Advances in Bioresearch
Adv. Biores., Vol 16 (4) July 2025: 170-175

Print ISSN 0976-4585; Online ISSN 2277-1573
Journal's URL:http://www.soeagra.com/abr.html
CODEN: ABRDC3
DOI: 10.15515/abr.0976-4585.16.4.170175

Advances in Bioresearch

ORIGINAL ARTICLE

Effect of Copper Metal stress on Seed Germination Physiology of *Murraya koenigii* plant

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ABSTRACT
Stress in plants pertains to environmental conditions that adversely affect plant development. This research explores the effects of copper stress on the germination parameters of the *Murraya koenigii* plant. Through controlled experiments, we evaluate how different concentrations of copper influence the rate of seed germination, emergence, and overall seedling development. Consequently, in this study, *Murraya koenigii* seeds were collected, and various parameters such as the germination index, seed vigor index, and copper injury rate were analyzed in a growth chamber. Various concentrations of copper were tested over different time intervals. The germination rate was observed to decrease as the concentration of copper increased. At lower concentrations, specifically 2mM, germination was only slightly impacted; however, at higher concentrations, such as 5mM and 10mM, seed germination was significantly reduced due to copper toxicity. Therefore, this paper concludes that excessive copper levels are detrimental to seed germination. Seedling growth was assessed, and measurements were recorded 12 days post-treatment.
Keywords: Germination parameter, *Murraya koenigii,* copper stress, and seed germination.

Received 21.05.2025 Revised 21.06.2025 Accepted 21.07.2025

How to cite this article:
Dubey S, Umesh K and Kumari S. Effect of Copper Metal stress on Seed Germination Physiology of *Murraya koenigii* plant. Adv. Biores. Vol 16 [4] July 2025.170-175

INTRODUCTION

Plants derive a range of essential nutrients from the soil, including copper, which serves as a vital micronutrient necessary in minimal quantities for optimal growth and development. *Murraya koenigii*, commonly known as curry leaf or Kari Patta in Indian languages, belongs to the Rutaceae family, which includes around 150 genera and 1,600 species [1]. It is regarded as one of the most beneficial plants due to its nutritional and medicinal attributes. Abiotic stresses disrupt cellular homeostasis, affect plant metabolism, and interfere with essential physiological and biochemical processes. Metal stress constitutes a significant abiotic challenge to agricultural production globally. Copper (Cu) is an essential nutrient for humans, animals, and plants, participating significantly in a variety of morphological, physiological, and biochemical functions. Cu serves as a cofactor for numerous enzymes and is essential for respiration, photosynthesis, the antioxidant system, and signal transduction. Numerous studies have demonstrated that elevated copper concentrations can impede crop germination, growth, photosynthesis, and antioxidant function. Increasing Cu levels in curry plants resulted in a decline in germination rates, with roughly 60% of this decrease noted in plants subjected to 10 mM Cu stress [2, 3]. These studies demonstrate the adverse effects of copper stress on seed germination in curry plants. Seed germination represents a vital stage in plant development, signifying the onset of growth; consequently, this research endeavours to assess the influence of varying copper concentrations on seed germination, thereby examining the tolerance capabilities of plants under copper stress.

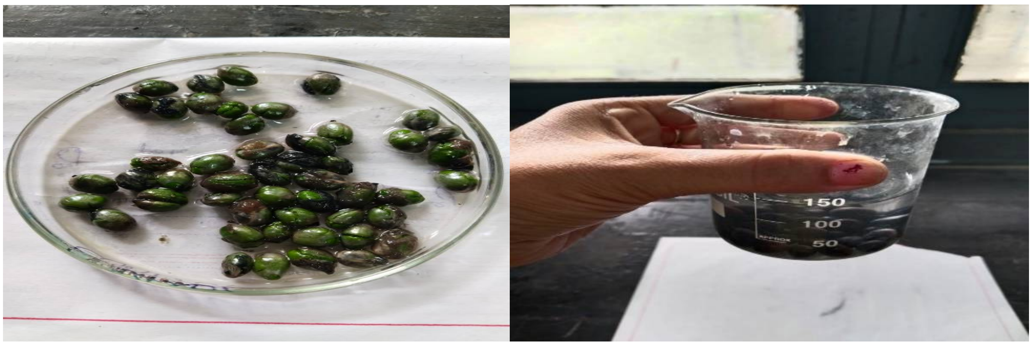
MATERIAL AND METHODS

Collection of seed

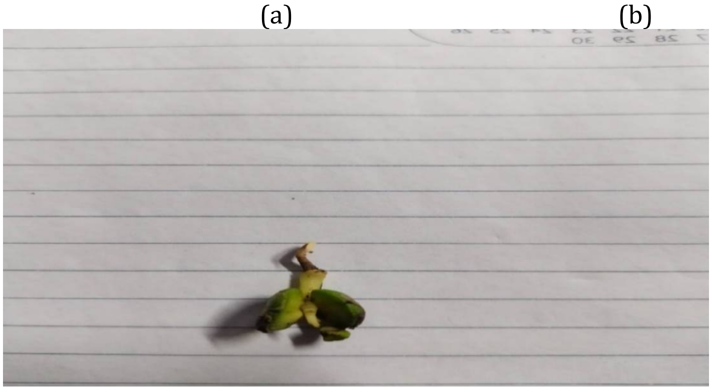
Seeds of *Murraya koenigii* were collected from the Botanical Garden of D.D.U. Campus Gorakhpur university Gorakhpur.

Germination studies

The seeds underwent surface sterilization using 0.1% mercuric chloride for a duration of 1 minute [3], followed by extensive rinsing with distilled water to mitigate fungal infection during the germination process. The seeds were subsequently germinated within sterilized Petri dishes measuring 120 mm in diameter. All the Petri dishes were initially cleaned using tap water, subsequently rinsed with distilled water, and then sterilized at 120˚C for 15 minutes in a hot air oven. The Petri dish is organized according to a fully randomized block design, comprising 3 replications. Solutions of 2, 5, and 10 mM copper were formulated in distilled water utilizing copper sulfate ($CuSO_4.5H_2O$). The unmanipulated group served as the control in the study. All experiments were conducted in triplicate. Eleven seeds were positioned on double-layer Whatman paper and imbibed for 72 hours. Subsequently, 10 ml of the appropriate concentration of copper sulfate solution was added to each Petri dish (fig. 1), with fresh solution applied every other day to mitigate contamination and sustain concentration. The Petri dish was maintained for a duration of 12 days during this time and was subjected to daily observation. Each day, 10 ml of distilled water was added to the Petri dishes. Data was collected for germination, seed vigor index, and relative rate of copper injury.



(a) (b)



(c)

Figure 1. (a) Collected seeds, (b) Stored seeds and (c) Germinated seed

Control 2mM 5mM 10mM

Figure 2. Germination of *Murraya Koengii* seeds in different copper concentrations

Germination percentage

Radicle and plumule emergence were taken as an index of germination. The germination percentage was recorded on the 12th day.
Germination percentage was calculated by the formula [2].
Germination percentage = No. of seeds germinated /Total no. of seeds ×100
Seed Vigor Index
It was calculated according to Baki and Andeson, [4]. In this, radical length was measured after 12 days of treatment.
Seed Vigor index = Radical length× Percent germination
Relative copper injury rate
It was calculated by following the formula of Li [5].
Relative copper injury rate = Germination % in control-Germination % in copper treatment/Germination percentage in control.
Statistical analysis
The experimental design comprised complete randomized blocks with 3 replicates. The results were evaluated by analysis of variance using statistical analysis system software SPSS 16.0, and treatment means were considered significantly different at $p<0.05$.

RESULT
Germination percentage
It was recorded 12 days after giving treatment. Radicle and plumule emergence were taken as an index of germination. Maximum germination percentage was observed in plants treated with control and slight reduction occurred in 2 mM having mild stress of copper and minimum reduction was observed in 10 mM having severe concentration of copper thus seed germinates but growth is not proper.

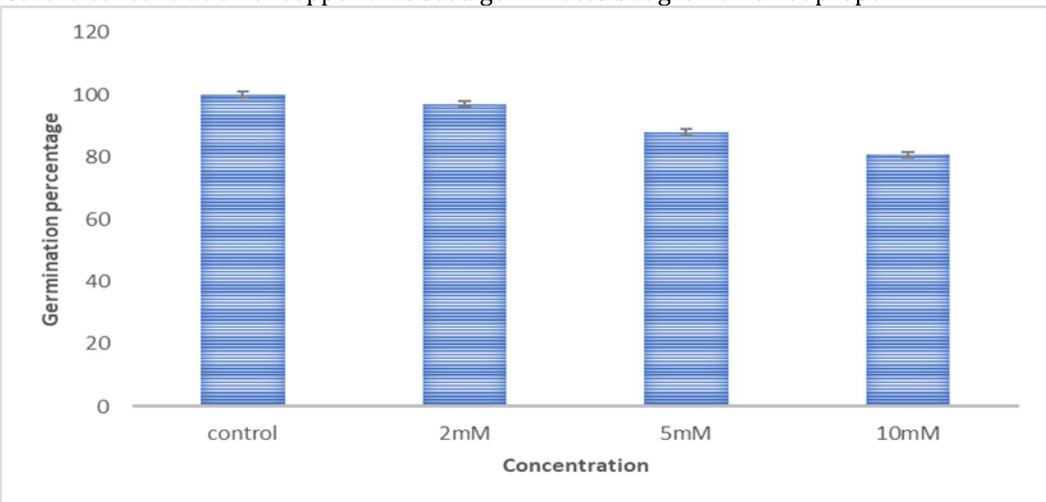


Figure 3: Germination percentage of *Murraya koenigii* seed at different copper concentrations
Seed Vigor index
The vigor index was assessed 12 days following the intervention. The highest Vigor index was noted in the control group, where seedlings developed naturally and exhibited normal lengths, with only a minor decrease at 2 mM and a significant decline at 10 mM.

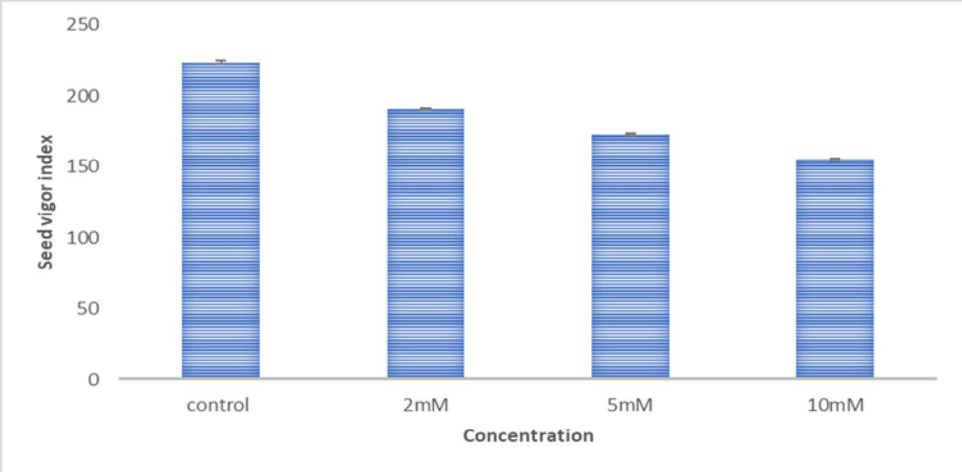


Figure -4: Seed vigor index of *Murraya koenigii* seed at different copper concentrations.

Relative copper injury rate

It was recorded after 12 days of treatment. The maximum copper injury rate was observed in 10 mM as germination is severely affected, followed by a decline in 5 mM and 2 mM with no injury rate in control.

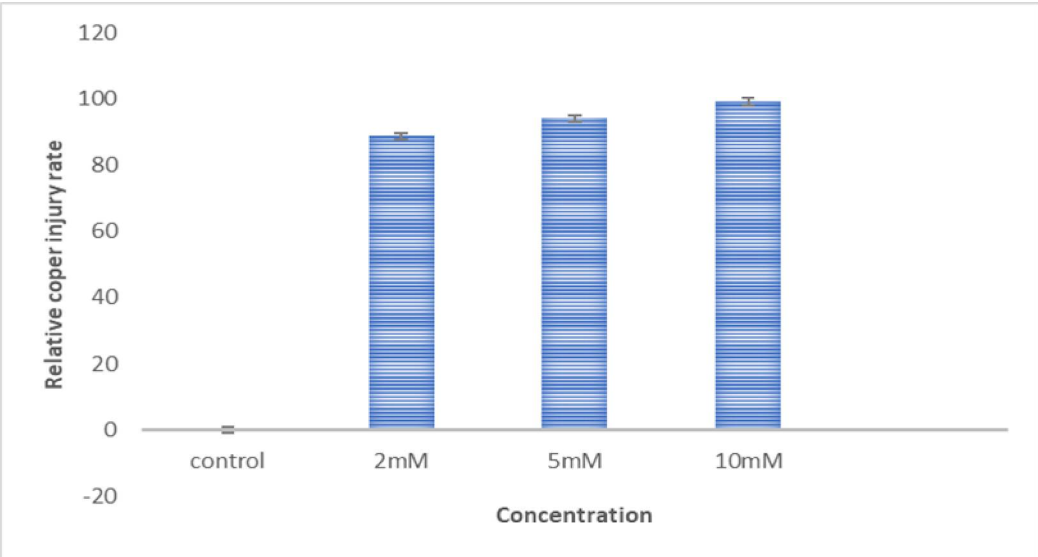


Figure -5: Copper injury rate of *Murraya koenigii* seed at different concentrations of copper.

Table -1 Effects of different concentrations of copper on seed germination, seed vigor index, and copper injury rate of *Murraya koenigii* seeds. (Average± standard error)

Treatments (Copper concentration mM)	Germination percentage	Seed vigor index	Copper injury rate
control	100±0	223.3±17.63	0±0
2mM	96.96 ±3.03	190.6±5.25	88.69±5.57
5mM	87.87± 3.03	172.7±5.49	94±2.08
10mM	80.66 ±1.20	154.9±10.16	99.12±0.03

Source of variation	Sum of square	Df	Mean of square	F	P- Value	F crit
Between-group	29975.06	2	14987.531	14.25	0.0016	4.25
Within group	9460.138	9	1051.12			
Total	39435.20	11				
LSD	**59.88**					

Table 2: Summary of analysis of variance for all the analyzed parameters using the ANOVA method.

The F-value is 14.26 and the p-value is 0.0016. There is a significant difference between the treatment groups, as indicated by the p-value (0.0016). The LSD value stands at 59.88, If the difference between the means of the two groups exceeds 59.88, they are considerably different.
In conclusion, the treatments show a statistically significant effect, indicating that at least one differs from others. More pairwise comparisons with LSD can reveal which specific treatments vary.

DISCUSSION

Copper (Cu) serves as an essential micronutrient for the growth of plants; however, when present in excessive amounts, it can become toxic, negatively impacting both seed germination and the maturation of seedlings. This study investigated the effects of varying copper concentrations (control, 2 mM, 5 mM, and 10 mM) on the germination rates of *Murraya koenigii* (curry leaf) seeds.
The results obtained indicate that higher copper concentrations are associated with a reduction in germination percentage. Seeds in the control group (0 mM Cu) demonstrated the highest germination rate, suggesting that optimal conditions promote effective seed development. However, at a concentration of 2 mM Cu, there was a slight decrease in germination. This suggests that even low levels of copper can begin to affect the physiological processes essential for germination.
At 5 mM Cu, there was a significant drop in germination, likely due to increased copper toxicity, which adversely affects water uptake and enzymatic activity. The most severe effects were observed at a copper concentration of 10 mM, where germination was markedly hindered or entirely suppressed. These findings are consistent with earlier research indicating that excessive copper reduces seed metabolism and obstructs root and shoot development [6-8].
Other plant species exposed to copper have shown comparable responses [9]. Similarly, studies on rice (Oryza sativa) have indicated that copper stress adversely affects early seedling growth and root elongation [10]. These investigations corroborate our findings that high levels of copper detrimentally affect seed germination and the initial growth of *Murraya koenigii* seeds.

CONCLUSION

This research investigates how copper (Cu) stress impacts the germination of *Murraya koenigii* seeds. The findings indicate that elevated copper levels negatively influence the germination percentage, with concentrations of 5 mM and 10 mM leading to a significant reduction in germination rates compared to the control group. In contrast, at a lower concentration of 2 mM, only minor inhibition was observed, suggesting that while copper is an essential nutrient, excessive amounts can jeopardize seed development. Future studies could focus on strategies to mitigate copper toxicity, such as using soil amendments or selecting plant species that exhibit copper tolerance. Gaining insights into the effects of heavy metals on plants is vital for promoting sustainable agriculture and effective environmental management.

ACKNOWLEDGMENT

The authors extend their heartfelt thanks to Prof. Poonam Tandon (V.C. Of D.D.U. Gorakhpur University Gorakhpur) and my respected supervisor Dr. Kumari Sunita (Assistant Professor) for support and guidance.

CONFLICT OF INTEREST

The authors declare that they have NO conflict of interest.

REFERENCES

1. Aebi, H., (1984). Catalase in vitro. Methods Enzymology 105, 121–126.
2. Antreich S. (2012). Heavy metal stress in plants - a closer look. Protocol of the project practicum "Heavy metal stress in plants, University of Vienna, 1-13.
3. AOSA. (1983). Seed Vigor Hand Testing Book, pp. 122-128. Contribution No. 32 to the Handbook on Seed Testing. Association of Official Seed Analysis. Springfield, USA.
4. Abdul-Baki AA, Anderson JD (1973). Vigor determination in soybean seed by multiple criteria 1. Crop Science 13: 630-633
5. Li, Q., Chen, H. H., Qi, Y. P., Ye, X., Yang, L. T., Huang, Z. R. & Chen, L. S. (2019). Excess copper effects on growth, uptake of water and nutrients, carbohydrates, and PSII photochemistry revealed by OJIP transients in citrus seedlings. Environmental Science and Pollution Research, 26(29), 30.188–30.205. DOI: https://doi.org /10.1007/s11356-019-06170-2
6. Asada, K., (2006). Production and scavenging of reactive oxygen species in chloroplasts and their functions. Plant Physiology 141, 391–396

7. Asgharipour MR, Rafiei M. (2011). Effect of salinity on germination and seedling growth of lentils. Aust J Basic App Sci 5(11).2002-2004
8. Wang et al. (2004). Copper-induced stress and antioxidative responses in roots of *Brassica juncea* L. Botanical Studies—Academia Sinica 45, 203–212
9. Sarvajeet Singh, Nafees A. Khan, Rahat Nazar and Naser A. Anjum. (2008). Photosynthetic Traits and Activities of Antioxidant Enzymes in Blackgram (*Vigna mungo* L. Hepper) Under Cadmium Stress. American Journal of Plant Physiology 3, 25–32
10. Ahsan N, Lee DG, Lee SH, Kang KY, Lee JJ, Kim PJ, Yoon HS, Kim JS, Lee BH. (2007). Excess copper induced physiological and proteomic changes in germinating rice seeds. Chemosphere.;67(6):1182-93. doi: 10.1016/j.chemosphere.2006.10.075. Epub 2006 Dec 19. PMID: 17182080.