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# Influence of Foliar Spray with Different Doses of Zinc (ZnSO<sub>4</sub>) at Various Crop Stages on Seed Quality Parameters of Harvested Seeds

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# Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

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# **ABSTRACT**

A laboratory experiment was conducted to analyze the influence of foliar spray and stages of zinc application on seed quality parameters of harvested radish seeds cv. Japanese White in laboratory of the Department of Seed Science and Technology, Dr Yashwant Singh Parmar University of Horticulture and Forestry, Nauni, Solan (HP). There were 12 treatment combinations as 0g/l zinc at

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bolting stage, 0g/l bolting +14 days after first application, 0g/l at bolting +14days +28 days after first application 1g/l zinc (ZnSO<sub>4</sub>) at bolting stage, 1g/l bolting +14 days after first application, 1g/l at bolting +14days +28 days after first application, 2g/l at bolting, 2g/l at bolting +14 days after first application, 2g/l at bolting +14 days after first application, 3g/l at bolting +14 days after first application, 3g/l at bolting +14days +28 days after first application. The experiment was laid out in CRD (completely randomized design) as well as RCBD(Randomized complete block design) design Among treatment combinations Zn<sub>2</sub>S<sub>1</sub> (2g l<sup>-1</sup> at the time of bolting) was found superior in every aspect of quality attributes i.e. 1000 seed weight (14.36g), germination of harvested seeds (85.75%), seedling length (24.20cm), seedling dry weight (11.31mg), seedling vigour index-I (2,075.08), seedling vigour index-II (966.77), electrical conductivity (0.017 dSm<sup>-1</sup>), accelerated ageing test (78.25%) and cold test (81.00%).

Keywords: Radish; micronutrients; zinc; seed quality parameter.

# 1. INTRODUCTION

Radish is a cruciferae family root crop that is grown in both tropical and temperate regions of the world. It is native to Europe and Asia. Radish seed output per unit area is low, however, it could be increased with better management foliar practices such as micronutrient supplementation. Micronutrients are essential for germination and seedling growth, from cell wall development to respiration, photosynthesis and chlorophyll generation. The administration of micronutrients accelerates the uptake of all plant nutrients while simultaneously generating a defence mechanism against insect - pests and enhancing diseases. hence growth productivity [1]. Micronutrients are required in minimal dose, even then lack of these might hinder growth and production. Foliar spray is the most recent type of crop nutrition, consisting of micronutrients given to leaves in liquid form. Foliar spray is 6 to 20 times more effective than soil application [2]. Micronutrients such as zinc are critical for plant growth, development and biomass production because they are required for the creation of Indole Acetic Acid (IAA). It also aids in energy production, protein and growth regulator synthesis [3], pollen development, sexual fertilization and germination [4], greater seed yield, and fruit number. It also improves growth, hormone biosynthesis, starch creation, seed production and maturation [5]. Zinc also improves the viability and vigour of seedlings. It is a necessary component of ribosomes and is critical to their structural integrity [6]. Zinc plays numerous minor roles in plant growth, and a consistent and continuous supply is essential for optimal growth and output [7]. The Zn ion is the form of zinc that is available to plants. Because of its adsorption with soil particles and less interaction with crop roots, foliar application of Zn <sup>2+</sup> foliar spray is more effective than soil treatment [8].

# 2. MATERIALS AND METHODS

To study the effect of foliar application of zinc on quality of harvested seeds of radish, seed analysis was done at laboratory of the Department of Seed Science and Technology, Dr Yashwant Singh Parmar University Horticulture and Forestry, Nauni, Solan (HP). After completion of field trial harvested seeds of radish cv. Japanese White were evaluated in laboratory for different seed quality parameters. In field trial, 12 different treatmentswere laid out by combining 4 different doses of zinc (ZnSO<sub>4</sub>) viz. Zn<sub>1</sub>: 0gl<sup>-1</sup>, Zn<sub>2</sub>:1gl<sup>-1</sup>,Zn<sub>3</sub>: 2gl<sup>-1</sup> and Zn<sub>4</sub>: 3gl<sup>-1</sup> along with 3 different stages of application S<sub>1</sub>: Bolting, S<sub>2</sub>: Bolting + 14 days after first treatment, S<sub>3</sub>: Bolting + 14 days after first application + 28 days after first application. Observations under laboratory conditions were recorded for 1000 seed weight, germination per cent, seedling length, seedling dry weight, seed vigourindex-I, Seed vigour index-II, electrical conductivity, accelerated ageing test and cold test as per procedures. Experiments standard conducted with 4 replications [9] and results were statistically analyzed by using completely randomized block design [10] following windowbased computer application OPSTAT developed by Sheoran [11] and post hoc test Dunken Multiple range Test (DMRT).

# 3. RESULTS AND DISCUSSION

# 3.1 1000 Seed Weight

Data (Table 1) depicted that zinc levels, stages of foliar spray and their combinations had a significant influence on 1000 seed weight. Among the levels of zinc, 1000 seed weight was uncurvingly maximum (13.55 g) up to 2g I<sup>-1</sup> (Zn<sub>3</sub>) and thereafter a significant reduction was observed when zinc was applied @ 3g I<sup>-1</sup>and minimum 1000 seed weight (12.28 g) was noticed in Zn<sub>1</sub> (Water spray). As far as stages of

application were concerned, the highest 1000 seed weight (13.20 g) was observed with foliar application of zinc at S<sub>1</sub> (Bolting stage) and after that there was a linear decrease and lowest (12.99 g) 1000 seed weight was observed at S<sub>3</sub> (Bolting + 14 days + 28 days after first application). However, treatment combinations Zn<sub>3</sub>S<sub>1</sub> (2g l<sup>-1</sup> foliar spray of zinc applied at bolting stage) produced maximum 1000 seed weight (14.36 g) among all interactions and minimum (12.28 g) was observed in treatment combination Zn<sub>1</sub>S<sub>2</sub> (Water spray at bolting + 14 days after first application) and Zn<sub>1</sub>S<sub>3</sub> (Water spray at bolting + 14 days + 28 days after first application) was at par with Zn<sub>1</sub>S<sub>1</sub>. The increase in 1000 seed weight could be attributed to zinc as it is responsible for improved seed filling due to its role in pollen germination, cell division, sugar transfer from source to sink and seed development Masuthi et al., [12]. Pariari et al., [13] in fenugreek and Lakshami et al., [14] in black gram, Rahman et al., [15], in okra, Reza et al., [16] in lentil, Matinez et al., [17] in soybean support these findings. Zinc spray at bolting stage might have boosted the accumulation of assimilates in the seed, resulting in maximum 1000 seed weight as compared to later stages of zinc application.

# 3.2 Germination Percentage

In accordance with (Table 1), germination percentage of harvested seeds was significantly

influenced by zinc levels, stages of application and their interaction. Among zinc levels, there was a significant increase in germination (80.92 %) up to  $Zn_3$  (2g  $I^{-1}$ ) and minimum (70.83 %) was recorded in Zn<sub>1</sub> (Water spray). However, in case of stages of application, maximum germination (80.62 %) was found during first stage of foliar application S<sub>1</sub> (Bolting stage) and foliar application at subsequent stages led to decrease in germination of the harvested seeds and minimum (74.19 %) was registered in S<sub>3</sub> (Bolting + 14 days + 28 days after first application). Whereas, in interaction, maximum germination (85.75 %) was observed in interaction Zn<sub>3</sub>S<sub>1</sub> (2g I<sup>-1</sup> foliar spray at bolting stage) while minimum germination (70.75 %) was found in Zn<sub>1</sub>S<sub>2</sub> (Water spray at bolting + 14 days after first application) and Zn<sub>1</sub>S<sub>3</sub> (Water spray at bolting + 14 days + 28 days after first application) which was at par with Zn<sub>1</sub>S<sub>1</sub>.The increased germination of harvested seeds may attributed to zinc's involvement in numerous catalytic activities and the breakdown of complex chemicals into their simplest form, such as glucose, amino acids and fatty acids. Zinc is also aggregation of food responsible for the molecules such as proteins and carbohydrates, well rapid transport as their source to growing seed, resulting in increased germination of harvested seeds [18]. Yoganand [19] in bell pepper and Kiran [20] in brinjal also noticed.

Table 1. Effect of foliar application of zinc, stages of application and their interaction on 1000 seed weight (g), germination % seeds, seedling length (cm), seedling dry weight(mg), seed vigour index-l in radish cv. Japanese White

Crop Stage	Zn dose	1000 seed weight (g)	Germination percentage	Seedling length (cm)	Seedling dry weight (mg)	SVI-I
S <sub>1</sub>	Zn₁	12.29	71.00(8.48)*	19.64	9.18	1,394.25
	Zn <sub>2</sub>	12.28	70.75(8.47)	19.97	9.16	1,412.12
	Zn <sub>3</sub>	12.28	70.75(8.47)	20.17	9.12	1,426.71
	Zn <sub>4</sub>	13.47	83.00(9.16)	21.82	10.04	1,811.68
S <sub>2</sub>	Zn₁	12.28	70.75(8.47)	19.97	9.16	1,412.12
	Zn <sub>2</sub>	13.44	77.00(8.83)	22.23	10.83	1,712.62
	Zn <sub>3</sub>	13.26	81.50(9.08)	20.82	9.33	1,706.78
	Zn <sub>4</sub>	13.23	77.50(8.86)	21.95	10.64	1,700.58
S <sub>3</sub>	Zn₁	12.28	70.75(8.47)	20.17	9.12	1,426.71
	Zn <sub>2</sub>	13.45	74.75(8.70)	21.26	9.45	1,589.61
	Zn <sub>3</sub>	13.02	75.50(8.75)	21.37	9.83	1,613.89
	Zn <sub>4</sub>	13.22	75.75(8.76)	21.12	9.43	1,599.28
CD						
Zn		0.06	0.07	0.47	0.04	40.00
S		0.05	0.06	0.41	0.04	34.65
Zn X S		0.10	0.12	0.82	0.72	69.29

\*Figures in parenthesis represent the square root transformed value

# 3.3 Seedling Length (cm)

It is apparent from the data (Table 1) that there was a significant impact of zinc levels, stages of application and treatment combinations on seedling length. In case of zinc levels, maximum seedling length (22.13 cm) was observed in Zn<sub>3</sub> (2g l-1) with a linear increase up to this level and at higher application of zinc (3g I-1), seedling length was decreased significantly, whereas minimum seedling length (19.93 cm) was recorded in Zn<sub>1</sub> (Water spray). Among stages of application, maximum seedling length (21.75 cm) was noticed in S<sub>1</sub> (Bolting stage) while minimum (20.98 cm) was found in last stage of foliar application i.e.S<sub>3</sub> (Bolting + 14 days + 28 days after first application). Amongst treatment combinations, significantly maximum seedling length (24.20 cm) was registered in Zn<sub>3</sub>S<sub>1</sub> (2g l-1 zinc application at the time of bolting) and minimum (19.64) was obtained in Zn<sub>1</sub>S<sub>1</sub> (Water spray at the time of bolting) being at par with Zn<sub>1</sub>S<sub>2</sub> and Zn<sub>1</sub>S<sub>3</sub>. The increased seedling length could be attributed to higher metabolic activity of indole acetic acid and auxins [21], which could have resulted in rapid cell division and cell elongation, resulting in increased seedling length. The results coincide with the findings of Kiran [20] in brinjal, Reza et al., [16] in lentil.

# 3.4 Seedling Dry Weight (mg)

According to data (Table 1), the zinc levels, stages of foliar application and their interaction significantly influenced the seedling dry weight. A continuous increase in the seedling dry weight (10.16 mg) was observed up to Zn<sub>3</sub> (2g l-1) and there after a decrease was recorded in Zn4 however, minimum seedling dry weight (9.16 mg) was recorded in Zn<sub>1</sub> (Water spray). In stages of application, maximum seedling dry weight (10.04 mg) was obtained in S<sub>1</sub> (Bolting stage) and afterwards, there was a sharp reduction in it having minimum seedling dry weight (9.46 mg) in S<sub>3</sub> (Bolting + 14 days + 28 days after first application). On the other hand, among the interactions, maximum seedling dry weight (11.31 mg) was recorded in Zn<sub>3</sub>S<sub>1</sub> (2g l<sup>-1</sup> ZnSO<sub>4</sub> at the time of bolting) which was at par with  $Zn_2S_2$  and  $Zn_4S_2$  and minimum (9.12 mg) was observed in Zn<sub>1</sub>S<sub>3</sub> (Water spray done at the time of bolting + 14 days + 28 days after first application) which was at par with Zn<sub>1</sub>S<sub>1</sub> and Zn<sub>1</sub>S<sub>2</sub> along with majority of other treatment combinations. Maximum seedling dry weight may be due to longer seedling length and lipid consumption via the glyoxylate cycle, which is

the primary pathway that elevated the faster growth and seedling development to reach autotrophic stages sufficiently early and produce relatively more dry weight [22]. In brinjal, the results concur with the studies conducted by Kiran [20], Reza et al., [16] in lentil.

# 3.5 Seedling Vigour Index-I

Examination of the data (Table 1) revealed thatseed vigour index-I was significantly affected by the zinc levels, stages of application and their interaction. Among different zinc levels, maximum SVI-I (1798.58) was observed in Zn<sub>3</sub> (2g l-1) up to which there was uncurvingly increase in SVI-I and after that there was a decline in SVI-I and recording minimum (1411.02) in Zn<sub>1</sub> (Water spray). In stages of application, maximum SVI-I (1761.72) was obtained in S<sub>1</sub> (Bolting stage) and at later stages there was reduction in SVI-I with minimum (1557.37) being observed in S<sub>3</sub> (Bolting + 14 days + 28 days after first application). On the other hand, among treatment combinations. maximum SVI-I (2075.08) was recorded in  $Zn_3S_1$  (2g  $I^{-1}$  at the time of bolting) which was superior among all the combinations, however minimum (1394,25) was found in Zn<sub>1</sub>S<sub>1</sub> (Water spray at the time of bolting) which was at par with Zn<sub>1</sub>S<sub>2</sub> and Zn<sub>1</sub>S<sub>3</sub>.The findings are similar to that of Kiran [20] in brinjal. The highest seed vigour index-I could be owing to the fact that SVI-I is a derived parameter as SV-I was determined by multiplying germination percentage and seedling length (cm), therefore as germination percentage and seedling length increased, so did the seed vigour index-I.

# 3.6 Seedling Vigour Index- II

Analysis of the data (Table 3) depicted a significant difference in seed vigour index-II when zinc was sprayed at different levels and stages. A linear increase in SVI-II (822.64) was discernible with zinc up to 2g I-1 (Zn<sub>3</sub>) and then there was a decline in SVI-II at Zn<sub>4</sub> and minimum value (648.49) was obtained in Zn<sub>1</sub> (Water spray). Similarly, among different stages of application, maximum SVI-II (810.78) was recorded in S<sub>1</sub> (Bolting stage) and then there was a decrease in SVI-II with every stage of zinc application with minimum SVI-II (701.90) in S<sub>3</sub> (Bolting + 14 days + 28 days after first application). However, in case of interactions, maximum (966,77) SVI-II was observed in Zn<sub>3</sub>S<sub>1</sub> (2g I<sup>-1</sup> at the time of bolting) and minimum (645.07) in observed in Zn<sub>1</sub>S<sub>3</sub> (Water spray at bolting + 14 days + 28 days after first application) had statistical similarity with Zn<sub>1</sub>S<sub>1</sub> and Zn<sub>1</sub>S<sub>2</sub>. The highest seed

Table 2. DMRT Analysis table

Treatment Name	Treatment Mean	Least Significant Difference	Treatment Mean	Least Significant Difference	Treatment Mean	Least Significant Difference	Treatment Mean	Least Significant Difference
1000seed weight (g)		Germination(%)		Seedling Length (cm)		Seedling dry weight (mg)		
Zn₁S₁	12.283 <sup>b</sup>	1.749	71 <sup>d</sup>	3.271	19.643 <sup>b</sup>	4.409	9.185ª	3.082
Zn₁S₂	12.28 <sup>b</sup>	1.749	70.75 <sup>d</sup>	3.271	19.968 <sup>ab</sup>	4.409	9.165 <sup>a</sup>	3.082
Zn₁S₃	12.677 <sup>ab</sup>	1.749	70.75 <sup>d</sup>	3.271	20.168ab	4.409	9.118 <sup>a</sup>	3.082
Zn₂S₁	13.437 <sup>ab</sup>	1.73	82.75 <sup>ab</sup>	3.182	21.34ab	4.409	9.615 <sup>a</sup>	3.082
Zn <sub>2</sub> S <sub>2</sub>	13.447 <sup>ab</sup>	1.702	77°	3.271	22.233ab	4.146	10.835a	2.898
Zn₂S₃	14.357a	0	74.75 <sup>c</sup>	3.271	21.265ab	4.409	9.45 <sup>a</sup>	3.082
Zn₃S₁	13.26 <sup>ab</sup>	1.749	85.75 <sup>a</sup>	0	24.203a	0	11.308a	0
Zn <sub>3</sub> S <sub>2</sub>	13.02 <sup>ab</sup>	1.749	81.5 <sup>b</sup>	3.235	20.825ab	4.409	9.333a	3.082
Zn <sub>3</sub> S <sub>3</sub>	13.473 <sup>ab</sup>	1.645	75.5 <sup>c</sup>	3.271	21.375ab	4.409	9.827 <sup>a</sup>	3.082
Zn <sub>4</sub> S <sub>1</sub>	13.227 <sup>ab</sup>	1.749	83 <sup>ab</sup>	3.075	21.825ab	4.361	10.045a	3.048
Zn <sub>4</sub> S <sub>2</sub>	13.217 <sup>ab</sup>	1.749	77.5 <sup>c</sup>	3.271	21.95ab	4.29	10.643a	2.998
Zn <sub>4</sub> S <sub>3</sub>	12.283 <sup>b</sup>	1.749	75.75 <sup>c</sup>	3.271	21.118ab	4.409	9.428a	3.082

Table 3. Effect of foliar application of zinc, stages of application and their interaction on seed vigour index-II, electrical conductivity (dSm<sup>-1</sup>), accelerated ageing test, cold test in radish cv.

Japanese White

Crop Stage	Zn dose	SVI-II	Electrical conductivity (dSm <sup>-1</sup> )	Accelerated ageing test	Cold test
S <sub>1</sub>	Zn₁	651.95	0.041	70.50(8.45)	70.50(8.46)
	Zn <sub>2</sub>	794.40	0.018	76.00(8.77)	76.75(8.82)
	Zn <sub>3</sub>	966.77	0.017	78.25(8.90)	81.00(9.05)
	Zn <sub>4</sub>	830.00	0.019	75.00(8.72)	73.25(8.62)
S <sub>2</sub>	Zn₁	648.43	0.041	70.00(8.43)	71.00(8.48)
	Zn <sub>2</sub>	834.30	0.025	73.25(8.62)	74.00(8.66)
	Zn <sub>3</sub>	759.15	0.023	73.00(8.60)	80.75(9.04)
	Zn <sub>4</sub>	824.78	0.028	71.50(8.51)	73.50(8.63)
<b>S</b> <sub>3</sub>	Zn₁	645.07	0.041	70.00(8.43)	70.50(8.46)
	Zn <sub>2</sub>	759.15	0.023	73.00(8.60)	80.75(9.04)
	Zn <sub>3</sub>	741.98	0.031	71.75(8.53)	75.00(8.71)
	Zn <sub>4</sub>	714.17	0.038	71.50(8.51)	73.50(8.63)
CD					
Zn		12.81	0.001	0.05	0.08
S		11.10	0.001	0.04	0.07
Zn X S		22.19	0.001	0.08	0.14

\*Figures in parenthesis represent the square root transformed value

vigour index-II could be because SVI-II is also a derived metric determined by multiplying germination (%) by seedling dry weight (mg). Thus, when the germination (%) and seedling dry weight increased, similar trend in SVI-II was obvious.

# 3.7 Electrical Conductivity (dS m<sup>-1</sup>)

In insight into the data (Table 3) showed that zinc levels, stages of application and their interaction had a significant influence on conductivity. In case of different levels of zinc, minimum electrical conductivity (0.023 dS m<sup>-1</sup>) was noticed in Zn<sub>3</sub> (2g I-1) and thereafter there was an increase in electrical conductivity with zinc applied @ 3g I-1 while maximum EC (0.041 dS m-1) was recorded in Zn<sub>1</sub> (Water spray). Regarding different stages of application, minimum EC (0.024 dS  $m^{-1}$ ) was obtained in  $S_1$ (Bolting stage) and after that EC of the seed was found to increase with every subsequent stage of application and maximum value (0.036 dS m<sup>-1</sup>) was recorded in S<sub>3</sub> (Bolting + 14 days + 28 days after first application). Among different treatment combinations, minimum EC (0.017 dS m<sup>-1</sup>) was registered in Zn<sub>3</sub>S<sub>1</sub> (2g I<sup>-1</sup> at the time of bolting) whereas, maximum EC (0.041 dS m-1) was recorded in Zn<sub>1</sub>S<sub>1</sub> (Water spray at the time of bolting), Zn<sub>1</sub>S<sub>2</sub> (Water spray at the time of bolting + 14 days first application) and Zn<sub>1</sub>S<sub>3</sub> (Water spray at the time of bolting + 14 days + 28 days after first application). The electrical conductivity

of seed is greatly dependent on seed vigour. The electrical conductivity of the seed is inversely proportional to its vigour, as seed vigour is greatest when electrical conductivity is lowest. The decrease in EC with zinc administration can be explained by the fact that this element is known to promote cell wall integrity, reducing solute leaking from the seed. In brinjal, the results concur with the findings of Kiran [20].

# 3.8 Accelerated Ageing Test

From the data (Table 3), it is extrapolated that there were significant differences among different zinc levels, stages of application and their interaction for this parameter. Among zinc levels, maximum germination (74.33 %) for the parameter was obtained inZn<sub>3</sub> (2g I <sup>-1</sup>) which was rectilinear maximum and minimum germination (70.17 %) was recorded in Zn<sub>1</sub> (Water spray). Whereas, in case of stages of application, maximum germination (74.93 %) was obtained in S<sub>1</sub> (Bolting stage) and thereafter there was a reduction in germination (%) of seed with every consecutive stage of application with minimum germination (71.31 %) being observed in S<sub>3</sub> (Bolting + 14 days + 28 days after first application). The improvement in germination (%) of the accelerated ageing test could be related to role of zinc increasing cell membrane integrity. As a result, the seed's vitality was increased, which lasted longer during the ageing process and resulted in a greater germination percentage

after the accelerated ageing test. Artificial ageing damages DNA and mRNA, resulting in biochemical degradation of the reserved material in the seed, which increases seed vigour [23].

# 3.9 Cold Test

An insight into the data (Table 3) revealed a significant effect of different levels of zinc, stages of application and their interaction on this parameter. Maximum germination (78.92 %) in cold test was recorded in Zn<sub>3</sub> (2g l<sup>-1</sup>) up to which there was linear increase and reduced as concentration of zinc application was increased to 3g l<sup>-1</sup> while minimum germination (70.67 %) was observed in Zn<sub>1</sub> (Water spray). Among stages of application, maximum germination (75.37 %) was obtained in S<sub>1</sub> (Bolting stage) and then, it declined with every subsequent stage reaching minimum (73.42 %) at S<sub>3</sub> (Bolting stage + 14 days + 28 days after first application). Amongst treatment combinations, maximum germination (81.00%) in cold test was found in Zn<sub>3</sub>S<sub>1</sub> (2g l<sup>-1</sup> with foliar zinc at the time of bolting) minimum germination (70.50%) observed in Zn<sub>1</sub>S<sub>1</sub> (Water spray at the time of bolting) and Zn<sub>1</sub>S<sub>3</sub> (Water spray at the time of bolting + 14 days + 28 days after first application). The cold test was created to mimic harsh field conditions and assess the capacity of seeds to emerge. The germination percentage of Zn<sub>3</sub>S<sub>1</sub> was higher because foliar application of zinc during the bolting stage might have increased photosynthates transport, available nutrients in seeds and cell membrane integrity, thereby inclination preserving the structural macromolecules and ion transport systems, resulting in greater vigour and germination [24].

# 4. CONCLUSION

Among different zinc levels,  $Zn_3(2g\ l^{-1})$  was considerable superior to other levels for majority of seed quality parameters. In case of stages of application,  $S_1$  (Bolting stage) showed its superiority above all other stages. In treatment combinations,  $Zn_3S_1$  ( $2g\ l^{-1}$  at the time of bolting) was superior over other treatment combinations for most of seed quality parameters namely, 1000 seed weight (g), germination percentage of harvested seeds, seedling length (cm), seedling dry weight (mg), seed vigour index-I, seed vigour index-II, accelerated ageing test, cold test.

# **DISCLAIMER (ARTIFICIAL INTELLIGENCE)**

Author(s) hereby declare that NO generative Al technologies such as Large Language Models

(ChatGPT, COPILOT, etc) and text-to-image generators have been used during writing or editing of this manuscript.

#### **COMPETING INTERESTS**

Authors have declared that no competing interests exist.

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