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Optimization Of Nanoparticles Based Seed Priming Treatments for Improving Germination Potential and Vigor Performance in Lentil (*Lens Culinaris* Medik) Under Laboratory Conditions

Shailendra Tiwari[1]* • J.S. Chauhan[1] • Vinay Chamoli[2] • Meenakshi Joshi[1] • Sonali Rajput[1]

[1]*Department of Seed Science and Technology, H.N.B.G.U, Srinagar, Garhwal*
[2]*Department of Agriculture, JB Institute of Technology (JBIT), Dehradun*

*Corresponding Author Email id: shailendratiwari@hnbgu.ac.in

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Abstract: The present investigation, conducted in September 2024, aimed to optimize seed priming treatments using various nanoparticles with different concentrations to enhance germination and seed quality of lentil (*Lens culinaris* Medik.) under lab conditions. The experiment was conducted using five seed priming treatments, excluding hydro-priming and control: Silicon dioxide (SiO_2), Zinc oxide (ZnO), Iron oxide (Fe2o3), Titanium Dioxide (TiO_2) and Copper oxide (CuO) with different concentrations (50 ppm, 100 ppm, 150 ppm and 200 ppm), hydro-priming, and an untreated (control) under laboratory conditions. Each treatment was replicated four times in a completely randomized design (CRD). Seeds were primed overnight and then evaluated for germination percentage (first and final counts), seedling length (cm), seedling fresh weight (g), seedling dry weight (g), and seed vigor indices. Hydropriming was a highly effective and low-cost technique, resulting in a massive improvement in the final germination percentage, and its early growth characteristics were highly significant. Among the nutrient treatments, the highest levels enhanced germination/vigor traits: ZnO (200 ppm), $Fe2_{O}3$ (50 ppm), SiO_2 (100 ppm), TiO_2 (150 ppm), and CuO (200 ppm). In most cases, low or high levels compared to the optimum also minimized the advantages, whereas both high levels of Fe2o3, TiO_2, and CuO displayed inhibitory effects on some traits.

Keywords: Seed priming • Nanoparticles • Germination and vigor • Lentil

Introduction

Early seed germination, seedling development, and plant growth are critical functions in the plant lifecycle that directly impact establishment and crop production (Bewley *et al* 2013). In pulse-like lentils (*Lens culinaris* Medik), seed vigor and uniform germination are crucial for achieving the best possible yield, especially when environmental conditions are variable. A pre-sowing treatment, seed priming, has helped regulate biochemical and physiological processes and emergence as a valuable method to enhance germination percentage, seedling vigor, and stress resistance (Farooq *et al* 2019).

Innovative developments have recently been made in nanotechnology, introducing nanoparticle-based seed priming as a promising alternative method. Due to their large surface area and reactivity, nanoparticles can efficiently deliver seeds, bioactive chemicals, and nutrients, stimulate metabolic activities, and enhance the performance of the seed (Mahakham *et al* 2017). It has been found that nanoparticles, such as ZnO, SiO2, and Fe3O4, can increase water uptake, enhance antioxidant enzyme activity, and decrease germination time in various crops (Chaparro *et al* 2025; Gupta *et al* 2022). Equally, titanium and copper are critical micronutrients that enhance enzyme activity and increase seed stress resistance. Furthermore, the absorption of water, hydro priming, introduced as a pre-sowing hydration technique, can also enhance absorption and enable faster, synchronized seed germination. The current

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research study aims to assess and enhance such treatments to improve seed germination and seed quality in lentils.

Materials and Methods

To understand the effect of treatment using nanoparticle-based and hydro-priming on germination and seed quality, the experiment was conducted in September 2024, under laboratory conditions in the Department of Seed Science and Technology, School of Agriculture and Allied Sciences, Chauras Campus, H.N.B. Garhwal University, Srinagar Garhwal, Uttarakhand. Five interventions, including nanoparticle zinc oxide (ZnO), copper oxide (CuO), iron oxide (Fe_2O_3), titanium dioxide (TiO_2), and silicon dioxide (SiO_2), were utilized at four concentrations: 50 ppm, 100 ppm, 150 ppm, and 200 ppm. In addition to these, a hydro-priming treatment was also employed, in which seeds were treated with distilled water overnight (12 hours) and then allowed to dry before being subjected to a germination test. Comparison was made with unprimed seeds as well. Surface sterilization of lentil (**VL-126**) seeds was performed using 70% ethanol, following cleaning with ethanol and allowing them to dry. The cleaning process was completed, and the seeds were washed with distilled water for one minute. The seeds were soaked in nanoparticle priming solutions overnight, followed by shade-drying to restore their original moisture content levels. The experiment was conducted in a completely randomized design (CRD) at four replications. Seeds were exposed to the treatment, after which germination trays of moist towel paper were used to incubate the seeds at 20 ± 1°C with a relative humidity of 70-80%. The percentage germination was noted, as well as the seedling's length (cm), fresh weight of seedling (g), dry weight of seedling (g), and seedling vigor index.

Results and Discussion

The analysis of variance exhibited the application of different nanoparticles, hydropriming, and control on germination and seedling parameters in Lentil (*Lens culinaris* Medik).

Zinc oxide (ZnO) priming treatments significantly improved germination and seedling vigor compared to hydropriming and the control (Table 1). Seeds primed with 200 ppm ZnO show an initial germination percentage (95.5%) and final germination percentage (98%), followed by ZnO at 100 ppm (94.5% and 98%). The 200 ppm Zn treatment recorded the highest performance among all zinc oxide doses, the most extended shoot length (15.04 cm) and root length (9.54 cm), Fresh weight (1.659 g) and dry weight (0.1618 g), and the height vigor index I^{st} (2292.53) and vigour index II^{nd} (3.987).

Table 1: Effect of Zinc Oxide (ZnO) Nanoparticles on Germination and Seedling Parameters in Lentil

Treatment	Dose (PPM)	FC (%)	FG (%)	SL (cm)	RL (cm)	SFW (g)	SDW (g)	VI 1	VI 2
Zinc oxide (ZnO)	T1 (50)	88.5	93.25	8.982	5.85	1.188	0.1572	1452.46	2.329
	T2 (100)	94.5	98.0	10.76	6.788	1.257	0.1502	1715.17	2.645
	T3 (150)	89.0	92.5	13.025	8.872	1.217	0.1535	2025.1	3.367
	T4 (200)	**95.5**	**98.0**	**15.04**	**9.54**	**1.659**	**0.1618**	**2292.535**	**3.987**
Mean		**91.875**	**95.4375**	**11.9519**	**7.7625**	**1.3303**	**0.1557**	**1871.3163**	**3.0819**
Sem		1.5069	1.2055	0.7584	0.3783	0.0833	0.0048	95.3742	0.2270
Sed		2.1311	1.7048	1.0726	0.5350	0.1178	0.0068	134.8794	0.3210
CV%		3.2804	2.5262	12.6915	9.7465	12.5184	6.1866	10.1933	14.7309
CD%		(4.643, 6.509) *	(3.714, 5.2073) **	(2.337, 3.2763) **	(1.1656, 1.6341) **	(0.256, 0.3597) **	(0.014, 0.0208)	(293.877, 411.9946) **	(0.699, 0.9806) **

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Overall, the results confirm that 200 ppm enhances seedling length, fresh and dry weight, and vigor in lentils under overnight priming conditions. The differences among treatments were highly significant at par for all germination-related traits, final germination percentage, shoot length, root length, fresh weight, and vigor index ($p \leq 0.01$). While the first count germination, the treatments were significant at par ($p \leq 0.05$). Moreover, it is also reported that ZnO seed priming has wider advantages, with the research results demonstrating enhanced seed vigor, early development, and biomass accumulation across several types of crops (Donia & Carbone 2023; Prajapati *et al* 2024).

Silicon dioxide (SiO_2) priming treatments had a positive effect on seed germination and vigor compared to hydropriming and the unprimed control (Table 2).

Table 2: Effect of Silicon Dioxide (SiO_2) Nanoparticles on Germination and Seedling Parameters in Lentil

Treatment	Dose (PPM)	FC (%)	FG (%)	SL (cm)	RL (cm)	SFW (g)	SDW (g)	VI 1	VI 2
Silicon dioxide (SiO_2)	T5 (50)	89.0	92.5	11.7625	9.145	1.4557	0.1635	1924.57	3.4057
	T6 (100)	**92.75**	**98.5**	**12.8875**	**9.525**	**1.5452**	**0.169**	**2207.6**	**3.7838**
	T7 (150)	90.0	94.5	11.53	9.458	1.5125	0.1685	1984.615	3.5402
	T8 (200)	92.5	95.0	11.525	8.328	1.445	0.1642	1885.795	3.2629
Mean		**91.0625**	**95.125**	**11.9262**	**9.1137**	**1.4896**	**0.1663**	**2000.645**	**3.4981**
Sem		0.8385	1.0508	0.3923	0.2772	0.1196	0.0065	60.7834	0.1727
Sed		1.1859	1.4860	0.5548	0.3920	0.1691	0.0092	85.9607	0.2442
CV%		1.8416	2.2093	6.5791	6.0835	16.0540	7.8296	6.0764	9.8716
CD%		(2.583, 3.6222) *	(3.237, 4.5392) *	(1.208, 1.6947)	(0.8542, 1.1975)*	(0.368, 0.5165)	(0.020, 0.0281)	(187.292, 262.5703) *	(0.532, 0.7459)

The 100 ppm SiO_2 treatment recorded the best performance, as indicated by First count (92.75%), Final germination percentage (98.5%), root length (9.525 cm), and Vigor Index (I^{st}) (22.76) with significant improvement ($p \leq 0.05$) compared to other doses, hydropriming, and the control. Seed primed with 50 ppm and 150 ppm showed moderate improvements, while 200 ppm SiO_2 slightly declined these parameters. For final germination, shoot length, fresh weight, and Vigor index II were non-significant among SiO_2 treatments, indicating that silicon concentration had a minimal effect on these characters. Overall, 100 ppm SiO_2 proved to be the most effective dose for improving early germination and vigor traits in lentil seeds under overnight priming conditions. Nanopriming with SiO2 presents a promising, economical, and scalable approach to enhance germination, early growth, and drought resistance in jalapeno pepper cultivation under semi-arid conditions (Chaparro *et al* 2025). Combining silicic and humic acids improved seedling growth and reduced oxidative damage in lentil plants under salt stress conditions (Rao *et al* 2023).

Iron oxide (Fe_2O_3), priming treatments have a marked influence on seed germination and early seedling growth of lentils (Table 3).

Among the tested doses, 50 ppm Iron emerged as the most effective, recording highly significant ($p \leq 0.01$) improvements over the control. Seeds primed with 50 ppm iron exhibited rapid and uniform germination, accompanied by strong seedling growth and higher biomass accumulation. In contrast, the higher concentrations of 100 ppm, 150 ppm, and 200 ppm iron failed to produce a comparable response.

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Table 3: Effect of Iron Oxide (Fe2o3) Nanoparticles on Germination and Seedling Parameters in Lentil

Treatment	Dose (PPM)	FC (%)	FG (%)	SL (cm)	RL (cm)	SFW (g)	SDW (g)	VI 1	VI 2
Iron oxide (Fe2O3)	**T9 (50)**	**89.5**	**94.5**	**9.972**	**7.318**	**1.326**	**0.176**	**1639.125**	**3.017**
	T10 (100)	0.0	2.5	0.0	0.0	0.0	0.0	0.0	0.0
	T11 (150)	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
	T12 (200)	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Mean		**22.375**	**24.25**	**2.4931**	**1.8294**	**0.3315**	**0.0439**	**409.7812**	**0.7542**
Sem		0.4787	1.0607	0.4559	0.5331	0.0341	0.0050	98.7727	0.1665
Sed		0.6770	1.5	0.6447	0.7540	0.0482	0.0071	139.6857	0.2355
CV%		4.2790	8.7477	36.5713	58.2871	20.5431	22.7098	48.2075	44.1564
CD%		(1.475, 2.0679) **	(3.268, 4.5818) **	(1.404, 1.9693) **	(1.6428, 2.3031) **	(0.104, 0.1471) **	(0.015, 0.0216) **	(304.349, 426.6755) **	(0.513, 0.7193) **

The superiority of 50 ppm iron across all key germination and vigor traits highlights its potential as the optimal dose for improving seedling establishment and early growth in lentil under overnight priming conditions. Thus, 50 ppm Fe_3O_4 priming emerges as a promising strategy to accelerate lentil seed germination and vigor, consistent with analogous findings in mungbean crop studies employing iron oxide nanoparticle priming (Triveni *et al* 2021). According to Khan *et al* (2019), seed priming with 100 ppm showed maximum values of germination percentage, shoot length, shoot fresh weight and dry weight, root length, fresh weight of the root, and root dry weight. Seed priming with FeSo4 significantly affects the growth of *Vigna radiata L.* under NaCl stress and improves germination, growth, and biochemical parameters.

Copper oxide (CuO) priming treatments influenced germination and seedling of vigor of lentil seeds, with the highest concentration of 200 ppm copper producing the best performance (First count 97%, final germination 97.5 %, shoot length 10.052 (cm), root length 8.202 (cm), fresh weight 1.394 (g), dry weight 0.1625 (g), vigor index 1771.68 and vigor index II 2.958) and first count, shoot length, root length and vigor indices highly significant ($p \leq 0.01$). Treatments showed significant ($p \leq 0.05$) improvement in fresh weight and significant ($p \leq 0.05$) over the control (Table 4).

Table 4: Effect of Copper Oxide (CuO) Nanoparticles on Germination and Seedling Parameters in Lentil

Treatment	Dose (PPM)	FC (%)	FG (%)	SL (cm)	RL (cm)	SFW (g)	SDW (g)	VI 1	VI 2
Copper oxide (CuO)	T13 (50)	92.0	97.0	6.642	3.548	1.105	0.1505	992.81	1.552
	T14 (100)	87.5	94.5	6.315	2.338	1.09	0.1582	817.655	1.367
	T15 (150)	92.0	97.0	6.86	4.558	0.952	0.163	1108.745	1.848
	T16 (200)	**97.0**	**97.5**	**10.052**	**8.202**	**1.394**	**0.1625**	**1771.68**	**2.958**
Mean		**92.125**	**96.5**	**7.4675**	**4.6612**	**1.1351**	**0.1586**	**1172.7225**	**1.9312**
SEm		1.0704	0.8660	0.5573	0.4792	0.0934	0.0068	98.3166	0.1589
SEd		1.5138	1.2247	0.7881	0.6777	0.1320	0.0096	139.0406	0.2246
CV%		2.3239	1.7949	14.9259	20.5619	16.4525	8.5562	16.7672	16.4508
CD%		(3.298, 4.6240) **	(2.668, 3.7410)	(1.717, 2.4074) **	(1.4766, 2.0701) **	(0.287, 0.4034) *	(0.020, 0.0293)	(302.943, 424.7050) **	(0.489, 0.6862) **

For other characters, namely final germination percentage and dry weight, differences among copper treatments were non-significant ($p > 0.05$), suggesting a limited influence of copper

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concentration on these traits. The findings confirm that 200 ppm copper is the most effective dose for better specific germination and vigor parameters in lentil under overnight priming conditions.

Titanium dioxide (TiO_2) priming treatments showed considerable variation in their effect on lentil germination and seedling growth parameters (Table 5).

Table 5: Effect of Titanium Dioxide (TiO_2) Nanoparticles on Germination and Seedling Parameters in Lentil

Treatment	Dose (PPM)	FC (%)	FG (%)	SL (cm)	RL (cm)	SFW (g)	SDW (g)	VI 1	VI 2
Titanium Dioxide (TiO2)	T17 (50)	96.5	97.5	9.817	6.95	1.23	0.165	1637.415	2.728
	T18 (100)	97.0	98.0	10.135	6.935	1.197	0.161	1684.735	2.684
	T19 (150)	**98.0**	**99.0**	**11.808**	**8.972**	**1.461**	0.1515	**2036.44**	**3.157**
	T20 (200)	93.0	96.0	6.648	3.41	1.006	0.1552	963.86	1.564
Mean		**96.125**	**97.625**	**9.6019**	**6.5669**	**1.2235**	**0.1582**	**1580.6125**	**2.5329**
Sem		0.75	0.5590	0.9911	1.1347	0.0955	0.0090	201.7923	0.3016
Sed		1.0607	0.7906	1.4017	1.6047	0.1351	0.0127	285.3774	0.4265
CV%		1.5605	1.1452	20.6445	34.5589	15.618	11.361	25.5334	23.8145
CD%		(2.311, 3.2398) **	(1.722, 2.4148) *	(3.054, 4.2815) *	(3.4964, 4.9017) *	(0.294, 0.4127) *	(0.027, 0.0388)	(621.784, 871.6967) *	(0.929, 1.3028) *

Among the tested concentrations 150 ppm titanium showed most effective, recoded highly significant ($p \leq 0.01$) improvement in first count (98.0 %) and significant (($p \leq 0.05$) improvement in final germination (99.00 %), shoot length (11.808 cm), root length (8.972 cm), fresh weight (1.461 g), vigor index Ist (2036.44) and vigor index IInd (3.157) compared to the hydropriming, control and other Titanium doses. The 150 ppm treatment promoted faster and more uniform germination and balanced seedling development, resulting in greater fresh biomass and a superior vigor index. Lower concentrations, 50 ppm and 100 ppm, showed moderate effects, while the high concentration, 200 ppm, led to a decline in most traits, indicating reduced success at high doses. These results showed 150 ppm titanium as the most suitable dose for improving germination efficiency and early seedling vigor in lentil under overnight seed priming conditions.

This finding aligns with previous studies, which have shown that titanium dioxide (TiO_2) nanoparticles improve germination metrics in lentils and other plants. For example, Feizi *et al* (2020) reported that the application of 300 ppm TiO_2 nanoparticles significantly improved the mean germination time by 39% and the seed germination rate by 62% in lentils. Additionally, TiO_2 effectively mitigated the adverse effects of cadmium toxicity, maintaining shoot length and enhancing seedling performance under stress. These

effects suggest that titanium nanoparticles play a role in promoting seed germination. Overall, these findings consolidate the understanding that titanium-based seed priming, especially at optimized concentrations such as 150 ppm, enhances seed germination performance and seedling vigor in lentils by promoting more uniform germination, faster emergence, and greater early biomass accumulation. Additionally, titanium dioxide nanoparticles exhibit protective effects against abiotic stresses, supporting plant growth resilience.

Hydropriming markedly improved lentil germination and seedling growth parameters compared to the control (unprimed). The treatments resulted in a significant ($p \leq 0.05$) enhancement in final germination percentage and highly significant ($p \leq 0.01$) improvements

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in first count germination, shoot length, root length, fresh weight, and vigor indices (Table 6).

Table 6: Effect of Hydropriming on Germination and Seedling Parameters in Lentil

Treatment	FC (%)	FG (%)	SL (cm)	RL (cm)	SFW (g)	SDW (g)	VI 1	VI 2
Hydropriming (T21)	91.5	95.0	4.108	2.778	0.8	0.176	653.6	1.179
Control (T 22)	75.0	92.0	1.24	0.905	0.514	0.17	196.615	0.387
Mean	**83.25**	**93.5**	**2.6738**	**1.8413**	**0.6568**	**0.173**	**425.1075**	**0.7833**
SEm	1.6202	0.7071	0.3079	0.3322	0.0361	0.0122	57.1072	0.1420
SEd	2.2913	1.0000	0.4355	0.4698	0.0510	0.0173	80.7618	0.2009
CV%	3.8923	1.5125	23.0344	36.0870	10.9882	14.1392	26.8672	36.2627
CD%	(5.606, 8.4948) **	(2.446, 3.7074) *	(1.065, 1.6146) **	(1.1497, 1.7419) **	(0.124, 0.1892) **	(0.042, 0.0641)	(197.616, 299.4185) **	(0.491, 0.7447) **

Note: *, **: significant at $P \leq 0.05$ and $P \leq 0.01$ probability levels, respectively. FC: First Count germination, FG: Final germination, SL: Shoot length, RL: Root length, SFW: Seedling Fresh weight, SDW: Seedling Dry weight, VI 1: Vigor index Ist, and VI 2: Vigor index IInd

Hydroprimed seeds exhibited faster germination, longer seedlings, and higher seedling vigor, indicating enhanced seed metabolic activity and improved establishment potential. However, differences in dry weight between hydroprimed and control treatments were non-significant ($p > 0.05$). Seed priming enhances germination, root initiation, and dry seedling weight (Azary et al 2020). Saglam *et al*. (2010) observed a higher germination percentage and improved seedling growth rate with hydropriming under increased water stress. Seedling emergence rate was also enhanced by seed priming with water (Golezani *et al* 2008).

Conclusion

The current study indicated a significant increase in germination parameters and seedling vigor at early growth stages of lentil with both hydropriming and nutrient-based seed priming compared to the unprimed control. Hydropriming was an efficient and cost-effective method, demonstrating significant improvement in early growth characteristics. Out of the nutrient treatments, the optimum level of each nutrient differs, as ZnO at 200 ppm, SiO_2 at 100 ppm, Fe_2O_3 at 50 ppm, TiO_2 at 150 ppm, and Cu at 200 ppm were found to have the most effective influence on significant germination and vigor qualities. The levels lower or higher than the optimum tend to diminish the benefits, and higher doses of Fe_2O_3, TiO_2, and CuO demonstrated reduced positive effects on selected traits. Throughout the treatments, germination, shoot and root elongation, fresh weight, and vigor index were significantly altered, whereas dry weight was not significantly changed in most cases. Although these results suggest that optimized priming treatment could improve the performance of lentil seeds, further testing under field conditions should aim to confirm the laboratory findings in actual farming conditions.

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