S., Nayak, L., Ngangkham, U., Samantaray, S., & Sharma, S. G. (2017). Bioavailability of iron and zinc as affected by phytic acid content in rice grain. *Journal of food biochemistry*, 41(6), e12413.
- Kumar, D., Dhar, S., Kumar, S., Meena, D. C., & Meena, R. B. (2019). Effect of zinc application on yield attributes and yield of maize and wheat in maize-wheat cropping system. *International journal of current microbiology and applied sciences*, 8(1), 1931-1941.
- Kumar, P., Yadava, R. K., Gollen, B., Kumar, S., Verma, R. K., & Yadav, S. (2011). Nutritional contents and medicinal properties of wheat: a review. *Life Sciences and Medicine Research*, 22(1), 1-10.
- Kumssa, D. B., Joy, E. J., Ander, E. L., Watts, M. J., Young, S. D., Walker, S., & Broadley, M. R. (2015). Dietary calcium and zinc deficiency risks are decreasing but remain prevalent. *Scientific reports*, 5(1), 10974.
- Kuroha, T., Nagai, K., Gamuyao, R., Wang, D. R., Furuta, T., Nakamori, M., ... & Ashikari, M. (2018). Ethylene-gibberellin signaling underlies adaptation of rice to periodic flooding. *Science*, 361(6398), 181-186.

- Lassi, Z. S., Kurji, J., de Oliveira, C. S., Moin, A., & Bhutta, Z. A. (2020). Zinc supplementation for the promotion of growth and prevention of infections in infants less than six months of age. *Cochrane database of systematic reviews*, 4.
- Lassi, Z. S., Moin, A., & Bhutta, Z. A. (2016). Zinc supplementation for the prevention of pneumonia in children aged 2 months to 59 months. *Cochrane database of systematic reviews*, 12.
- Leff, B., Ramankutty, N., & Foley, J. A. (2004). Geographic distribution of major crops across the world. *Global biogeochemical cycles*, 18, 1-27.
- Leite, C. M. D. C., Muraoka, T., Colzato, M., & Alleoni, L. R. F. (2019). Soil-applied Zn effect on soil fractions. *Scientia agricola*, 77, e20180124.
- Li, S., Liu, X., Zhou, X., Li, Y., Yang, W., & Chen, R. (2019). Improving zinc and iron accumulation in maize grains using the zinc and iron transporter ZmZIP5. *Plant and Cell Physiology*, 60(9), 2077-2085.
- Lindsay, W. L., & Norvell, W. (1978). Development of a DTPA soil test for zinc, iron, manganese, and copper. *Soil science society of America journal*, 42(3), 421-428.
- Ling, X. B., Wei, H. W., Wang, J., Kong, Y. Q., Wu, Y. Y., Guo, J. L., ... & Li, J. K. (2016). Mammalian metallothionein-2A and oxidative stress. *International journal of molecular sciences*, 17(9), 1483.
- Liu, D. Y., Zhang, W., Pang, L. L., Zhang, Y. Q., Wang, X. Z., Liu, Y. M., ... & Zou, C. Q. (2017). Effects of zinc application rate and zinc distribution relative to root distribution on grain yield and grain Zn concentration in wheat. *Plant and soil*, 411, 167-178.
- Liu, H. E., Wang, Q. Y., Rengel, Z., & Zhao, P. (2015). Zinc fertilization alters flour protein composition of winter wheat genotypes varying in gluten content. *Plant soil and environment 61*:195-200.
- Livingstone, C. (2015). Zinc: physiology, deficiency, and parenteral nutrition. *Nutrition in clinical practice*, *30*(3), 371-382.

- Long, D. Y. (2019). Molecular Cytogentic Identification of BC1F8 Generation of Common Wheat–Aegilops Geniculata Roth SY159 Progeny. Xianyang: northwest A&F university.
- Lowe, N. M., Hall, A. G., Broadley, M. R., Foley, J., Boy, E., & Bhutta, Z. A. (2024). Preventing and controlling zinc deficiency across the life course: A call to action. *Advances in nutrition*, 100181.
- Lowe, N. M., Woodhouse, L. R., & King, J. C. (1998). A comparison of the shortterm kinetics of zinc metabolism in women during fasting and following a breakfast meal. *British journal of nutrition*, 80(4), 363-370.
- Lu, J., Stewart, A. J., Sleep, D., Sadler, P. J., Pinheiro, T. J., & Blindauer, C. A. (2012). A molecular mechanism for modulating plasma Zn speciation by fatty acids. *Journal of the American chemical society*, 134(3), 1454-1457.
- Luo, A., Qian, Q., Yin, H., Liu, X., Yin, C., Lan, Y., ... & Chu, C. (2006). EUI1, encoding a putative cytochrome P450 monooxygenase, regulates internode elongation by modulating gibberellin responses in rice. *Plant and cell physiology*, 47(2), 181-191.
- Madhu, U., Begum, M., Salam, A., & Sarkar, S. K. (2018). Influence of sowing date on the growth and yield performance of wheat (*Triticum aestivum* L.) varieties. *Archives of agriculture and environmental science*, *3*(1), 89-94.
- Maity, A., Babu, K. D., & Sarkar, A. (2019). Guidelines for fertilizer use in pomegranate orchards based on seasonal uptake and partitioning of nutrients. *Scientia horticulturae*, 252, 138-148.
- Malecki, J. J., Kadzikiewicz-Schoeneich, M., & Szostakiewicz-Hołownia, M. (2016). Concentration and mobility of copper and zinc in the hypergenic zone of a highly urbanized area. *Environmental earth sciences*, 75, 1-13.
- Maqbool, M. A., & Beshir, A. (2019). Zinc biofortification of maize (*Zea mays* L.): Status and challenges. *Plant breeding*, *138*(1), 1-28.
- Martinez-Esteso, M. J., Nørgaard, J., Brohée, M., Haraszi, R., Maquet, A., & O'Connor, G. (2016). Defining the wheat gluten peptide fingerprint via a discovery and targeted proteomics approach. *Journal of proteomics*, 147, 156-168.

- Mathpal, B., Srivastava, P. C., Pachauri, S. P., Shukla, A. K., & Shankhdhar, S. C. (2023). Role of Gibberellic Acid and Cytokinin in Improving Grain Zn Accumulation and Yields of Rice (*Oryza sativa* L.). *Journal of soil science and plant nutrition*, 23(4), 6006-6016.
- Mathpal, B., Srivastava, P. C., Pachauri, S. P., Shukla, A. K., Pant, N. C., & Shankhdhar, S. C. (2022). Enhancing translocation and remobilization of zinc in wheat by the application of plant growth regulators. *Israel journal of plant sciences*, 69(1-2), 61-68.
- Mathpal, B., Srivastava, P. C., Shankhdhar, D., & Shankhdhar, S. C. (2015). Zinc enrichment in wheat genotypes under various methods of zinc application. *Plant, soil and environment*, *61*(4), 171-175.
- Meena, D., Tiwari, R., & Singh, O. P. (2014). Effect of nutrient spray on growth, fruit yield and quality of aonla. *Annals of plant and soil research*, *16*(3), 242-245.
- Mi, X., Wang, X., Wu, H., Gan, L., Ding, J., & Li, Y. (2017). Characterization and expression analysis of cytokinin biosynthesis genes in *Fragaria vesca*. *Plant* growth regulation, 82, 139-149.
- Mirzapour, M. H., & Khoshgoftar, A. H. (2006). Zinc application effects on yield and seed oil content of sunflower grown on a saline calcareous soil. *Journal of plant nutrition*, 29(10), 1719-1727.
- Mogazy, A. M., & Hanafy, R. S. (2022). Foliar spray of biosynthesized zinc oxide nanoparticles alleviate salinity stress effect on *Vicia faba* plants. *Journal of soil science and plant nutrition*, 22(2), 2647-2662.
- Moradinezhad, F., & Ranjbar, A. (2024). Foliar application of fertilizers and plant growth regulators on pomegranate fruit yield and quality: a review. *Journal of plant nutrition*, 47(5), 797-821.
- Mu, S., Yamaji, N., Sasaki, A., Luo, L., Du, B., Che, J., ... & Ma, J. F. (2021). A transporter for delivering zinc to the developing tiller bud and panicle in rice. *The Plant Journal*, 105(3), 786-799.
- Munir, T., Rizwan, M., Kashif, M., Shahzad, A., Ali, S., Amin, N., ... & Imran, M. (2018). Effect of zinc oxide nanoparticles on the growth and Zn uptake in wheat (*Triticum aestivum* L.) By seed priming method. *Digest journal of nanomaterials & biostructures*, 13(1).

- Nagar, S., Ramakrishnan, S., Singh, V. P., Singh, G. P., Dhakar, R., Umesh, D. K., & Arora, A. (2015). Cytokinin enhanced biomass and yield in wheat by improving N-metabolism under water limited environment. *Indian journal of plant physiology*, 20, 31-38.
- Nakamura, T., Nishiyama, S., Futagoishi-Suginohara, Y., Matsuda, I., & Higashi, A. (1993). Mild to moderate zinc deficiency in short children: effect of zinc supplementation on linear growth velocity. *The journal of pediatrics*, 123(1), 65-69.
- Nakandalage, N., Nicolas, M., Norton, R. M., Hirotsu, N., Milham, P. J., & Seneweera, S. (2016). Improving rice zinc biofortification success rates through genetic and crop management approaches in a changing environment. *Frontiers in plant science*, 7, 764.
- Neumann, K., Verburg, P. H., Stehfest, E., & Müller, C. (2010). The yield gap of global grain production: A spatial analysis. *Agricultural systems*, 103(5), 316-326.
- Niharika, Singh, N. B., Singh, A., Khare, S., Yadav, V., Bano, C., & Yadav, R. K. (2021). Mitigating strategies of gibberellins in various environmental cues and their crosstalk with other hormonal pathways in plants: a review. *Plant molecular biology reporter*, 39, 34-49.
- Noulas, C., Tziouvalekas, M., & Karyotis, T. (2018). Zinc in soils, water and food crops. *Journal of trace elements in medicine and biology*, *49*, 252-260.
- Obaid, E. A., & Al-Hadethi, M. E. A. (2013). Effect of foliar application with manganese and zinc on pomegranate growth, yield and fruit quality. *Journal of horticultural science & ornamental plants*, 5(1), 41-45.
- Olechnowicz, J., Tinkov, A., Skalny, A., & Suliburska, J. (2018). Zinc status is associated with inflammation, oxidative stress, lipid, and glucose metabolism. *The journal of physiological sciences*, 68(1), 19-31.
- Olsen, L. I., & Palmgren, M. G. (2014). Many rivers to cross: the journey of zinc from soil to seed. *Frontiers in plant science*, *5*, 73677.
- Olsen, S. R. (1954). *Estimation of available phosphorus in soils by extraction with sodium bicarbonate* (No. 939). US department of agriculture.

- Palmer, C. M., & Guerinot, M. L. (2009). Facing the challenges of Cu, Fe and Zn homeostasis in plants. *Nature chemical biology*, 5(5), 333-340.
- Panigrahi, S., Pankaj, Y. K., Kumar, V., Kumar, R., & Singh, S. K. (2022). Studies on effects of terminal heat stress on yield stability, grain iron and zinc contents in wheat (*Triticum aestivum* L.). *Indian journal of genetics and plant breeding*, 82(03), 289-298.
- Pavhane, S. B., Waghmare, G. M., Gawade, R. N., Shinde, V. N., Khandare, V. S., Ismail, S., & More, S. S. (2022). Effect of fertilizer and chelated micronutrient on fruit set, retention, cracking and fruit drop of pomegranate (*Punica granatum* L.) Cv. Bhagwa. *The pharma innovation journal*, 11(1), 124-128.
- Peck, A. W., McDonald, G. K., & Graham, R. D. (2008). Zinc nutrition influences the protein composition of flour in bread wheat (Triticum aestivum L.). *Journal* of Cereal Science, 47(2), 266-274.
- Pedas, P., Schjoerring, J. K., & Husted, S. (2009). Identification and characterization of zinc-starvation-induced ZIP transporters from barley roots. *Plant Physiology and Biochemistry*, 47(5), 377-383.
- Persson, D. P., De Bang, T. C., Pedas, P. R., Kutman, U. B., Cakmak, I., Andersen, B., ... & Husted, S. (2016). Molecular speciation and tissue compartmentation of zinc in durum wheat grains with contrasting nutritional status. *New phytologist*, 211(4), 1255-1265.
- Piper, C. S. (2019). Soil and plant analysis. Scientific publishers.
- Pompano, L. M., & Boy, E. (2021). Effects of dose and duration of zinc interventions on risk factors for type 2 diabetes and cardiovascular disease: a systematic review and meta-analysis. *Advances in nutrition*, 12(1), 141-160.
- Prasad, A. S. (2009). Zinc: role in immunity, oxidative stress and chronic inflammation. *Current opinion in clinical nutrition & metabolic care*, 12(6), 646-652.
- Prasad, R. (2022). Cytokinin and its key role to enrich the plant nutrients and growth under adverse conditions-an update. *Frontiers in genetics*, *13*, 883924.

- Prentice, A. M., Gershwin, M. E., Schaible, U. E., Keusch, G. T., Victora, C. G., & Gordon, J. I. (2008). New challenges in studying nutrition-disease interactions in the developing world. *The journal of clinical investigation*, 118(4), 1322-1329.
- Prerostova, S., Dobrev, P. I., Gaudinova, A., Knirsch, V., Körber, N., Pieruschka, R., ... & Vankova, R. (2018). Cytokinins: Their impact on molecular and growth responses to drought stress and recovery in Arabidopsis. *Frontiers in plant science*, 9, 655.
- Pullakhandam, R., Agrawal, P. K., Peter, R., Ghosh, S., Reddy, G. B., Kulkarni, B., ... & Johnston, R. (2021). Prevalence of low serum zinc concentrations in Indian children and adolescents: findings from the Comprehensive National Nutrition Survey 2016–18. *The American Journal of Clinical Nutrition*, 114(2), 638-648.
- Rai-Kalal, P., & Jajoo, A. (2021). Priming with zinc oxide nanoparticles improve germination and photosynthetic performance in wheat. *Plant physiology and biochemistry*, 160, 341-351.
- Ram, H., Rashid, A., Zhang, W., Duarte, A. Á., Phattarakul, N., Simunji, S., ... & Cakmak, I. (2016). Biofortification of wheat, rice and common bean by applying foliar zinc fertilizer along with pesticides in seven countries. *Plant* and soil, 403, 389-401.
- Raza, M. A. S., Zaheer, M. S., Saleem, M. F., Khan, I. H., Ahmad, S., & Iqbal, R. (2020). Drought ameliorating effect of exogenous applied cytokinin in wheat. *Pakistan journal of agricultural sciences*, 57(3), 725-733.
- Read, S. A., Obeid, S., Ahlenstiel, C., & Ahlenstiel, G. (2019). The role of zinc in antiviral immunity. Advances in nutrition, 10(4), 696-710.
- Rehman, A., Farooq, M., Ozturk, L., Asif, M., & Siddique, K. H. (2018). Zinc nutrition in wheat-based cropping systems. *Plant and Soil*, 422, 283-315.
- Rehman, A., Farooq, M., Ullah, A., Nadeem, F., Im, S. Y., Park, S. K., & Lee, D. J. (2020). Agronomic biofortification of zinc in Pakistan: Status, benefits, and constraints. *Frontiers in sustainable food systems*, 4, 591722.

- Ren, Y., Li, X., Liang, J., Wang, S., Wang, Z., Chen, H., & Tang, M. (2023). Brassinosteroids and gibberellic acid actively regulate the zinc detoxification mechanism of *Medicago sativa* L. seedlings. *BMC plant biology*, 23(1), 75.
- Rengel, Z., & Graham, R. D. (1995). Wheat genotypes differ in Zn efficiency when grown in chelate-buffered nutrient solution: I. Growth. *Plant and Soil*, 176, 307-316.
- Riley, M. M., Gartrell, J. W., Brennan, R. F., Hamblin, J., & Coates, P. (1992). Zinc deficiency in wheat and lupins in Western Australia is affected by the source of phosphate fertiliser. *Australian Journal of Experimental Agriculture*, 32(4), 455-463.
- Rose, T. J., Impa, S. M., Rose, M. T., Pariasca-Tanaka, J., Mori, A., Heuer, S., ... & Wissuwa, M. (2013). Enhancing phosphorus and zinc acquisition efficiency in rice: a critical review of root traits and their potential utility in rice breeding. *Annals of botany*, 112(2), 331-345.
- Ruz, M., Castillo-Duran, C., Lara, X., Codoceo, J., Rebolledo, A., & Atalah, E. (1997). A 14-mo zinc-supplementation trial in apparently healthy Chilean preschool children. *The American journal of clinical nutrition*, 66(6), 1406-1413.
- Sabagh, A. E., Mbarki, S., Hossain, A., Iqbal, M. A., Islam, M. S., Raza, A., ... & Farooq, M. (2021). Potential role of plant growth regulators in administering crucial processes against abiotic stresses. *Frontiers in agronomy*, *3*, 648694.
- Safa, A., Hakimi, L., Pypker, T. G., & Khosropour, E. (2020). The effect of ZnSO4 and KNO3 on quantitative and qualitative properties of *Punica granatum* L. *Journal of plant nutrition*, 43(9), 1286-1292.
- Saha, S., Chakraborty, M., Sarkar, D., Batabyal, K., Mandal, B., Murmu, S., ... & Bell, R. W. (2017). Rescheduling zinc fertilization and cultivar choice improve zinc sequestration and its bioavailability in wheat grains and flour. *Field crops research*, 200, 10-17.
- Salam, A., Khan, A. R., Liu, L., Yang, S., Azhar, W., Ulhassan, Z., ... & Gan, Y. (2022). Seed priming with zinc oxide nanoparticles downplayed

ultrastructural damage and improved photosynthetic apparatus in maize under cobalt stress. *Journal of hazardous materials*, 423, 127021.

- Sasaki, A., Yamaji, N., & Ma, J. F. (2014). Overexpression of OsHMA3 enhances Cd tolerance and expression of Zn transporter genes in rice. *Journal of experimental botany*, 65(20), 6013-6021.
- Satoh-Nagasawa, N., Mori, M., Nakazawa, N., Kawamoto, T., Nagato, Y., Sakurai, K., ... & Akagi, H. (2012). Mutations in rice (*Oryza sativa*) heavy metal ATPase 2 (OsHMA2) restrict the translocation of zinc and cadmium. *Plant* and Cell Physiology, 53(1), 213-224.
- Sattar, A., Wang, X., Ul-Allah, S., Sher, A., Ijaz, M., Irfan, M., ... & Skalicky, M. (2022). Foliar application of zinc improves morpho-physiological and antioxidant defense mechanisms, and agronomic grain biofortification of wheat (*Triticum aestivum* L.) under water stress. *Saudi journal of biological sciences*, 29(3), 1699-1706.
- Savi, G. D., Bortoluzzi, A. J., & Scussel, V. M. (2013). Antifungal properties of Zinccompounds against toxigenic fungi and mycotoxin. *International journal of food science & technology*, 48(9), 1834-1840.
- Shah, D., Sachdev, H. S., Gera, T., De-Regil, L. M., & Peña-Rosas, J. P. (2016). Fortification of staple foods with zinc for improving zinc status and other health outcomes in the general population. *Cochrane database of systematic reviews*, 6.
- Shang, M., Wang, X., Zhang, J., Qi, X., Ping, A., Hou, L., ... & Li, M. (2017). Genetic regulation of GA metabolism during vernalization, floral bud initiation and development in Pak Choi (*Brassica rapa* ssp. chinensis Makino). *Frontiers in plant science*, 8, 1533.
- Shang, Y., Hasan, M. K., Ahammed, G. J., Li, M., Yin, H., & Zhou, J. (2019). Applications of nanotechnology in plant growth and crop protection: a review. *Molecules*, 24(14), 2558.
- Shekari, F., Mohammadi, H., Pourmohammad, A., Avanes, A., & Benam, M. B. K. (2015). Spring wheat yielding and the content of protein and zinc in its grain

depending on zinc fertilisation. *Electron journal of polish agricultural universities*, 18(1), 1-12.

- Shewry, P. R. (2007). Improving the protein content and composition of cereal grain. *Journal of cereal science*, 46(3), 239-250.
- Shewry, P. R. (2009). The HEALTHGRAIN programme opens new opportunities for improving wheat for nutrition and health. *Nutrition Bulletin*, *34*(2), 225–231.
- Shewry, P. R. (2009). Wheat. Journal of experimental botany, 60(6), 1537-1553.
- Shewry, P. R., Powers, S., Field, J. M., Fido, R. J., Jones, H. D., Arnold, G. M., ... & Darlington, H. (2006). Comparative field performance over 3 years and two sites of transgenic wheat lines expressing HMW subunit transgenes. *Theoretical and Applied Genetics*, 113, 128-136.
- Shukla, A. K., Tiwari, P. K., Pakhare, A., & Prakash, C. (2016). Zinc and iron in soil, plant, animal and human health. *Indian journal of fertilisers*, *12*(11), 133-149.
- Simmonds, D. H. (1989). Wheat and wheat quality in Australia. Csiro Publishing, 31-61.
- Sinclair, S. A., & Krämer, U. (2012). The zinc homeostasis network of land plants. Biochimica et Biophysica Acta (BBA)-Molecular Cell Research, 1823(9), 1553-1567.
- Singh, J., Aulakh, G. S., & Singh, S. (2023). Effect of seed priming on growth and yield of late sown wheat (*Triticum aestivum*) in central plain region of Punjab. *Research on crops*, 24(1), 1-7.
- Singh, S., Riar, A. S., Kaur, P., & Singh, G. (2022). Assessing impact of temperature change on phenology and grain yield of wheat by using Infocrop model. *Agricultural Research Journal*, 59(6), 1151-1158.
- Singh, V., Kaur, S., Singh, J., Kaur, A., & Gupta, R. K. (2021). Rescheduling fertilizer nitrogen topdressing timings for improving productivity and mitigating N<sub>2</sub>O emissions in timely and late sown irrigated wheat (*Triticum aestivum* L.). Archives of agronomy and soil science, 67(5), 647-659.
- Slavin, J. (2004). Whole grains and human health. *Nutrition research reviews*, 17(1), 99-110.
- Slavin, J. L., Jacobs, D., Marquart, L. E. N., & Wiemer, K. (2001). The role of whole grains in disease prevention. *Journal of the American Dietetic Association*, 101(7), 780-785.

- Sofy, M. R., Elhindi, K. M., Farouk, S., & Alotaibi, M. A. (2020). Zinc and paclobutrazol mediated regulation of growth, upregulating antioxidant aptitude and plant productivity of pea plants under salinity. *Plants*, *9*(9), 1197.
- Sperotto, R. A., Ricachenevsky, F. K., Waldow, V. D. A., Müller, A. L. H., Dressler, V. L., & Fett, J. P. (2013). Rice grain Fe, Mn and Zn accumulation: How important are flag leaves and seed number?. *Plant soil and environment*, 59, 262-266.
- Srivastava, R. K., Satyavathi, C. T., Mahendrakar, M. D., Singh, R. B., Kumar, S., Govindaraj, M., & Ghazi, I. A. (2021). Addressing iron and zinc micronutrient malnutrition through nutrigenomics in pearl millet: Advances and prospects. *Frontiers in genetics*, 12, 723472.
- Stewart, C. P., Dewey, K. G., & Ashorn, P. (2010). The undernutrition epidemic: an urgent health priority. *The lancet*, *375*(9711), 282.
- Stomph, T., Jiang, W., & Struik, P. C. (2009). Zinc biofortification of cereals: rice differs from wheat and barley. *Trends in plant science*, 14(3).
- Subbiah, B. V., & Asija, G. L. (1956). A rapid procedure for the estimation of available nitrogen in soils. *Current science*, 25, 259-260.
- Sudha, S., & Stalin, P. (2017). Zinc deficiency in soil and role of zinc in human and plant. *International journal of farm sciences*, 7(4), 30-38.
- Suganya, A., Saravanan, A., & Manivannan, N. (2020). Role of zinc nutrition for increasing zinc availability, uptake, yield, and quality of maize (Zea mays L.) grains: An overview. Communications in soil science and plant analysis, 51(15), 2001-2021.
- Supraja, N., Prasad, T. N. V. K. V., Gandhi, A. D., Anbumani, D., Kavitha, P., & Babujanarthanam, R. (2018). Synthesis, characterization and evaluation of antimicrobial efficacy and brine shrimp lethality assay of *Alstonia scholaris* stem bark extract mediated ZnONPs. *Biochemistry and biophysics reports*, 14, 69-77.
- Suzuki, M., Bashir, K., Inoue, H., Takahashi, M., Nakanishi, H., & Nishizawa, N. K. (2012). Accumulation of starch in Zn-deficient rice. *Rice*, *5*, 1-8.

- Swamy, B. M., Rahman, M. A., Inabangan-Asilo, M. A., Amparado, A., Manito, C., Chadha-Mohanty, P., ... & Slamet-Loedin, I. H. (2016). Advances in breeding for high grain zinc in rice. *Rice*, 9, 1-16.
- Takahashi, R., Bashir, K., Ishimaru, Y., Nishizawa, N. K., & Nakanishi, H. (2012). The role of heavy-metal ATPases, HMAs, in zinc and cadmium transport in rice. *Plant signaling & behavior*, 7(12), 1605-1607.
- Tamura, Y. (2021). The role of zinc homeostasis in the prevention of diabetes mellitus and cardiovascular diseases. *Journal of atherosclerosis and thrombosis*, 28(11), 1109-1122.
- Tao, Z. Q., Wang, D. M., Chang, X. H., Wang, Y. J., Yang, Y. S., & Zhao, G. C. (2018). Effects of zinc fertilizer and short-term high temperature stress on wheat grain production and wheat flour proteins. *Journal of integrative* agriculture, 17(9), 1979-1990.
- Tavallali, V., Rahemi, M., Maftoun, M., Panahi, B., Karimi, S., Ramezanian, A., & Vaezpour, M. (2009). Zinc influence and salt stress on photosynthesis, water relations, and carbonic anhydrase activity in pistachio. *Scientia horticulturae*, 123(2), 272-279.
- Tepfer, D. A., & Fosket, D. E. (1978). Hormone-mediated translational control of protein synthesis in cultured cells of Glycine max. *Developmental biology*, 62(2), 486-497.
- Thounaojam, T. C., Thounaojam, T. M., & Upadhyaya, H. (2021). Role of zinc oxide nanoparticles in mediating abiotic stress responses in plant. In *Zinc-based nanostructures for environmental and agricultural applications* 323-337.
- Toor, M. D., Adnan, M., Javed, M. S., Habibah, U., Arshad, A., Din, M. M., & Ahmad, R. (2020). Foliar application of Zn: Best way to mitigate drought stress in plants; A review. *International journal of applied research*, 6(8), 16-20.
- Topping, D. (2007). Cereal complex carbohydrates and their contribution to human health. *Journal of Cereal Science*, *46*(3), 220-229.

- Uauy, C., Distelfeld, A., Fahima, T., Blechl, A., & Dubcovsky, J. (2006). A NAC gene regulating senescence improves grain protein, zinc, and iron content in wheat. *Science*, 314(5803), 1298-1301.
- Ullah, A., Manghwar, H., Shaban, M., Khan, A. H., Akbar, A., Ali, U., ... & Fahad, S. (2018). Phytohormones enhanced drought tolerance in plants: a coping strategy. *Environmental science and pollution research*, 25, 33103-33118.
- Ullah, M. I., Mahpara, S., Bibi, R., Shah, R. U., Ullah, R., Abbas, S., ... & Khan, M. I. (2021). Grain yield and correlated traits of bread wheat lines: Implications for yield improvement. *Saudi journal of biological sciences*, 28(10), 5714-5719.
- Verret, F., Gravot, A., Auroy, P., Leonhardt, N., David, P., Nussaume, L., ... & Richaud, P. (2004). Overexpression of AtHMA4 enhances root-to-shoot translocation of zinc and cadmium and plant metal tolerance. *FEBS letters*, 576(3), 306-312.
- Walker, E. L., & Waters, B. M. (2011). The role of transition metal homeostasis in plant seed development. *Current opinion in plant biology*, *14*(3), 318-324.
- Wang, B., Wei, H., Xue, Z., & Zhang, W.H. (2017). Gibberellins regulate iron defciency-response by infuencing iron transport and translocation in rice seedlings (*Oryza sativa* L.). *Annals of botany*, 119, 945–956.
- Wang, J., Mao, H., Zhao, H., Huang, D., & Wang, Z. (2012). Different increases in maize and wheat grain zinc concentrations caused by soil and foliar applications of zinc in Loess Plateau, China. *Field crops research*, 135, 89-96.
- Wang, J., Qin, H., Zhou, S., Wei, P., Zhang, H., Zhou, Y., ... & Huang, R. (2020). The ubiquitin-binding protein OsDSK2a mediates seedling growth and salt responses by regulating gibberellin metabolism in rice. *The plant cell*, 32(2), 414-428.
- Wang, M., Kong, F., Liu, R., Fan, Q., & Zhang, X. (2020). Zinc in wheat grain, processing, and food. *Frontiers in nutrition*, *7*, 124.
- Wang, X., Mao, X., Yan, A., Tan, T., Yang, Y., & Wan, Y. (2016). Simultaneous determination of nine plant growth regulators in navel oranges by liquid chromatography-triple quadrupole tandem mass spectrometry. *Food analytical methods*, 9, 3268-3277.

- Wang, X., Zhong, F., Woo, C. H., Miao, Y., Grusak, M. A., Zhang, X., ... & Jiang, L. (2017). A rapid and efficient method to study the function of crop plant transporters in Arabidopsis. *Protoplasma*, 254, 737-747.
- Wang, Y. X., Specht, A., & Horst, W. J. (2011). Stable isotope labelling and zinc distribution in grains studied by laser ablation ICP-MS in an ear culture system reveals zinc transport barriers during grain filling in wheat. *New phytologist*, 189(2), 428-437.
- Wang, Z., Liu, Q., Pan, F., Yuan, L., & Yin, X. (2015). Effects of increasing rates of zinc fertilization on phytic acid and phytic acid/zinc molar ratio in zinc biofortified wheat. *Field crops research*, 184, 58-64.
- Waters, B. M., & Sankaran, R. P. (2011). Moving micronutrients from the soil to the seeds: genes and physiological processes from a biofortification perspective. *Plant science*, 180(4), 562-574.
- Weisany, W., Mohammadi, M., Tahir, N. A. R., Aslanian, N., & Omer, D. A. (2021). Changes in growth and nutrient status of maize (*Zea mays L.*) in response to two zinc sources under drought stress. *Journal of soil science and plant nutrition*, 21, 3367-3377.
- Wessells, K. R., & Brown, K. H. (2012). Estimating the global prevalence of zinc deficiency: results based on zinc availability in national food supplies and the prevalence of stunting. *Plos one*, 7(11), e50568.
- Wessels, I., Fischer, H. J., & Rink, L. (2021). Dietary and physiological effects of zinc on the immune system. *Annual review of nutrition*, *41*, 133-175.
- Wessels, I., Maywald, M., & Rink, L. (2017). Zinc as a gatekeeper of immune function. *Nutrients*, 9(12), 1286.
- White, P. J., & Broadley, M. R. (2011). Physiological limits to zinc biofortification of edible crops. *Frontiers in plant science*, *2*, 80.
- Williams, L. E., & Mills, R. F. (2005). P1B-ATPases–an ancient family of transition metal pumps with diverse functions in plants. *Trends in plant science*, 10(10), 491-502.

- Wissuwa, M., Ismail, A. M., & Yanagihara, S. (2006). Effects of zinc deficiency on rice growth and genetic factors contributing to tolerance. *Plant physiology*, 142(2), 731-741.
- Wu, C. Y., Lu, L. L., Yang, X. E., Feng, Y., Wei, Y. Y., Hao, H. L., ... & He, Z. L. (2010). Uptake, translocation, and remobilization of zinc absorbed at different growth stages by rice genotypes of different Zn densities. *Journal of agricultural and food chemistry*, 58(11), 6767-6773.
- Xia, J., Cai, L. H., Wu, H., MacKintosh, F. C., & Weitz, D. A. (2021). Anomalous mechanics of Zn2+-modified fibrin networks. *Proceedings of the national* academy of sciences, 118(10), e2020541118.
- Xie, Y., Zhang, Y., Han, J., Luo, J., Li, G., Huang, J., ... & Chen, L. (2018). The intronic cis element SE1 recruits trans-acting repressor complexes to repress the expression of elongated uppermost internode1 in rice. *Molecular plant*, 11(5), 720-735.
- Xi-wen, Y., Xiao-hong, T., Xin-chun, L., William, G., & Yu-xian, C. (2011). Foliar zinc fertilization improves the zinc nutritional value of wheat (*Triticum aestivum* L.) grain. *African journal of biotechnology*, 10(66), 14778-14785.
- Xu, J., Luo, X., Wang, Y., & Feng, Y. (2018). Evaluation of zinc oxide nanoparticles on lettuce (*Lactuca sativa* L.) growth and soil bacterial community. *Environmental science and pollution research*, 25, 6026-6035.
- Xu, K., Xu, X., Fukao, T., Canlas, P., Maghirang-Rodriguez, R., Heuer, S., ... & Mackill, D. J. (2006). Sub1A is an ethylene-response-factor-like gene that confers submergence tolerance to rice. *Nature*, 442(7103), 705-708.
- Yamaguchi, N., Ishikawa, S., Abe, T., Baba, K., Arao, T., & Terada, Y. (2012). Role of the node in controlling traffic of cadmium, zinc, and manganese in rice. *Journal of experimental botany*, 63(7), 2729-2737.
- Yamaji, N., Xia, J., Mitani-Ueno, N., Yokosho, K., & Feng Ma, J. (2013). Preferential delivery of zinc to developing tissues in rice is mediated by P-type heavy metal ATPase OsHMA2. *Plant physiology*, 162(2), 927-939.

- Yang, D., Luo, Y., Kong, X., Huang, C., & Wang, Z. (2021). Interactions between exogenous cytokinin and nitrogen application regulate tiller bud growth via sucrose and nitrogen allocation in winter wheat. *Journal of plant growth regulation*, 40, 329-341.
- Yin, H., Gao, X., Stomph, T., Li, L., Zhang, F., & Zou, C. (2016). Zinc concentration in rice (*Oryza sativa* L.) grains and allocation in plants as affected by different zinc fertilization strategies. *Communications in soil science and plant analysis*, 47(6), 761-768.
- Zaheer, M. S., Ali, H. H., Iqbal, M. A., Erinle, K. O., Javed, T., Iqbal, J., ... & Dessoky, E. S. (2022). Cytokinin production by *Azospirillum brasilense* contributes to increase in growth, yield, antioxidant, and physiological systems of wheat (*Triticum aestivum* L.). *Frontiers in microbiology*, 13, 886041.
- Zaheer, M. S., Raza, M. A. S., Saleem, M. F., Erinle, K. O., Iqbal, R., & Ahmad, S. (2019). Effect of rhizobacteria and cytokinins application on wheat growth and yield under normal vs drought conditions. *Communications in soil science and plant analysis*, 50(20), 2521-2533.
- Zahid, G., Iftikhar, S., Shimira, F., Ahmad, H. M., & Kaçar, Y. A. (2023). An overview and recent progress of plant growth regulators (PGRs) in the mitigation of abiotic stresses in fruits: A review. *Scientia horticulturae*, 309, 111621.
- Zahra, S., Shaheen, T., Qasim, M., Hussain, M., Zulfiqar, S., & Shaukat, K. (2023). Genome-wide survey of HMA gene family and its characterization in wheat (*Triticum aestivum*). *PeerJ*, 11, e14920.
- Zanewich, K. P., & Rood, S. B. (1995). Vernalization and gibberellin physiology of winter canola (endogenous gibberellin (GA) content and metabolism of [3H]
  GA1 and [3H] GA20. *Plant physiology*, *108*(2), 615-621.
- Zarea, M. J., & Karimi, N. (2023). Grain yield and quality of wheat are improved through post-flowering foliar application of zinc and 6-benzylaminopurine under water deficit condition. *Frontiers in plant science*, 13, 1068649.
- Zhang, D. X., Wang, M. Y., Lin, W. B., Qu, S., Ji, L., Xu, C., ... & Dong, K. (2023). Recent advances in emerging application of functional materials in sample

pretreatment methods for liquid chromatography-mass spectrometry analysis of plant growth regulators: A mini-review. *Journal of chromatography A*, 464130.

- Zhang, H., Sun, X., & Dai, M. (2022). Improving crop drought resistance with plant growth regulators and rhizobacteria: Mechanisms, applications, and perspectives. *Plant communications*, 3(1).
- Zhang, X., Kong, J., Yu, L., Wang, A., Yang, Y., Li, X., & Wang, J. (2024). Functional characterization of *Fagopyrum tataricum* ZIP gene family as a metal ion transporter. *Frontiers in Plant Science*, 15, 1373066.
- Zhao A, Zhang L, Ning P, Chen Q, Wang B, Zhang F, Yang X, Zhang Y (2021) Zinc in cereal grains: Concentration, distribution, speciation, bioavailability, and barriers to transport from roots to grains in wheat. *Critical reviews in food science and nutrition*, 62(28), 7917-7928.
- Zhao, Y., Hu, Y., Dai, M., Huang, L., & Zhou, D. X. (2009). The WUSCHEL-related homeobox gene WOX11 is required to activate shoot-borne crown root development in rice. *The plant cell*, 21(3), 736-748.
- Zhiguo, E., Tingting, L. I., Chen, C. H. E. N., & Lei, W. A. N. G. (2018). Genomewide survey and expression analysis of P1B-ATPases in rice, maize and sorghum. *Rice Science*, 25(4), 208-217.
- Zhou, H., Zhang, T., Harmon, J. S., Bryan, J., & Robertson, R. P. (2007). Zinc, not insulin, regulates the rat α-cell response to hypoglycemia in vivo. *Diabetes*, 56(4), 1107-1112.
- Zhu, Y., Nomura, T., Xu, Y., Zhang, Y., Peng, Y., Mao, B., ... & He, Z. (2006). Elongated uppermost internode encodes a cytochrome P450 monooxygenase that epoxidizes gibberellins in a novel deactivation reaction in rice. *The plant cell*, 18(2), 442-456.
- Zhuang, Q. S. (2003). Chinese wheat improvement and pedigree analysis. *Beijing: China agriculture*, 550-7.
- Zou, C. Q., Zhang, Y. Q., Rashid, A., Ram, H., Savasli, E., Arisoy, R. Z., ... & Cakmak, I. (2012). Biofortification of wheat with zinc through zinc fertilization in seven countries. *Plant and soil*, 361, 119-130.

Zou, C., Gao, X., Shi, R., Fan, X., & Zhang, F. (2008). Micronutrient deficiencies in crop production in China. *Micronutrient deficiencies in global crop* production, 127-148.

# Annexure 1

Month	Meteorological weeks	-	Temperature (°C)		Relative Humidity (%)	Wind speed (km/hr)
		Max.	Min.	_		
er	<b>46</b> (12-18 Nov)	26.7	13.6	0.0	56.0	2.3
November	<b>47</b> (19-25 Nov)	24.3	13.3	0.0	57.0	1.4
Nov	<b>48</b> (26- 2 Dec)	21.4	11.0	0.0	56.4	0.6
	<b>49</b> (3 –9 Dec)	21.3	11.4	0.0	66.4	1.4
nber	<b>50</b> (10-16 Dec)	19.9	9.1	0.0	65.3	1.7
December	<b>51</b> (17 – 23 Dec)	20.1	9.0	0.0	67.1	0.9
Ď	<b>52</b> (24- 31 Dec)	18.9	8.8	0.1	65.0	1.0
	<b>1</b> (1–7 Jan)	16.3	8.4	9.0	74.1	1.1
ary	<b>2</b> (8 –14 Jan)	14.6	10.3	6.1	76.0	3.1
January	<b>3</b> (15–21 Jan)	17.4	11.4	0.3	74.9	2.0
ſ	<b>4</b> (22– 28 Jan)	14.9	11.7	0.0	76.9	2.3
	<b>5</b> (29 – 4 Feb)	16.4	8.4	0.1	78.3	3.0
ary	<b>6</b> (5–11 Feb)	12.6	8.7	0.0	77.3	3.0
February	<b>7</b> (12 - 18 Feb)	12.6	8.7	0.0	77.3	3.0
ų	8 (19 -25 Feb)	17.1	8.4	0.0	75.7	3.2
	<b>9</b> (26 – 4 March)	19.6	10.4	0.0	68.7	3.0
	10 (5-11 March)	17.1	9.3	2.3	60.9	3.1
March	11(12–18 March)	21.3	14.3	0.0	54.7	1.1
Μ	12(19–25 March)	26.9	19.9	0.0	55.1	2.3
	<b>13</b> (26 - 1 April)	31.9	21.6	0.0	52.4	2.3
	14 (2– 8 April)	31.4	21.0	0.0	51.1	4.0
ii	<b>15</b> (9 – 15 April)	33.1	24.1	0.0	47.3	3.4
April	<b>16</b> (16–22 April)	41.3	27.1	0.1	44.6	4.0
	<b>17</b> (23–29 April)	39.4	28.7	0.0	41.7	12.3

Weekly mean meteorological data recorded for growing season 2021-22.

# Annexure 2

Month	Meteorological weeks	Temperature (°C)		Rainfall (mm)	Relative Humidity (%)	Wind speed (km/hr)
		Max.	Min.	_		
er	<b>46</b> (12-18 Nov)	26.7	13.6	0.0	56.0	2.3
November	<b>47</b> (19-25 Nov)	24.3	13.4	0.0	57.0	2.3
Nov	<b>48</b> (26- 2 Dec)	22.6	11.0	0.0	64.4	0.3
	<b>49</b> (3 –9 Dec)	27.0	10.9	0.0	86.6	3.3
nber	<b>50</b> (10-16 Dec)	26.7	11.4	0.0	89.1	8.4
December	<b>51</b> (17 – 23 Dec)	24.6	9.9	0.0	90.4	4.6
Ă	<b>52</b> (24- 31 Dec)	21.6	8.6	0.3	94.3	7.3
	<b>1</b> (1– 7 Jan)	14.4	5.5	0.0	94.1	6.6
ary	<b>2</b> (8 –14 Jan)	13.5	8.4	0.3	95.6	8.9
January	<b>3</b> (15–21 Jan)	14.4	5.9	0.0	91.4	6.7
ſ	<b>4</b> (22– 28 Jan)	18.7	7.6	4.9	84.1	7.7
	<b>5</b> (29 – 4 Feb)	20.1	8.7	1.8	94.6	11.4
ary	<b>6</b> (5–11 Feb)	23.7	11.8	0.0	80.3	10.1
February	<b>7</b> (12 - 18 Feb)	26.0	10.8	0.0	63.0	9.6
ų	8 (19 -25 Feb)	28.1	14.3	0.0	92.3	9.9
	<b>9</b> (26 – 4 March)	27.4	14.0	0.0	66.3	9.1
	10 (5-11 March)	29.4	14.0	0.0	46.4	8.7
March	11(12–18 March)	27.8	16.7	2.1	74.4	10.6
Μ	12(19–25 March)	24.7	13.9	5.5	93.3	1.8
	<b>13</b> (26 - 1 April)	27.6	15.7	0.3	88.7	10.1
	14 (2-8 April)	27.8	14.2	0.3	65.1	2.9
ii	<b>15</b> (9 – 15 April)	35.5	16.1	0.0	75.7	3.4
April	<b>16</b> (16–22 April)	35.3	17.2	1.3	79.6	4.9
	17 (23– 29 April)	35.0	18.2	0.0	76.9	9.4

Weekly mean meteorological data recorded for growing season 2022-23.

### **APPENDIX III**

## ANOVA TABLES

```
Plant height 30 DAS 2021-22
Analysis of Variance Table
Response: Yield
      Df Sum Sq Mean Sq F value Pr(>F)
block
          1.32
                  0.66
                        0.7530 0.478217
       2
       1 529.21 529.21 985.3207 0.001013 **
V
                 0.54
       2
          1.07
Ea
       2 12.68
                  6.34
                        1.8227 0.222717
Zn
v:Zn
       2
         0.23 0.11 0.0324 0.968260
       8 27.82
                 3.48
Eb
Н
       3
          1.41 0.47 0.5336 0.662173
      3 0.84 0.28 0.3185 0.811888
H:v
      6 3.63 0.61 0.6898 0.659180
H:Zn
H:v:Zn 6 3.17 0.53 0.6012 0.727423
Ec
     36 31.62 0.88
- - -
Signif. codes: 0 (***' 0.001 (**' 0.01 (*' 0.05 (.' 0.1 (') 1
cv(a) = 5.7 %, cv(b) = 14.5 %, cv(c) = 7.3 %, Mean = 12.88056
```

```
Plant height 60 DAS 2021-22
Analysis of Variance Table
Response: Yield
      Df Sum Sq Mean Sq F value Pr(>F)
       2 1.91
                 0.96
                        0.4644 0.632194
block
       1 565.66 565.66 203.7044 0.004873 **
V
       2
          5.55
                 2.78
Ea
       2 109.62
                54.81 13.3631 0.002817 **
Zn
      2 0.03 0.02 0.0038 0.996253
v:Zn
      8 32.81
                4.10
Eb
       3 25.56 8.52 4.1433 0.012738 *
Η
      3 0.77 0.26 0.1251 0.944625
H:v
         0.58 0.10 0.0471 0.999513
H:Zn
      6
                        0.0521 0.999349
H:v:Zn 6 0.64 0.11
      36 74.03
               2.06
Ec
- - -
Signif. codes: 0 (***' 0.001 (**' 0.01 (*' 0.05 (.' 0.1 (') 1
cv(a) = 4.3 %, cv(b) = 5.2 %, cv(c) = 3.7 %, Mean = 38.57653
```

Plant height 90 DAS 2021-22 Analysis of Variance Table Response: Yield Df Sum Sq Mean Sq F value Pr(>F) 2 20.81 10.41 3.3536 0.046176 \* block 1 628.41 628.41 69.6353 0.014058 \* V Ea 2 18.05 9.02 2 236.98 118.49 15.5062 0.001768 \*\* Zn v:Zn 2 0.50 0.25 0.0325 0.968156 Eb 8 61.13 7.64 3 111.24 37.08 11.9506 1.402e-05 \*\*\* Н 3 2.30 0.77 0.2472 0.862773 H:v H:Zn 6 4.69 0.78 0.2519 0.955329 H:v:Zn 6 5.22 0.87 0.2803 0.942522 Ec 36 111.70 3.10 \_ \_ \_ Signif. codes: 0 (\*\*\*' 0.001 (\*\*' 0.01 (\*' 0.05 (.' 0.1 (' 1 cv(a) = 6.2 %, cv(b) = 5.7 %, cv(c) = 3.6 %, Mean = 48.80486

```
Plant height 120 DAS 2021-22
Analysis of Variance Table
Response: Yield
      Df Sum Sq Mean Sq F value Pr(>F)
block
                  1.45
                        0.2898 0.7501606
      2
           2.89
       1 2452.92 2452.92 4358.8588 0.0002293 ***
V
                 0.56
Ea
      2
           1.13
       2 145.75 72.87
Zn
                       92.6388 2.935e-06 ***
      2 0.59 0.30 0.3780 0.6968628
v:Zn
                 0.79
Eb
      8
           6.29
       3 115.05 38.35 7.6853 0.0004288 ***
н
      3
          0.16 0.05 0.0105 0.9984997
H:v
          2.46 0.41
                        0.0822 0.9976351
      6
H:Zn
          1.42 0.24 0.0476 0.9994978
H:v:Zn 6
      36 179.63
                 4.99
Ec
- - -
Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
cv(a) = 1 %, cv(b) = 1.1 %, cv(c) = 2.9 %, Mean = 77.87292
```

```
Plant height at harvest 2021-22
Analysis of Variance Table
Response: Yield
      Df Sum Sq Mean Sq F value Pr(>F)
block
       2
           19.7
                   9.9
                         2.1653 0.129428
       1 6619.2 6619.2 618.2265 0.001614 **
V
Ea
       2 21.4
                  10.7
Zn
       2 154.8
                 77.4 15.1353 0.001909 **
                  0.2
v:Zn
       2
           0.3
                       0.0322 0.968483
Eb
       8 40.9
                  5.1
       3 81.6 27.2 5.9706 0.002062 **
H
H:v
       3
          0.8
                 0.3
                        0.0598 0.980543
      6 42.1 7.0
                       1.5413 0.192921
H:Zn
H:v:Zn 6 22.6
                  3.8
                        0.8262 0.557355
Ec
      36 164.1
                 4.6
_ _ _
Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
cv(a) = 3.6 %, cv(b) = 2.5 %, cv(c) = 2.3 %, Mean = 91.36486
```

```
Tillers 30 DAS 2021-22
Analysis of Variance Table
Response: Yield
      Df Sum Sq Mean Sq F value
                                    Pr(>F)
block
       2
            93.0
                   46.5
                          0.6802 0.5129177
       1 16972.4 16972.4 324.2308 0.0030700 **
V
           104.7
                   52.3
Ea
       2
       2 4168.4 2084.2 25.5023 0.0003379 ***
Zn
       2
                   87.1
                         1.0656 0.3887899
v:Zn
          174.2
       8 653.8
                   81.7
Eb
           36.7 12.2
Н
       3
                        0.1788 0.9100756
                  0.4 0.0060 0.9993537
H:v
       3
            1.2
H:Zn
       6
            37.7
                   6.3 0.0919 0.9967693
                   1.8
                          0.0262 0.9999113
H:v:Zn 6
            10.8
Ec
      36 2461.7
                   68.4
- - -
Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
cv(a) = 3.8 %, cv(b) = 4.8 %, cv(c) = 4.3 %, Mean = 190.1044
```

Tillers 60 DAS 2021-22 Analysis of Variance Table Response: Yield Df Sum Sq Mean Sq F value Pr(>F) 545 273 0.8938 0.4180017 block 2 33867 50.8283 0.0191119 \* 1 33867 V 666 Ea 2 1333 Zn 2 27740 13870 21.2054 0.0006343 \*\*\* 265 133 0.2028 0.8205287 v:Zn 2 Eb 8 5233 654 3 7478 2493 8.1703 0.0002813 \*\*\* H H:v 3 31 10 0.0343 0.9913249 H:Zn 6 312 52 0.1702 0.9831476 H:v:Zn 6 106 18 0.0580 0.9991159 36 10983 305 Ec \_ \_ \_ Signif. codes: 0 '\*\*\*' 0.001 '\*\*' 0.01 '\*' 0.05 '.' 0.1 ' ' 1 cv(a) = 5.8 %, cv(b) = 5.8 %, cv(c) = 3.9 %, Mean = 443.6028

	Tillers 90 DAS 2021-22								
Analysis of Variance Table									
Respons	se:	Yield							
			Mean Sq	F value	Pr(>F)				
block	2	729	365	1.1966	0.3139635				
V	1	50924	50924	107.6506	0.0091618	**			
Ea	2	946	473						
				16.4862	0.0014534	**			
v:Zn	2	178	89	0.1093	0.8977653				
Eb	8	6528	816						
Н	3	7708	2569	8.4344	0.0002244	***			
H:v	3	34	11	0.0374	0.9901691				
H:Zn	6	285	47	0.1557	0.9866268				
H:v:Zn	6	103	17	0.0565	0.9991792				
Ec	36	10966	305						
Signif	. cc	des: (	3 (***) (	0.001 '**	0.01 '*'	0.05 '.' 0.1 '' 1			
cv(a) =	= 5.	8 %, c	v(b) = 7	.6 %, cv(	() = 4.6 %	, Mean = 377.6942			
				•					

Tillers 120 DAS 2021-22 Analysis of Variance Table Response: Yield Df Sum Sq Mean Sq F value Pr(>F) block 143 71 0.3690 0.694025 2 1 59144 59144 549.0833 0.001816 \*\* V Ea 2 215 108 2 20648 10324 15.4010 0.001807 \*\* Zn 2 116 58 0.0862 0.918226 v:Zn 8 5363 Eb 670 2667 13.7699 3.871e-06 \*\*\* H 3 8001 H:v 3 3 0.0138 0.997748 8 6 395 H:Zn 66 0.3395 0.911327 15 H:v:Zn 6 3 0.0129 0.999989 Ec 36 6973 194 - - -Signif. codes: 0 '\*\*\*' 0.001 '\*\*' 0.01 '\*' 0.05 '.' 0.1 ' ' 1 cv(a) = 3 %, cv(b) = 7.6 %, cv(c) = 4.1 %, Mean = 342.0004

	LAI 30 DAS 2021-22							
Analysi	is of Variance Table							
Respons	se: Yield							
кезропз	Df Sum Sq Mean Sq F value Pr(>F)							
block	2 0.000392 0.000196 0.1048 0.900806							
	1 0.243296 0.243296 109.6397 0.008998 **							
	2 0.004438 0.002219							
	2 0.008930 0.004465 0.3871 0.691098							
	2 0.002659 0.001329 0.1153 0.892593							
	8 0.092277 0.011535							
	3 0.001557 0.000519 0.2773 0.841424							
	3 0.001341 0.000447 0.2388 0.868687							
	6 0.009156 0.001526 0.8153 0.565239							
	6 0.004722 0.000787 0.4205 0.860467							
	36 0.067383 0.001872							
	50 0.007585 0.001872							
Signif	codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1							
Jighth.								
cv(a) =	= 10.6 %, cv(b) = 24.1 %, cv(c) = 9.7 %, Mean = 0.4461856							
	-10.0 %, $CV(0) = 24.1$ %, $CV(0) = 9.7$ %, Mean = 0.4401830							

LAI 60 DAS 2021-22 Analysis of Variance Table Response: Yield Df Sum Sq Mean Sq F value Pr(>F) block 2 0.00351 0.00176 0.1574 0.854930 1 0.55785 0.55785 334.2156 0.002979 \*\* V 2 0.00334 0.00167 Ea Zn 2 0.21681 0.10841 6.9961 0.017510 \* 2 0.00251 0.00125 0.0809 0.923041 v:Zn 8 0.12396 0.01550 Eb Н 3 0.21892 0.07297 6.5431 0.001203 \*\* H:v 3 0.00321 0.00107 0.0959 0.961824 6 0.00287 0.00048 0.0429 0.999627 H:Zn H:v:Zn 6 0.02229 0.00371 0.3331 0.914981 36 0.40151 0.01115 Ec - - -Signif. codes: 0 (\*\*\*' 0.001 (\*\*' 0.01 (\*' 0.05 (.' 0.1 (' 1 cv(a) = 2 %, cv(b) = 6.1 %, cv(c) = 5.1 %, Mean = 2.055466

	LAI 90 DAS 2021-22								
Analys	Analysis of Variance Table								
		-							
Respon	se:	Yield							
	Df	Sum Sq	Mean Sq	F value	Pr(>F)				
block	2	0.19287	0.09644	2.9894	0.0629349				
V	1	1.39588	1.39588	23.0873	0.0406885	*			
Ea	2	0.12092	0.06046						
Zn	2	1.58658	0.79329	13.2460	0.0028940	**			
v:Zn	2	0.04845	0.02422	0.4045	0.6802432				
Eb	8	0.47911	0.05989						
Н	3	0.87761	0.29254	9.0685	0.0001319	***			
H:v	3	0.00308	0.00103	0.0318	0.9922409				
H:Zn	6	0.01592	0.00265	0.0823	0.9976266				
H:v:Zn	6	0.02079	0.00346	0.1074	0.9950459				
Ec	36	1.16131	0.03226						
Signif	. co	odes: 0	(***) 0	.001 '**	' 0.01 '*'	0.05 '.' 0.1 ' ' 1			
cv(a)	= 6	.1 %, cv	(b) = 6.2	1 %, cv(d	c) = 4.5 %	, Mean = 4.00581			

LAI 120 DAS 2021-22 Analysis of Variance Table Response: Yield Df Sum Sq Mean Sq F value Pr(>F) block 2 0.14827 0.07414 6.4738 0.0039653 \*\* 1 0.81533 0.81533 42.9894 0.0224801 \* V 2 0.03793 0.01897 Ea 2 1.33576 0.66788 24.4049 0.0003932 \*\*\* Zn v:Zn 2 0.04957 0.02478 0.9056 0.4420489 Fb 8 0.21893 0.02737 H 3 0.46687 0.15562 13.5894 4.38e-06 \*\*\* 3 0.00760 0.00253 0.2213 0.8809424 H:v 6 0.00916 0.00153 0.1333 0.9911281 H:Zn H:v:Zn 6 0.01293 0.00216 0.1882 0.9782265 36 0.41227 0.01145 Ec \_ \_ \_ Signif. codes: 0 '\*\*\*' 0.001 '\*\*' 0.01 '\*' 0.05 '.' 0.1 ' ' 1 cv(a) = 4.4 %, cv(b) = 5.3 %, cv(c) = 3.4 %, Mean = 3.109273

```
DMA 30 DAS 2021-22
Analysis of Variance Table
Response: Yield
      Df Sum Sq Mean Sq F value Pr(>F)
block
       2
         68.48 34.24 1.0425 0.36299
       1 690.06 690.06 90.4949 0.01087 *
V
Ea
       2 15.25 7.63
       2 167.89 83.95 2.2634 0.16635
Zn
v:Zn
       2 99.21 49.61 1.3374 0.31543
       8 296.71 37.09
Eb
       3
Н
           1.55 0.52 0.0157 0.99726
           2.14
                  0.71 0.0217 0.99558
H:v
       3
H:Zn
       6 10.81
                  1.80 0.0549 0.99924
            8.60 1.43 0.0436 0.99961
H:v:Zn 6
      36 1182.45 32.85
Ec
- - -
Signif. codes: 0 (***' 0.001 (**' 0.01 (*' 0.05 (.' 0.1 (' 1
cv(a) = 6.9 %, cv(b) = 15.2 %, cv(c) = 14.3 %, Mean = 40.02917
```

DMA 60 DAS 2021-22 Analysis of Variance Table Response: Yield Df Sum Sq Mean Sq F value Pr(>F) block 379 189 0.1671 0.846728 2 V 1 97770 97770 378.7031 0.002630 \*\* Ea 2 516 258 2 14384 7192 8.7769 0.009606 \*\* Zn v:Zn 2 590 295 0.3603 0.708226 Eb 8 6555 819 3 7456 2485 2.1925 0.105788 Н 3 148 0.1306 0.941247 H:v 444 H:Zn 6 378 63 0.0555 0.999219 H:v:Zn 6 532 89 0.0782 0.997936 Ec 36 40808 1134 - - -Signif. codes: 0 (\*\*\*' 0.001 (\*\*' 0.01 (\*' 0.05 (.' 0.1 (') 1 cv(a) = 7.2 %, cv(b) = 12.9 %, cv(c) = 15.1 %, Mean = 222.35

#### DMA 90 DAS 2021-22 Analysis of Variance Table Response: Yield Df Sum Sq Mean Sq F value Pr(>F)block 2 153 76 0.0651 0.937121 1 250384 250384 278.9125 0.003566 \*\* V Ea 2 1795 898 13183 13.2154 0.002915 \*\* Zn 2 26366 2 45 0.0453 0.955925 v:Zn 90 7980 998 Eb 8 3 15342 5114 4.3589 0.010189 \* Н 3 1273 424 0.3617 0.781032 H:v H:Zn 6 993 165 0.1410 0.989693 0.0594 0.999052 H:v:Zn 6 418 70 36 42236 Ec 1173 \_ \_ \_ Signif. codes: 0 (\*\*\*' 0.001 (\*\*' 0.01 (\*' 0.05 (.' 0.1 (' 1 cv(a) = 6.4 %, cv(b) = 6.8 %, cv(c) = 7.4 %, Mean = 464.5819

DMA 120 DAS 2021-22 Analysis of Variance Table Response: Yield Df Sum Sq Mean Sq F value Pr(>F) block 2 1575 788 0.3610 0.6995053 V 1 207905 207905 245.7345 0.0040448 \*\* Ea 2 1692 846 2 68323 34161 11.6800 0.0042351 \*\* Zn v:Zn 2 204 102 0.0349 0.9658408 8 23398 Eb 2925 3 47053 15684 7.1890 0.0006669 \*\*\* Н 3 1153 384 0.1762 0.9118169 H:v H:Zn 6 2574 429 0.1966 0.9756877 999 0.4578 0.8345963 H:v:Zn 6 5993 Ec 36 78542 2182 - - -Signif. codes: 0 (\*\*\*' 0.001 (\*\*' 0.01 (\*' 0.05 (.' 0.1 (' 1 cv(a) = 3.7 %, cv(b) = 6.9 %, cv(c) = 6 %, Mean = 778.9583

Effective tillers 2021-22								
Analys:	is d	of Varia	ance Tab	le				
Respons	se:	Yield						
	Df	Sum Sq	Mean Sq	F value	Pr(>F)			
block	2	29	14	0.0645	0.9376758			
V	1	47965	47965	1146.3853	0.0008712	***		
Ea	2	84	42					
Zn	2	20164	10082	24.5935	0.0003830	***		
v:Zn	2	322	161	0.3924	0.6877663			
Eb	8	3280	410					
Н	3	7877	2626	11.6975	1.689e-05	***		
H:v	3	18	6	0.0272	0.9938350			
H:Zn	6	343	57	0.2544	0.9542555			
H:v:Zn	6	12	2	0.0092	0.9999960			
Ec	36	8081	224					
Signif	. co	odes: (	) (***) (	0.001 (**)	0.01 (*)	0.05 '.' 0.1 ' ' 1		
cv(a)	= 2.	.2 %, c	(b) = 6	.8 %, cv(c	) = 5.1 %,	Mean = 296.1133		
. ,								

```
Spike length 2021-22
Analysis of Variance Table
Response: Yield
      Df Sum Sq Mean Sq F value Pr(>F)
       2 1.2025 0.6012 2.2909 0.11574
block
V
       1 16.1501 16.1501 34.5355 0.02776 *
Ea
       2 0.9353 0.4676
       2 4.5025 2.2512 7.0890 0.01693 *
Zn
       2 0.1753 0.0876 0.2760 0.76578
v:Zn
       8 2.5406 0.3176
Eb
       3 2.5871 0.8624 3.2858 0.03164 *
Н
       3 0.0415 0.0138 0.0527 0.98377
H:v
H:Zn
      6 0.3542 0.0590 0.2249 0.96605
H:v:Zn 6 0.2014 0.0336 0.1279 0.99206
Ec
      36 9.4483 0.2625
- - -
Signif. codes: 0 (***' 0.001 (**' 0.01 (*' 0.05 (.' 0.1 (' 1
cv(a) = 7 %, cv(b) = 5.8 %, cv(c) = 5.2 %, Mean = 9.7875
```

```
Number of grains per spike 2021-22
Analysis of Variance Table
Response: Yield
      Df Sum Sq Mean Sq F value Pr(>F)
       2 86.11 43.06 2.4467 0.100850
block
V
       1 672.22 672.22 21.8807 0.042790 *
Ea
       2 61.44 30.72
       2 317.19 158.60 5.4157 0.032571 *
Zn
v:Zn
       2 1.69 0.85 0.0289 0.971585
       8 234.28
                29.28
Eb
       3 280.11 93.37 5.3060 0.003926 **
Н
H:v
       3
           9.44
                  3.15 0.1789 0.910010
H:Zn
       6 9.14
                  1.52 0.0866 0.997265
H:v:Zn 6 3.31
                  0.55 0.0313 0.999851
Ec
      36 633.50 17.60
_ _ _
Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
cv(a) = 13.1 \%, cv(b) = 12.8 \%, cv(c) = 9.9 \%, Mean = 42.27778
```

1000 – grain weight 2021-22 Analysis of Variance Table Response: Yield Df Sum Sq Mean Sq F value Pr(>F) 2 3.583 1.792 0.9258 0.4054275 block V 1 36.125 36.125 32.1111 0.0297588 \* Ea 2 2.250 1.125 2 66.333 33.167 25.2698 0.0003488 \*\*\* Zn 2 0.333 0.167 0.1270 0.8824879 v:Zn Eb 8 10.500 1.313 3 60.597 20.199 10.4378 4.396e-05 \*\*\* Η 3 0.931 0.310 0.1603 0.9223422 H:v H:Zn 6 0.778 0.130 0.0670 0.9986670 H:v:Zn 6 2.778 0.463 0.2392 0.9605439 Ec 36 69.667 1.935 \_ \_ \_ Signif. codes: 0 '\*\*\*' 0.001 '\*\*' 0.01 '\*' 0.05 '.' 0.1 ' ' 1 cv(a) = 2.5 %, cv(b) = 2.7 %, cv(c) = 3.3 %, Mean = 41.875

	Grain yield 2021-22									
Analysi	Analysis of Variance Table									
Deener		V: JJ								
Respons				_						
		1.00			Pr(>F)					
block	2	10.450	5.225	0.7964	0.458752					
V	1	299.635	299.635	34.1616	0.028047	*				
Ea	2	17.542	8.771							
Zn	2	220.078	110.039	11.9088	0.003997	**				
v:Zn	2	4.652	2.326	0.2517	0.783374					
Eb	8	73.921	9.240							
Н	3	128.009	42.670	6.5032	0.001249	**				
H:v	3	0.946	0.315	0.0480	0.985824					
H:Zn	6	6.086	1.014	0.1546	0.986880					
H:v:Zn	6	0.304	0.051	0.0077	0.999998					
Ec	36	236.208	6.561							
Signif.	. C(	odes: 0	·*** 0	.001 ***	° 0.01 '*	0.05 '.'	0.1 '	ر	1	
-									_	
cv(a) =	= 6	.8 %, cv	(b) = 7	%, cv(c)	= 5.9 %,	Mean = $43$	.45722			

Straw yield 2021-22 Analysis of Variance Table Response: Yield Df Sum Sq Mean Sq F value Pr(>F) 0.0288 0.9716395 block 2 0.58 0.29 1 968.00 968.00 132.7543 0.0074487 \*\* V Ea 2 14.58 7.29 2 487.75 243.87 12.8779 0.0031548 \*\* Zn 0.2002 0.8225301 v:Zn 2 7.58 3.79 Eb 8 151.50 18.94 3 266.33 88.78 8.7642 0.0001699 \*\*\* Н 1.59 0.1572 0.9243425 H:v 3 4.78 5.93 0.5855 0.7395340 H:Zn 6 35.58 H:v:Zn 6 8.64 1.44 0.1421 0.9894787 36 364.67 10.13 Ec \_ \_ \_ Signif. codes: 0 (\*\*\*' 0.001 (\*\*' 0.01 (\*' 0.05 (.' 0.1 (' 1 cv(a) = 4.6 %, cv(b) = 7.3 %, cv(c) = 5.4 %, Mean = 59.33333

#### Harvest index 2021-22 Analysis of Variance Table Response: Yield Df Sum Sq Mean Sq F value Pr(>F) 4.031 2.0154 0.6855 0.51030 block 2 8.795 8.7954 7.1866 0.11553 V 1 Ea 2 2.448 1.2239 Zn 2 0.369 0.1844 0.0393 0.96169 v:Zn 2 4.234 2.1170 0.4506 0.65247 Eb 8 37.583 4.6979 H 3 35.755 11.9183 4.0537 0.01398 \* H:v 3 0.395 0.1316 0.0448 0.98721 3.914 0.6524 0.2219 0.96716 H:Zn 6 H:v:Zn 6 1.696 0.2827 0.0962 0.99634 36 105.843 2.9401 Ec \_ \_ \_ Signif. codes: 0 (\*\*\*' 0.001 (\*\*' 0.01 (\*' 0.05 (.' 0.1 (') 1 cv(a) = 2.6 %, cv(b) = 5.1 %, cv(c) = 4.1 %, Mean = 42.30713

```
Grain protein content 2021-22
Analysis of Variance Table
Response: Yield
      Df Sum Sq Mean Sq F value Pr(>F)
       2 0.690 0.345 0.9770 0.386206
block
V
       1 51.069 51.069 24.4353 0.038572 *
       2 4.180 2.090
Ea
       2 14.222 7.111 15.7896 0.001669 **
Zn
v:Zn
       2 0.004 0.002 0.0044 0.995619
Eb
       8 3.603 0.450
H
       3 3.563 1.188 3.3609 0.029177 *
       3 0.095 0.032 0.0892 0.965493
H:v
H:Zn
      6 0.365 0.061 0.1722 0.982645
H:v:Zn 6 0.346 0.058 0.1634 0.984852
Ec
      36 12.721 0.353
_ _ _
Signif. codes: 0 (***' 0.001 (**' 0.01 (*' 0.05 (.' 0.1 (') 1
cv(a) = 13 %, cv(b) = 6 %, cv(c) = 5.3 %, Mean = 11.13145
```

```
Grain phytic acid content 2021-22
Analysis of Variance Table
Response: Yield
       Df
           Sum Sq Mean Sq F value Pr(>F)
block
       2 0.075709 0.037855 7.4733 0.0019292 **
V
       1 0.008216 0.008216 0.1341 0.7493445
Ea
       2 0.122560 0.061280
       2 0.277155 0.138578 13.9295 0.0024772 **
Zn
v:Zn
       2 0.001601 0.000801 0.0805 0.9234146
Eb
       8 0.079588 0.009949
Н
       3 0.112206 0.037402 7.3839 0.0005601 ***
H:v
       3 0.000502 0.000167 0.0330 0.9917967
       6 0.000156 0.000026 0.0051 0.9999993
H:Zn
H:v:Zn 6 0.000695 0.000116 0.0229 0.9999405
       36 0.182351 0.005065
Ec
- - -
Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 '' 1
cv(a) = 23.9 %, cv(b) = 9.6 %, cv(c) = 6.9 %, Mean = 1.037334
```

Grain Zn content 2021-22 Analysis of Variance Table Response: Yield Df Sum Sq Mean Sq F value Pr(>F)block 2 0.09 0.05 5.2300e-02 0.94910 1 626.43 626.43 1.4652e+05 6.825e-06 \*\*\* V Ea 2 0.01 0.00 2 545.07 272.53 6.6462e+02 1.281e-09 \*\*\* Zn 1.81 4.4159e+00 0.05103 . v:Zn 2 3.62 Eb 8 3.28 0.41 Н 3 82.30 27.43 3.0639e+01 5.133e-10 \*\*\* H:v 3 1.75 0.58 6.5110e-01 0.58746 6 1.68 0.28 3.1340e-01 0.92582 H:Zn H:v:Zn 6 5.85 0.97 1.0881e+00 0.38806 36 32.23 0.90 Ec \_ \_ \_ Signif. codes: 0 '\*\*\*' 0.001 '\*\*' 0.01 '\*' 0.05 '.' 0.1 ' ' 1 cv(a) = 0.2 %, cv(b) = 2.1 %, cv(c) = 3.2 %, Mean = 29.80021

```
Steam Zn content 2021-22
Analysis of Variance Table
Response: Yield
      Df Sum Sq Mean Sq F value Pr(>F)
block
          2.00
                 1.00
                        0.4680 0.630011
       2
V
      1 406.93 406.93 253.0354 0.003929 **
          3.22
                 1.61
Ea
       2
      2 197.10 98.55 67.0964 1.002e-05 ***
Zn
         6.22
                3.11 2.1171 0.182833
v:Zn
      2
Eb
      8 11.75
                 1.47
      3 49.80 16.60 7.7674 0.000399 ***
Н
         0.80 0.27 0.1243 0.945129
H:v
       3
      6 6.63 1.11 0.5174 0.791196
H:Zn
H:v:Zn 6 0.50 0.08 0.0388 0.999722
Ec
      36 76.94
                2.14
Signif. codes: 0 (***) 0.001 (**) 0.05 (.' 0.1 (') 1
cv(a) = 6\%, cv(b) = 5.7\%, cv(c) = 6.9\%, Mean = 21.28764
```

Leaf Zn content 2021-22 Analysis of Variance Table Response: Yield Df Sum Sq Mean Sq F value Pr(>F) block 2 0.404 0.202 0.5723 0.56926 V 1 78.292 78.292 45.4100 0.02132 \* Ea 2 3.448 1.724 2 51.550 25.775 50.7288 2.853e-05 \*\*\* Zn 2 2.177 1.089 2.1423 0.17985 v:Zn Eb 8 4.065 0.508 H 3 24.636 8.212 23.2660 1.504e-08 \*\*\* H:v 3 0.299 0.100 0.2824 0.83778 6 1.984 0.331 0.9371 0.48074 H:Zn H:v:Zn 6 0.138 0.023 0.0651 0.99877 36 12.707 0.353 Ec \_ \_ \_ Signif. codes: 0 (\*\*\*' 0.001 (\*\*' 0.01 (\*' 0.05 (.' 0.1 (') 1 cv(a) = 14.8 %, cv(b) = 8.1 %, cv(c) = 6.7 %, Mean = 8.849444

#### Soil Zn content 2021-22 Analysis of Variance Table Response: Yield Df Sum Sq Mean Sq F value Pr(>F) 2 0.000605 0.000303 22.8783 3.873e-07 \*\*\* block 1 0.000004 0.000004 0.0045 0.9524560 V Ea 2 0.001751 0.000876 Zn 2 0.081815 0.040907 31.9127 0.0001539 \*\*\* 2 0.000008 0.000004 0.0031 0.9969109 v:Zn 8 0.010255 0.001282 Eb 3 0.000105 0.000035 2.6420 0.0640525 . H H:v 3 0.000012 0.000004 0.3000 0.8251341 6 0.000131 0.000022 1.6532 0.1610713 H:Zn H:v:Zn 6 0.000024 0.000004 0.3000 0.9327837 Ec 36 0.000476 0.000013 Signif. codes: 0 '\*\*\*' 0.001 '\*\*' 0.01 '\*' 0.05 '.' 0.1 ' ' 1 cv(a) = 6.1 %, cv(b) = 7.4 %, cv(c) = 0.7 %, Mean = 0.4867667

Plant height 30 DAS 2022-23 Analysis of Variance Table Response: Yield Df Sum Sq Mean Sq F value Pr(>F) block 1.25 0.62 0.4448 0.64445 2 1 523.26 523.26 81.9836 0.01198 \* V 6.38 Ea 2 12.77 2 10.05 5.03 1.3076 0.32259 Zn 2 1.54 0.77 0.1999 0.82282 v:Zn 8 30.75 3.84 Eb 3 1.65 0.55 0.3917 0.75966 Н 0.04 0.0294 0.99310 H:v 3 0.12 6 5.57 0.93 0.6619 0.68063 H:Zn H:v:Zn 6 2.81 0.47 0.3337 0.91465 Ec 36 50.48 1.40 \_ \_ \_ Signif. codes: 0 '\*\*\*' 0.001 '\*\*' 0.01 '\*' 0.05 '.' 0.1 ' ' 1 cv(a) = 18.3 %, cv(b) = 14.2 %, cv(c) = 8.6 %, Mean = 13.81306

	Plant height 60 DAS 2022-23								
Analys:	is (	of Varia	ance Tabi	le					
Respons	se:	Yield							
	Df	Sum Sq	Mean Sq	F value	Pr(>F)				
block	2	53.98	26.99	20.4199	1.183e-06	***			
V	1	453.36	453.36	69.0738	0.0141703	*			
Ea	2	13.13	6.56						
Zn	2	219.66	109.83	30.7833	0.0001749	***			
v:Zn	2	6.77	3.39	0.9489	0.4267697				
Eb	8	28.54	3.57						
				27.5766	1.935e-09	***			
H:v	3	1.52	0.51	0.3826	0.7661344				
H:Zn	6	6.87	1.15	0.8663	0.5289063				
H:v:Zn	6	2.27	0.38	0.2859	0.9398464				
of Rocky IV Second Colors			1.32						
Signif	. co	odes:	3 (***) (	0.001 (**	*' 0.01 '*	' 0.05'.' 0.1'' 1			
0									
cv(a) :	= 6	.4 %, c	v(b) = 4	.7 %, cv	(c) = 2.9	%, Mean = 39.86375			
	0700								

Plant height 90 DAS 2022-23 Analysis of Variance Table Response: Yield Df Sum Sq Mean Sq F value Pr(>F) block 2 444.44 222.22 99.0157 2.331e-15 \*\*\* 1 1155.92 1155.92 53.1067 0.018314 \* V Ea 43.53 21.77 2 Zn 2 338.12 169.06 16.8142 0.001364 \*\* 2.65 1.33 0.1320 0.878238 v:Zn 2 Eb 8 80.44 10.05 Н 3 226.30 75.43 33.6106 1.544e-10 \*\*\* 3 0.80 0.27 0.1185 0.948664 H:v H:Zn 6 7.10 1.18 0.5272 0.783818 H:v:Zn 6 11.37 1.90 0.8445 0.544287 Ec 36 80.80 2.24 \_ \_ \_ Signif. codes: 0 '\*\*\*' 0.001 '\*\*' 0.01 '\*' 0.05 '.' 0.1 ' ' 1 cv(a) = 7.8 %, cv(b) = 5.3 %, cv(c) = 2.5 %, Mean = 60.15347

	Plant height 120 DAS 2022-23								
Analys	Analysis of Variance Table								
Respon	se:	Yield							
	Df	Sum Sq	Mean Sq	F value	Pr(>F)				
block	2	465.8	232.9	80.4831	5.174e-14	***			
V	1	4976.0	4976.0	433.4122	0.002299	**			
Ea	2	23.0	11.5						
Zn	2	334.4	167.2	18.4438	0.001009	**			
v:Zn	2	1.8	0.9	0.0998	0.906124				
Eb	8	72.5	9.1						
Н	3	195.3	65.1	22.4976	2.224e-08	***			
H:v	3	1.1	0.4	0.1230	0.945933				
H:Zn	6	1.4	0.2	0.0792	0.997863				
H:v:Zn	6	1.8	0.3	0.1061	0.995209				
Ec	36	104.2	2.9						
Signif	. co	odes: (	) (***) (	0.001 <sup>(***</sup>	' 0.01'*'	0.05 '.' 0.1 ' ' 1			
cv(a)	= 4	%, cv(ł	o) = 3.6	%, cv(c)	= 2 %, Mea	an = 84.58944			

```
Plant height at harvest DAS 2022-23
Analysis of Variance Table
Response: Yield
      Df Sum Sq Mean Sq F value Pr(>F)
       2 1900.6 950.3 406.8617 < 2.2e-16 ***
block
       1 4755.7 4755.7 120.0992 0.008224 **
V
       2 79.2
                  39.6
Ea
Zn
       2 269.8
                  134.9 4.5557 0.047778 *
v:Zn
      2 1.6
                  0.8 0.0276 0.972830
       8 236.9 29.6
3 203.4 67.8 29.0325 1.017e-09 ***
Eb
Η
      30.90.30.13020.941493647.07.83.35170.009984**
H:v
H:Zn
H:v:Zn 6 24.6
                  4.1 1.7544 0.136626
Ec
      36 84.1
                   2.3
_ _ _
Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
cv(a) = 6.5 %, cv(b) = 5.6 %, cv(c) = 1.6 %, Mean = 96.79083
```

	Tillers 30 DAS 2022-23							
Analys	is o	of Varia	nce Table	e				
Respon	se:	Yield						
	Df	Sum Sq	Mean Sq	F value	Pr(>F)			
block	2	936.7	468.3	2.2614	0.1188			
V	1	2266.9	2266.9	4.5427	0.1667			
Ea	2	998.0	499.0					
Zn	2	10806.8	5403.4	42.1477	5.645e-05	***		
v:Zn	2	45.4	22.7	0.1772	0.8408			
Eb	8	1025.6	128.2					
Н	3	29.5	9.8	0.0475	0.9861			
H:v	3	2.0	0.7	0.0032	0.9997			
H:Zn	6	11.3	1.9	0.0091	1.0000			
H:v:Zn	6	16.0	2.7	0.0129	1.0000			
Ec	36	7455.7	207.1					
Signif	. co	odes: 0	·*** 0	.001 '**	0.01 '*'	0.05 '.' 0.1 ' ' 1		
cv(a)	= 9.	.6 %, cv	(b) = 4.9	9 %, cv(d	c) = 6.2 %	, Mean = 232.4722		
. /			. ,					

Tillers 60 DAS 2022-23 Analysis of Variance Table Response: Yield Df Sum Sq Mean Sq F value Pr(>F) block 554.4 277.2 2.1832 0.127380 2 1 27261.1 27261.1 39.1378 0.024611 \* V Ea 2 1393.1 696.5 Zn 2 17135.0 8567.5 21.8427 0.000574 \*\*\* 2 390.1 195.0 0.4973 0.625820 v:Zn 8 3137.9 392.2 Eb H 3 12134.2 4044.7 31.8575 3.107e-10 \*\*\* H:v 3 100.2 33.4 0.2629 0.851616 6 1241.0 206.8 1.6290 0.167502 H:Zn H:v:Zn 6 676.8 112.8 0.8885 0.513508 36 4570.7 127.0 Ec Signif. codes: 0 '\*\*\*' 0.001 '\*\*' 0.01 '\*' 0.05 '.' 0.1 ' ' 1 cv(a) = 5.4 %, cv(b) = 4.1 %, cv(c) = 2.3 %, Mean = 484.9028

```
Tillers 90 DAS 2022-23
Analysis of Variance Table
Response: Yield
      Df Sum Sq Mean Sq F value
                                  Pr(>F)
                 228.5 2.7578 0.0768464 .
block
          457.1
       2
       1 17681.8 17681.8 39.1697 0.0245921 *
V
Ea
       2
           902.8
                  451.4
       2 19655.8 9827.9 35.9127 0.0001009 ***
Zn
       2 465.1 232.6 0.8499 0.4627244
v:Zn
Eb
       8 2189.3 273.7
       3 9558.1 3186.0 38.4468 2.557e-11 ***
H
             7.3
H:v
      3
                    2.4 0.0295 0.9930640
      6 1064.7 177.4 2.1413 0.0722507 .
H:Zn
H:v:Zn 6 596.5 99.4 1.1997 0.3288027
      36 2983.3 82.9
Ec
_ _ _
Signif. codes: 0 (***' 0.001 (**' 0.01 (*' 0.05 (.' 0.1 (' 1
cv(a) = 5.1 %, cv(b) = 3.9 %, cv(c) = 2.2 %, Mean = 418.9656
```

Tillers 120 DAS 2022-23 Analysis of Variance Table Response: Yield Df Sum Sq Mean Sq F value Pr(>F) block 2 301.8 150.9 2.3621 0.10867 1 6523.8 6523.8 19.1939 0.04835 \* V Ea 2 679.8 339.9 2 17079.0 8539.5 40.4408 6.563e-05 \*\*\* Zn v:Zn 2 469.4 234.7 1.1114 0.37505 8 1689.3 211.2 Eb H 3 8284.2 2761.4 43.2317 5.068e-12 \*\*\* H:v 3 9.8 3.3 0.0511 0.98448 H:Zn 6 871.4 145.2 2.2736 0.05804 . H:v:Zn 6 484.7 80.8 1.2646 0.29790 36 2299.5 63.9 Ec \_ \_ \_ \_ Signif. codes: 0 (\*\*\*' 0.001 (\*\*' 0.01 (\*' 0.05 (.' 0.1 (' 1 cv(a) = 4.8 %, cv(b) = 3.8 %, cv(c) = 2.1 %, Mean = 386.8879

#### LAI 30 DAS 2022-23

Analysi	Analysis of Variance Table							
Respons	Yield							
	Sum Sq Mean Sq F value Pr(>F)							
block	0.001983 0.000991 0.5773 0.566500							
V	0.277156 0.277156 41.5840 0.023214 *							
Ea	0.013330 0.006665							
Zn	0.062237 0.031118 13.0138 0.003055 **							
v:Zn	0.001897 0.000948 0.3966 0.685145							
Eb	0.019129 0.002391							
Н	0.001320 0.000440 0.2563 0.856332							
H:v	0.003810 0.001270 0.7396 0.535403							
H:Zn	0.004852 0.000809 0.4709 0.825282							
H:v:Zn	0.007346 0.001224 0.7129 0.641468							
Ec	0.061823 0.001717							
Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1								
cv(a) =	6.6 %, cv(b) = 9.9 %, cv(c) = 8.4 %, Mean = 0.49	30785						

LAI 60 DAS 2022-23									
Analysi	Analysis of Variance Table								
Respons	se:	Yield							
	Df	Sum Sq	Mean Sq	F value	Pr(>F)				
block	2	0.00483	0.00242	0.1375	0.87199				
V	1	2.97559	2.97559	93.8861	0.01048	*			
Ea	2	0.06339	0.03169						
Zn	2	0.60646	0.30323	8.5990	0.01016	*			
v:Zn	2	0.00897	0.00448	0.1271	0.88236				
Eb	8	0.28211	0.03526						
Н	3	0.51194	0.17065	9.7073	7.836e-05	***			
H:v	3	0.00082	0.00027	0.0155	0.99733				
H:Zn	6	0.00531	0.00088	0.0503	0.99941				
H:v:Zn	6	0.06328	0.01055	0.6000	0.72838				
Ec	36	0.63285	0.01758						
Signif.		odes: 0	·***' 0	.001 '**	' 0.01 '*'	0.05 '.' 0.1 ' ' 1			
cv(a) =	= 6	.1 %, cv	(b) = 6.4	1 %, cv(d	c) = 4.5 %	, Mean = 2.941395			

#### LAI 90 DAS 2022-23

```
Analysis of Variance Table
Response: Yield
      Df Sum Sq Mean Sq F value
                                    Pr(>F)
       2 0.0276 0.0138
                          0.8173 0.4496472
block
       1 6.0293 6.0293 290.2292 0.0034278 **
V
Ea
       2 0.0415 0.0208
       2 1.9898 0.9949 23.3373 0.0004584 ***
Zn
v:Zn
       2 0.0007 0.0003
                        0.0079 0.9921564
       8 0.3410 0.0426
Eb
Н
       3 0.7079 0.2360 13.9837 3.346e-06 ***
H:v
       3 0.0038 0.0013 0.0759 0.9725849
       6 0.0581 0.0097
                          0.5740 0.7482988
H:Zn
H:v:Zn 6 0.0121 0.0020
                          0.1198 0.9933379
      36 0.6075 0.0169
EC
- - -
Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
cv(a) = 2.9 %, cv(b) = 4.1 %, cv(c) = 2.6 %, Mean = 4.996917
```

LAI 120 DAS 2022-23 Analysis of Variance Table Response: Yield Df Sum Sq Mean Sq F value Pr(>F) block 2 0.10272 0.05136 5.6587 0.007295 \*\* 1 1.12383 1.12383 23.2535 0.040415 \* V 2 0.09666 0.04833 Ea Zn 2 0.38152 0.19076 4.5856 0.047115 \* 2 0.00026 0.00013 0.0031 0.996933 v:Zn Eb 8 0.33280 0.04160 H 3 0.14175 0.04725 5.2055 0.004335 \*\* 3 0.00130 0.00043 0.0477 0.985968 H:v 6 0.01604 0.00267 0.2945 0.935608 H:Zn H:v:Zn 6 0.00279 0.00046 0.0512 0.999382 Ec 36 0.32676 0.00908 - - -Signif. codes: 0 (\*\*\*' 0.001 (\*\*' 0.01 (\*' 0.05 (.' 0.1 (' 1 cv(a) = 5.5 %, cv(b) = 5.1 %, cv(c) = 2.4 %, Mean = 4.026703

#### DMA 30 DAS 2022-23 Analysis of Variance Table Response: Yield Df Sum Sq Mean Sq F value Pr(>F) 82.86 41.43 1.2645 0.294621 block 2 V 1 18.30 18.30 1.3114 0.370687 2 27.91 13.95 Ea 2 1396.46 698.23 18.3098 0.001033 \*\* Zn 2 4.01 2.01 0.0526 0.949065 v:Zn Eb 8 305.08 38.13 0.76 0.25 0.0077 0.999052 3 Н 3 0.64 0.21 0.0065 0.999259 H:v 3.67 0.61 0.0187 0.999967 6 H:Zn H:v:Zn 6 10.18 1.70 0.0518 0.999359 36 1179.54 32.77 Ec \_ \_ \_ Signif. codes: 0 '\*\*\*' 0.001 '\*\*' 0.01 '\*' 0.05 '.' 0.1 ' ' 1 cv(a) = 7.4 %, cv(b) = 12.3 %, cv(c) = 11.4 %, Mean = 50.32917

DMA 60 DAS 2022-23 Analysis of Variance Table Response: Yield Df Sum Sq Mean Sq F value Pr(>F) block 300 150 1.4451 0.2490600 2 1 55217 55217 107.5988 0.0091662 \*\* V Ea 2 1026 513 8655 31.6605 0.0001583 \*\*\* Zn 2 17310 v:Zn 2 129 65 0.2367 0.7945989 Eb 8 2187 273 2440 23.5396 1.311e-08 \*\*\* 7321 Н 3 H:v 3 290 97 0.9325 0.4349977 H:Zn 6 1406 234 2.2606 0.0593104 . H:v:Zn 6 372 62 0.5986 0.7294114 Ec 36 3732 104 \_ \_ \_ Signif. codes: 0 '\*\*\*' 0.001 '\*\*' 0.01 '\*' 0.05 '.' 0.1 ' ' 1 cv(a) = 7.2 %, cv(b) = 5.3 %, cv(c) = 3.3 %, Mean = 312.7319

	DMA 90 DAS 2022-23							
Analys	Analysis of Variance Table							
Deenen		Viald						
Respon				_				
	Df	Sum Sq	Mean Sq	F value	Pr(>F)			
block	2	464	232	0.1171	0.889817			
V	1	401359	401359	45.4789	0.021289	*		
Ea	2	17650	8825					
Zn	2	44559	22279	13.2154	0.002915	**		
v:Zn	2	153	76	0.0453	0.955925			
Eb	8	13487	1686					
Н	3	25928	8643	4.3589	0.010189	*		
H:v	3	2151	717	0.3617	0.781032			
H:Zn	6	1678	280	0.1410	0.989693			
H:v:Zn	6	707	118	0.0594	0.999052			
Ec	36	71379	1983					
Signif	Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1							
cv(a)	= 1	5.4 %, (	cv(b) = 6	5.7 %, cv	v(c) = 7.3	3 %, Mean = 609.2899		

DMA 120 DAS 2022-23 Analysis of Variance Table Response: Yield Df Sum Sq Mean Sq F value Pr(>F) block 0.3267 0.723388 2 1274 637 1 408995 408995 245.7296 0.004045 \*\* V Ea 2 3329 1664 Zn 2 121826 60913 25.1824 0.000353 \*\*\* v:Zn 446 223 0.0923 0.912824 2 Eb 8 19351 2419 3 72310 24103 12.3638 1.038e-05 \*\*\* Н 3 7319 2440 1.2515 0.305537 H:v H:Zn 6 4761 794 0.4070 0.869448 H:v:Zn 6 2107 351 0.1801 0.980534 Ec 36 70182 1950 \_ \_ \_ Signif. codes: 0 '\*\*\*' 0.001 '\*\*' 0.01 '\*' 0.05 '.' 0.1 ' ' 1 cv(a) = 4.1 %, cv(b) = 5 %, cv(c) = 4.5 %, Mean = 991.9417

	Effective tillers 2022-23							
Analys	Analysis of Variance Table							
Respon	Response: Yield							
	Df	Sum Sq	Mean Sq	F value	Pr(>F)			
block	2	778.4	389.2	5.3401	0.009312	**		
V	1	2582.2	2582.2	37.3175	0.025766	*		
Ea	2	138.4	69.2					
Zn	2	17679.4	8839.7	183.8458	2.056e-07	***		
v:Zn	2	0.0	0.0	0.0004	0.999563			
Eb	8	384.7	48.1					
Н	3	15998.0	5332.7	73.1643	2.220e-15	***		
H:v	3	62.1	20.7	0.2839	0.836700			
H:Zn	6	561.4	93.6	1.2838	0.289291			
H:v:Zn	6	317.3	52.9	0.7255	0.631909			
Ec	36	2623.9	72.9					
Signif	. co	odes: 0	(***) Ø.	.001 '**'	0.01 (*)	0.05'.'0.1''1		
cv(a)	cv(a) = 2.5 %, cv(b) = 2.1 %, cv(c) = 2.6 %, Mean = 334.2692							

Spike length 2022-23 Analysis of Variance Table Response: Yield Df Sum Sq Mean Sq F value Pr(>F) 2 5.9003 2.9502 8.7619 0.0007936 \*\*\* block 1 17.3357 17.3357 25.2190 0.0374400 \* V Ea 2 1.3748 0.6874 2 5.7434 2.8717 7.0655 0.0170746 \* Zn 2 0.2084 0.1042 0.2564 0.7799549 v:Zn Eb 8 3.2515 0.4064 Н 3 3.3041 1.1014 3.2710 0.0321524 \* 3 0.0481 0.0160 0.0476 0.9860227 H:v H:Zn 6 0.4488 0.0748 0.2221 0.9670686 H:v:Zn 6 0.2533 0.0422 0.1254 0.9924641 36 12.1214 0.3367 Ec ---Signif. codes: 0 (\*\*\*' 0.001 (\*\*' 0.01 (\*' 0.05 (.' 0.1 (' 1 cv(a) = 7.6 %, cv(b) = 5.8 %, cv(c) = 5.3 %, Mean = 10.96587

#### Number of grains per spike 2022-23 Analysis of Variance Table Response: Yield Df Sum Sq Mean Sq F value Pr(>F)block 2 23.71 11.86 0.8699 0.427619 1 542.77 542.77 174.7553 0.005674 \*\* V Ea 2 6.21 3.11 2 447.26 223.63 16.9058 0.001340 \*\* Zn v:Zn 2 3.41 1.71 0.1291 0.880703 8 105.82 13.23 Eb 3 247.79 82.60 6.0597 0.001894 \*\* Н H:v 3 4.51 1.50 0.1103 0.953526 6 23.67 3.94 0.2894 0.938123 H:Zn H:v:Zn 6 10.17 1.69 0.1243 0.992638 36 490.69 Ec 13.63 \_ \_ \_ Signif. codes: 0 (\*\*\*' 0.001 (\*\*' 0.01 (\*' 0.05 (.' 0.1 (') 1 cv(a) = 3.5 %, cv(b) = 7.3 %, cv(c) = 7.4 %, Mean = 49.81896

1000 – grain weight 2022-23 Analysis of Variance Table Response: Yield Df Sum Sq Mean Sq F value Pr(>F) 8.3779 0.001029 \*\* block 2 4.316 2.158 1 51.140 51.140 22.6677 0.041396 \* V Ea 2 4.512 2.256 2 151.106 75.553 91.2058 3.116e-06 \*\*\* Zn 2 0.251 0.125 0.1515 0.861858 v:Zn Eb 8 6.627 0.828 3 139.871 46.624 181.0157 < 2.2e-16 \*\*\* Н 3 0.143 0.048 H:v 0.1846 0.906142 6 5.873 0.979 3.8003 0.004920 \*\* H:Zn H:v:Zn 6 0.190 0.032 0.1231 0.992827 Ec 36 9.272 0.258 - - -Signif. codes: 0 (\*\*\*' 0.001 (\*\*' 0.01 (\*' 0.05 (.' 0.1 (' 1 cv(a) = 3.5 %, cv(b) = 2.1 %, cv(c) = 1.2 %, Mean = 42.85306

```
Grain yield 2022-23
Analysis of Variance Table
Response: Yield
      Df Sum Sq Mean Sq F value
                                   Pr(>F)
       2 0.50
                  0.25
                         2.7765 0.0756131 .
block
       1 794.68 794.68 6029.1507 0.0001658 ***
V
Ea
       2
           0.26
                 0.13
       2 156.91 78.45 608.6940 1.817e-09 ***
Zn
       2 9.20 4.60 35.7069 0.0001030 ***
v:Zn
Eb
       8
           1.03
                 0.13
       3 150.74 50.25 561.4692 < 2.2e-16 ***
H
       3 0.53 0.18
                        1.9659 0.1365278
H:v
      6 8.11
                 1.35 15.1123 1.484e-08 ***
H:Zn
H:v:Zn 6 1.32
                 0.22 2.4625 0.0424700 *
Ec
      36 3.22
                 0.09
- - -
Signif. codes: 0 (***' 0.001 (**' 0.01 (*' 0.05 (.' 0.1 (' 1
cv(a) = 0.7 \%, cv(b) = 0.7 \%, cv(c) = 0.6 \%, Mean = 49.57222
```

Straw yield 2022-23 Analysis of Variance Table Response: Yield Df Sum Sq Mean Sq F value Pr(>F) block 2 2.92 1.46 3.8677 0.0300912 \* 1 1769.23 1769.23 7708.6547 0.0001297 \*\*\* V Ea 2 0.46 0.23 2 317.21 158.60 957.6251 2.994e-10 \*\*\* Zn v:Zn 2 24.06 12.03 72.6368 7.421e-06 \*\*\* Eb 8 1.32 0.17 3 344.77 114.92 304.6926 < 2.2e-16 \*\*\* Н 0.27 0.7115 0.5515246 H:v 3 0.81 8.4650 9.262e-06 \*\*\* H:Zn 6 19.16 3.19 0.47 1.2428 0.3080340 H:v:Zn 6 2.81 Ec 36 13.58 0.38 \_ \_ \_ Signif. codes: 0 (\*\*\*' 0.001 (\*\*' 0.01 (\*' 0.05 (.' 0.1 (' 1 cv(a) = 0.6 %, cv(b) = 0.5 %, cv(c) = 0.8 %, Mean = 74.25931

```
Harvest index 2022-23
Analysis of Variance Table
Response: Yield
      Df Sum Sq Mean Sq F value Pr(>F)
                                 0.02433 *
block
       2 0.275 0.1375
                       4.1276
       1 0.003 0.0031
                        0.5398
                                 0.53897
V
Ea
       2 0.011 0.0057
       2 0.115 0.0577 0.9679
                                 0.42030
Zn
       2 0.019 0.0093 0.1555
v:Zn
                                 0.85853
Eb
       8 0.477 0.0597
H
       3 63.909 21.3029 639.3871 < 2.2e-16 ***
H:v
       3 0.404 0.1346
                        4.0402
                                 0.01418 *
       6 4.478 0.7464 22.4026 8.534e-11 ***
H:Zn
H:v:Zn 6 0.309 0.0514 1.5442 0.19204
      36 1.199 0.0333
Ec
_ _ _
Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
cv(a) = 0.2 %, cv(b) = 0.6 %, cv(c) = 0.5 %, Mean = 40.03504
```

```
Grain protein content 2022-23
Analysis of Variance Table
Response: Yield
      Df Sum Sq Mean Sq F value Pr(>F)
block
       2 3.0120 1.5060
                         4.1403 0.024078 *
       1 22.2222 22.2222 168.8334 0.005871 **
V
Ea
       2 0.2632 0.1316
       2 15.0962 7.5481 9.5779 0.007532 **
Zn
v:Zn
       2 0.0039 0.0019 0.0024 0.997559
Eb
       8 6.3046 0.7881
Н
       3 3.8072 1.2691 3.4888 0.025429 *
H:v
      3 0.1272 0.0424 0.1165 0.949833
H:Zn
      6 0.3757 0.0626 0.1722 0.982657
H:v:Zn 6 0.3555 0.0593 0.1629 0.984965
      36 13.0950 0.3637
Ec
- - -
Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
cv(a) = 3.1 \%, cv(b) = 7.7 \%, cv(c) = 5.2 \%, Mean = 11.53583
```

```
Grain phytic acid content 2022-23
Analysis of Variance Table
Response: Yield
       Df Sum Sq Mean Sq F value
                                    Pr(>F)
       2 0.26616 0.13308 9.6228 0.0004488 ***
block
       1 0.03736 0.03736 0.2262 0.6812462
V
Ea
       2 0.33030 0.16515
       2 1.16581 0.58290 22.4692 0.0005215 ***
Zn
v:Zn
       2 0.00679 0.00339 0.1308 0.8792383
Eb
       8 0.20754 0.02594
       3 0.47174 0.15725 11.3704 2.155e-05 ***
Н
H:v
       3 0.00208 0.00069 0.0501 0.9849405
H:Zn
       6 0.00068 0.00011 0.0082 0.9999972
H:v:Zn 6 0.00288 0.00048 0.0347 0.9997983
       36 0.49787 0.01383
Ec
- - -
Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
cv(a) = 42.3 %, cv(b) = 16.7 %, cv(c) = 12.2 %, Mean = 0.9616667
```

Grain Zn content 2022-23 Analysis of Variance Table Response: Yield Df Sum Sq Mean Sq F value Pr(>F) block 2 0.65 0.32 1.2170 0.3079996 V 1 441.79 441.79 3457.4323 0.0002891 \*\*\* Ea 2 0.26 0.13 Zn 2 145.80 72.90 387.5418 1.089e-08 \*\*\* 2 0.05 0.02 v:Zn 0.1309 0.8791512 0.19 Eb 8 1.50 3 51.87 17.29 64.9983 1.332e-14 \*\*\* H 0.09 0.3528 0.7873299 H:v 3 0.28 H:Zn 6 4.26 0.71 2.6682 0.0302574 \* H:v:Zn 6 0.89 0.15 0.5568 0.7614801 Ec 36 9.58 0.27 \_ \_ \_ Signif. codes: 0 '\*\*\*' 0.001 '\*\*' 0.01 '\*' 0.05 '.' 0.1 ' ' 1 cv(a) = 0.9 %, cv(b) = 1.1 %, cv(c) = 1.3 %, Mean = 38.23681

```
Stem Zn content 2022-23
Analysis of Variance Table
Response: Yield
      Df Sum Sq Mean Sq F value Pr(>F)
           5.62
                  2.81 4.7637 0.014607 *
block
       2
       1 321.90 321.90 419.7427 0.002374 **
V
Ea
       2
          1.53
                  0.77
       2 208.88 104.44 256.2610 5.580e-08 ***
Zn
       2 0.58 0.29 0.7104 0.520016
v:Zn
       8 3.26
Eb
                 0.41
       3 41.66 13.89 23.5302 1.317e-08 ***
Н
         1.11 0.37 0.6277 0.601815
H:v
       3
H:Zn
      6 7.14
                 1.19 2.0154 0.088972 .
H:v:Zn 6 1.33
                 0.22 0.3752 0.889890
Ec
      36 21.25 0.59
_ _ _
Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1
cv(a) = 3.6 %, cv(b) = 2.6 %, cv(c) = 3.1 %, Mean = 24.63083
```

Leaf Zn content 2022-23 Analysis of Variance Table Response: Yield Df Sum Sq Mean Sq F value Pr(>F) 2 0.080 0.040 block 0.5026 0.609161 1 59.536 59.536 732.4006 0.001363 \*\* V Ea 2 0.163 0.081 2 22.007 11.003 75.2058 6.504e-06 \*\*\* Zn v:Zn 2 0.091 0.046 0.3125 0.740146 Eb 8 1.170 0.146 H 3 6.588 2.196 27.7086 1.824e-09 \*\*\* H:v 3 0.189 0.063 0.7963 0.504025 6 1.074 0.179 2.2576 0.059599 . H:Zn H:v:Zn 6 0.206 0.034 0.4341 0.851173 36 2.853 0.079 Ec \_ \_ \_ Signif. codes: 0 '\*\*\*' 0.001 '\*\*' 0.01 '\*' 0.05 '.' 0.1 ' ' 1 cv(a) = 3 %, cv(b) = 4 %, cv(c) = 2.9 %, Mean = 9.591833

	Soil Zn content 2022-23							
Analys	Analysis of Variance Table							
Respon	se: Yield							
	Df Sum Sq Mean Sq F value Pr(>F)							
block	2 0.000137 0.000069 3.3833 0.04504 *							
V	1 0.000007 0.000007 0.0120 0.92284							
Ea	2 0.001095 0.000548							
Zn	2 0.260403 0.130202 2334.1115 8.566e-12 ***							
v:Zn	2 0.000013 0.000007 0.1176 0.89055							
Eb	8 0.000446 0.000056							
Н	3 0.000048 0.000016 0.7927 0.50596							
H:v	3 0.000015 0.000005 0.2533 0.85843							
H:Zn	6 0.000107 0.000018 0.8765 0.52175							
H:v:Zn	6 0.000031 0.000005 0.2533 0.95472							
Ec	36 0.000731 0.000020							
Signif	. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 '' 1							
cv(a)	= 4.7 %, cv(b) = 1.5 %, cv(c) = 0.9 %, Mean = 0.5016692							
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S.no	Operation		Quantity/duration	Cost per quantity/hour	Total
1	Land preparation	Tractor cost	3 hr	500	1500
2	Layout preparation		5 labours	400 per day	2000
3	Seed		100 kg/ha	3775	3775
4	Sowing and fertilizer application		10 labours	400 per day	4000
5	Fertilizer				
	Nitrogen	Urea	225 kg/ha	268 per 50 kg/bag	1206
	Phosphorus	DAP	137.5 kg/ha	1350 per 50 kg/bag	3713
	Potassium	МОР	30 kg/ha	872 per 50 kg/bag	524
6	Labour for split dose		3 splits x 2 Labours per split (6)	400 per day	2400
7	Intercultural operations				
	Hand weeding		6 Labours	400 per day	2400
	Spraying	1 pre- emergence herbicide	1 Labour	400 per day	400
		1 post- emergence herbicide	1 Labour	400 per day	400
		1 plant protection chemicals	1 Labour	400 per day	400
8	Herbicides	Pre- emergence herbicide	Pendimethalin (3.75 litre/ha)	350 rupees per litre	1312
		Post- emergence	Total (40 g/ha)	500 rupees per	1250

# APPENDIX IV – COST OF CULTIVATION (FIXED COST) 2021-22 SEASON

		herbicide		16 g packet	
	Plant protection chemicals	Insecticide	Thiamethoxam (50 g/ha)	3000 rupees per 1 kg	150
9	Irrigation for cropping season		5 months	1000 per month	5000
10	Harvesting and shelling		10 labours x 2 days	400 per day	8000
11	Land lease and miscellaneous for cropping season				2000
12	Total				40430

# COST OF CULTIVATION (VARIABLE COST)

S. no	Treatment combination	Zn	Cytokinin	GA	Total cost
1	V725 Zn0 H0	0	0	0	0
2	V725 Zn0 H1	0	0	315	315
3	V725 Zn0 H2	0	350	0	350
4	V725 Zn0 H3	0	175	158	333
5	V725 Zn62.5 H0	6325	0	0	6325
6	V725 Zn62.5 H1	6325	0	315	6640
7	V725 Zn62.5 H2	6325	350	0	6675
8	V725 Zn62.5 H3	6325	175	158	6658
9	V725 Zn31.25,F H0	3846	0	0	3846
10	V725 Zn31.25,F H1	3846	0	315	4161
11	V725 Zn31.25,F H2	3846	350	0	4196
12	V725 Zn31.25,F H3	3846	175	158	4179
13	V752 Zn0 H0	0	0	0	0
14	V752 Zn0 H1	0	0	315	315
15	V752 Zn0 H2	0	350	0	350
16	V752 Zn0 H3	0	175	158	333
17	V752 Zn62.5 H0	6325	0	0	6325
18	V752 Zn62.5 H1	6325	0	315	6640
19	V752 Zn62.5 H2	6325	350	0	6675
20	V752 Zn62.5 H3	6325	175	158	6658
21	V752 Zn31.25,F H0	3846	0	0	3846
22	V752 Zn31.25,F H1	3846	0	315	4161
23	V752 Zn31.25,F H2	3846	350	0	4196
24	V752 Zn31.25,F H3	3846	175	158	4179

				Total
S. no	Treatment combination	Fixed cost	Variable cost	cost
1	V <sub>725</sub> Zn <sub>0</sub> H <sub>0</sub>	40430	0	40430
2	$V_{725} Zn_0 H_1$	40430	315	40745
3	V <sub>725</sub> Zn <sub>0</sub> H <sub>1</sub>	40430	350	40780
4	V <sub>725</sub> Zn <sub>0</sub> H <sub>2</sub>	40430	333	40763
5	V <sub>725</sub> Zn <sub>62.5</sub> H <sub>0</sub>	40430	6325	46755
6	$V_{725} Zn_{62.5} H_1$	40430	6640	47070
7	V <sub>725</sub> Zn <sub>62.5</sub> H <sub>2</sub>	40430	6675	47105
8	V725 Zn62.5 H3	40430	6658	47088
9	V725 Zn31.25,F H0	40430	3846	44276
10	V725 Zn31.25,F H1	40430	4161	44591
11	V725 Zn31.25,F H2	40430	4196	44626
12	V <sub>725</sub> Zn <sub>31.25,F</sub> H <sub>3</sub>	40430	4179	44609
13	V752 Zn0 H0	40430	0	40430
14	V <sub>752</sub> Zn <sub>0</sub> H <sub>1</sub>	40430	315	40745
15	V752 Zn0 H2	40430	350	40780
16	V752 Zn0 H3	40430	333	40763
17	V752 Zn62.5 H0	40430	6325	46755
18	V <sub>752</sub> Zn <sub>62.5</sub> H <sub>1</sub>	40430	6640	47070
19	V752 Zn62.5 H2	40430	6675	47105
20	V <sub>752</sub> Zn <sub>62.5</sub> H <sub>3</sub>	40430	6658	47088
21	V752 Zn31.25,F H0	40430	3846	44276
22	V752 Zn31.25,F H1	40430	4161	44591
23	V <sub>752</sub> Zn <sub>31.25,F</sub> H <sub>2</sub>	40430	4196	44626
24	V752 Zn31.25,F H3	40430	4179	44609

### TOTAL COST (FIXED + VARIABLE)

# Gross return = wheat yield $\times$ MSP of wheat (1975 rupees in 2021-22)

## **BENEFIT – COST RATIO**

		2021-22 sea	son		
S. no	Treatment combination	Cost of cultivation	Gross return	Net return	B:C ratio
1	V725 Zn0 H0	40430	83608	43178	2.07
2	V725 Zn0 H1	40745	85518	44773	2.10
3	V725 Zn0 H2	40780	88283	47503	2.16
4	V <sub>725</sub> Zn <sub>0</sub> H <sub>3</sub>	40763	86835	46072	2.13
5	V725 Zn62.5 H0	46755	86242	39487	1.84
6	V <sub>725</sub> Zn <sub>62.5</sub> H <sub>1</sub>	47070	90192	43122	1.92
7	V <sub>725</sub> Zn <sub>62.5</sub> H <sub>2</sub>	47105	92825	45720	1.97
8	V <sub>725</sub> Zn <sub>62.5</sub> H <sub>3</sub>	47088	91508	44420	1.94
9	V725 Zn31.25,F H0	44276	88875	44599	2.01
10	V <sub>725</sub> Zn <sub>31.25,F</sub> H <sub>1</sub>	44591	92167	47576	2.07
11	V <sub>725</sub> Zn <sub>31.25,F</sub> H <sub>2</sub>	44626	97433	52807	2.18
12	V <sub>725</sub> Zn <sub>31.25,F</sub> H <sub>3</sub>	44609	94800	50191	2.13
13	V <sub>752</sub> Zn <sub>0</sub> H <sub>0</sub>	40430	73733	33303	1.82
14	V <sub>752</sub> Zn <sub>0</sub> H <sub>1</sub>	40745	76367	35622	1.87
15	V752 Zn0 H2	40780	79988	39208	1.96
16	V <sub>752</sub> Zn <sub>0</sub> H <sub>3</sub>	40763	77815	37052	1.91
17	V752 Zn62.5 H0	46755	77025	30270	1.65
18	V752 Zn62.5 H1	47070	81963	34893	1.74
19	V752 Zn62.5 H2	47105	84925	37820	1.80
20	V752 Zn62.5 H3	47088	83280	36192	1.77
21	V752 Zn31.25,F H0	44276	81633	37357	1.84
22	V752 Zn31.25,F H1	44591	86242	41651	1.93
23	V752 Zn31.25,F H2	44626	90850	46224	2.04
24	V752 Zn31.25,F H3	44609	87769	43160	1.97

S.no	Operation		Quantity/duration	Cost per quantity/hour	Total
1	Land preparation	Tractor cost	3 hr	550	1650
2	Layout preparation		5 labours	400 per day	2000
3	Seed		100 kg/ha	3775	3775
4	Sowing and fertilizer application		10 labours	400 per day	4000
5	Fertilizer				
	Nitrogen	Urea	225 kg/ha	268 per 50 kg/bag	1206
	Phosphorus	DAP	137.5 kg/ha	1350 per 50 kg/bag	3713
	Potassium	МОР	30 kg/ha	872 per 50 kg/bag	524
6	Labour for split dose		3 splits x 2 Labours per split (6)	400 per day	2400
7	Intercultural operations				
	Hand weeding		6 Labours	400 per day	2400
	Spraying	1 pre- emergence herbicide	1 Labour	400 per day	400
		1 post- emergence herbicide	1 Labour	400 per day	400
		1 plant protection chemicals	1 Labour	400 per day	400
8	Herbicides	Pre- emergence herbicide	Pendimethalin (3.75 litre/ha)	350 rupees per litre	1312
		Post- emergence	Total (40 g/ha)	500 rupees per	1250

# APPENDIX V – COST OF CULTIVATION (FIXED COST) 2022-23 SEASON

		herbicide		16 g packet	
	Plant protection chemicals	Insecticide	Thiamethoxam (50 g/ha)	3000 rupees per 1 kg	150
9	Irrigation for cropping season		5 months	1000 per month	5000
10	Harvesting and shelling		10 labours x 2 days	400 per day	8000
11	Land lease and miscellaneous for cropping season				2500
12	Total				41080

# COST OF CULTIVATION (VARIABLE COST)

S. no	Treatment combination	Zn	Cytokinin	GA	Total cost
1	V725 Zn0 H0	0	0	0	0
2	V725 Zn0 H1	0	0	315	315
3	V725 Zn0 H2	0	350	0	350
4	V725 Zn0 H3	0	175	158	333
5	V725 Zn62.5 H0	6325	0	0	6325
6	V725 Zn62.5 H1	6325	0	315	6640
7	V725 Zn62.5 H2	6325	350	0	6675
8	V725 Zn62.5 H3	6325	175	158	6658
9	V725 Zn31.25,F H0	3846	0	0	3846
10	V725 Zn31.25,F H1	3846	0	315	4161
11	V725 Zn31.25,F H2	3846	350	0	4196
12	V725 Zn31.25,F H3	3846	175	158	4179
13	V752 Zn0 H0	0	0	0	0
14	V752 Zn0 H1	0	0	315	315
15	V752 Zn0 H2	0	350	0	350
16	V752 Zn0 H3	0	175	158	333
17	V752 Zn62.5 H0	6325	0	0	6325
18	V752 Zn62.5 H1	6325	0	315	6640
19	V752 Zn62.5 H2	6325	350	0	6675
20	V752 Zn62.5 H3	6325	175	158	6658
21	V752 Zn31.25,F H0	3846	0	0	3846
22	V752 Zn31.25,F H1	3846	0	315	4161
23	V752 Zn31.25,F H2	3846	350	0	4196
24	V752 Zn31.25,F H3	3846	175	158	4179

				Total
S. no	Treatment combination	Fixed cost	Variable cost	cost
1	V725 Zn <sub>0</sub> H <sub>0</sub>	41080	0	41080
2	V <sub>725</sub> Zn <sub>0</sub> H <sub>1</sub>	41080	315	41395
3	V725 Zn0 H2	41080	350	41430
4	V725 Zn0 H3	41080	333	41413
5	V725 Zn62.5 H0	41080	6325	47405
6	V <sub>725</sub> Zn <sub>62.5</sub> H <sub>1</sub>	41080	6640	47720
7	V <sub>725</sub> Zn <sub>62.5</sub> H <sub>2</sub>	41080	6675	47755
8	V725 Zn62.5 H3	41080	6658	47738
9	V725 Zn31.25,F H0	41080	3846	44926
10	V725 Zn31.25,F H1	41080	4161	45241
11	V <sub>725</sub> Zn <sub>31.25,F</sub> H <sub>2</sub>	41080	4196	45276
12	V <sub>725</sub> Zn <sub>31.25,F</sub> H <sub>3</sub>	41080	4179	45259
13	V752 Zn0 H0	41080	0	41080
14	$V_{752} Zn_0 H_1$	41080	315	41395
15	$V_{752} Zn_0 H_2$	41080	350	41430
16	V752 Zn0 H3	41080	333	41413
17	V752 Zn62.5 H0	41080	6325	47405
18	V752 Zn62.5 H1	41080	6640	47720
19	V752 Zn62.5 H2	41080	6675	47755
20	V752 Zn62.5 H3	41080	6658	47738
21	V752 Zn31.25,F H0	41080	3846	44926
22	V752 Zn31.25,F H1	41080	4161	45241
23	V <sub>752</sub> Zn <sub>31.25,F</sub> H <sub>2</sub>	41080	4196	45276
24	V752 Zn31.25,F H3	41080	4179	45259

### TOTAL COST (FIXED + VARIABLE)

# Gross return = wheat yield $\times$ MSP of wheat (2015 rupees in 2022-23)

## **BENEFIT – COST RATIO**

2022-23 season								
S. no	Treatment combination	Cost of cultivation	Gross return	Net return	B:C ratio			
1	V725 Zn0 H0	41080	98601	57521	2.40			
2	V725 Zn0 H1	41080	100683	59603	2.45			
3	V725 Zn0 H2	41080	104982	63902	2.56			
4	V725 Zn0 H3	41080	102899	61819	2.50			
5	V725 Zn62.5 H0	41080	103974	62894	2.53			
6	V725 Zn62.5 H1	41080	106191	65111	2.58			
7	V <sub>725</sub> Zn <sub>62.5</sub> H <sub>2</sub>	41080	110893	69813	2.70			
8	V725 Zn62.5 H3	41080	108475	67395	2.64			
9	V725 Zn31.25,F H0	41080	106728	65648	2.60			
10	V725 Zn31.25,F H1	41080	108340	67260	2.64			
11	V725 Zn31.25,F H2	41080	115997	74917	2.82			
12	V725 Zn31.25,F H3	41080	111228	70148	2.71			
13	V752 Zn0 H0	41080	87787	46707	2.14			
14	V752 Zn0 H1	41080	89600	48520	2.18			
15	$V_{752} Zn_0 H_2$	41080	93093	52013	2.27			
16	V752 Zn0 H3	41080	91280	50200	2.22			
17	V752 Zn62.5 H0	41080	88929	47849	2.16			
18	V <sub>752</sub> Zn <sub>62.5</sub> H <sub>1</sub>	41080	91212	50132	2.22			
19	V752 Zn62.5 H2	41080	96989	55909	2.36			
20	V752 Zn62.5 H3	41080	94638	53558	2.30			
21	V752 Zn31.25,F H0	41080	91212	50132	2.22			
22	V <sub>752</sub> Zn <sub>31.25,F</sub> H <sub>1</sub>	41080	93765	52685	2.28			
23	V <sub>752</sub> Zn <sub>31.25,F</sub> H <sub>2</sub>	41080	101690	60610	2.48			
24	V752 Zn31.25,F H3	41080	98131	57051	2.39			