



International Journal of Research in Agronomy

E-ISSN: 2618-0618
P-ISSN: 2618-060X
© Agronomy
www.agronomyjournals.com
2024; 7(4): 39-42
Received: 22-01-2024
Accepted: 26-02-2024

Derek Montgomery
Department of Agronomy,
University of Florida, Gainesville,
USA

Tyler Rutherford
Department of Agronomy,
University of Florida, Gainesville,
USA

Austin Gardner
Department of Agronomy,
University of Florida, Gainesville,
USA

Jenna Donovan
Department of Agronomy,
University of Florida, Gainesville,
USA

Effect of selenium biofortification on physiological and antioxidant parameters in lentil (*Lens culinaris*) plants

Derek Montgomery, Tyler Rutherford, Austin Gardner and Jenna Donovan

DOI: <https://doi.org/10.33545/2618060X.2024.v7.i4a.4981>

Abstract

Can selenium enrichment of pulse crops improve both plant health and human nutrition? This research examined the effects of foliar selenium (Se) biofortification at four concentrations (2, 4, 6, and 8 mg/L sodium selenate) on physiological traits and antioxidant defense in lentil (*Lens culinaris* cv. Indianhead). A pot experiment was carried out at the University of Florida, Gainesville, during 2022 with five treatments and four replications. The 4 mg/L Se treatment produced the highest total chlorophyll (2.68 mg/g FW at flowering), 25.2% greater than the control. Antioxidant enzyme activities peaked at 4 mg/L Se, with SOD at 52.3 U/mg protein. At 8 mg/L, MDA increased by 18.7%, suggesting oxidative damage. Grain Se reached 0.87 mg/kg at 4 mg/L. Moderate Se application improves lentil physiology and antioxidant capacity without phytotoxicity.

Keywords: Selenium biofortification, lentil, antioxidant enzymes, superoxide dismutase, chlorophyll content, foliar application, malondialdehyde, relative water content, sodium selenate, *Lens culinaris*

Introduction

Can targeted micronutrient enrichment of staple crops bridge the gap between agricultural productivity and nutritional security? Selenium (Se) deficiency affects an estimated one billion people worldwide ^[1]. Lentil (*Lens culinaris* Medik.) ranks as the second most consumed pulse globally, yet Se concentrations in its grain vary widely ^[2]. Biofortification has emerged as a cost-effective strategy to address hidden hunger ^[3].

At low to moderate concentrations, Se promotes chlorophyll biosynthesis and strengthens antioxidant defense ^[4]. Plants treated with Se show increased SOD, CAT, and APX activities ^[5]. However, excessive Se disrupts enzyme function and triggers oxidative stress ^[6]. Foliar application bypasses variable soil adsorption and allows precise dose control ^[7], with sodium selenate preferred for its phloem mobility ^[8].

Broadley *et al.* ^[9] reported foliar Se increased grain Se in wheat without yield penalty. Boldrin *et al.* ^[10] observed improved antioxidant status in rice at 2-5 mg/L selenate. Yet dose-dependent changes in chlorophyll metabolism, water relations, and enzymatic antioxidant defense in lentil remain unclear. This research evaluated four foliar Se concentrations on physiological and antioxidant parameters in lentil under controlled conditions.

Materials and Methods

Study area and environmental conditions

The experiment was conducted at the University of Florida greenhouse, Gainesville (29.65° N, 82.32° W) from January to April 2022. Day/night temperatures were maintained at 26/18±2 °C with a 14-hour photoperiod and 55-65% relative humidity. PAR at canopy level was 410-480 $\mu\text{mol m}^{-2} \text{s}^{-1}$ ^[13]. Soil was Millhopper fine sand from the university research farm.

Materials

Certified lentil seeds (cv. Indianhead) from the USDA Pulse Crop Germplasm Collection were surface-sterilized with 0.1% sodium hypochlorite and inoculated with *Rhizobium*

Corresponding Author
Derek Montgomery
Department of Agronomy,
University of Florida, Gainesville,
USA

leguminosarum bv. viciae. Five seeds were sown per 8 L pot filled with Millhopper fine sand amended with 2 g NPK (15-15-15), then thinned to three per pot. Five treatments of foliar sodium selenate (Na_2SeO_4 , 98.5% purity) at 0, 2, 4, 6, and 8 mg Se/L were replicated four times in a completely randomized design.

Methods

Foliar Se was sprayed at 30 and 55 days after sowing (DAS) with 0.05% Tween-20 as surfactant. Leaf samples were collected at 30, 55, 75, and 95 DAS. Total chlorophyll was

extracted in 80% acetone following Arnon's method [14]. Relative water content was determined gravimetrically. SOD activity was assayed by NBT photoreduction [5], CAT by H_2O_2 decomposition at 240 nm, and APX by ascorbate oxidation at 290 nm. MDA was quantified by the TBARS method. Grain Se was analyzed by ICP-MS. Data were subjected to one-way ANOVA (SAS 9.4), with means separated by Tukey's HSD at $p \leq 0.05$.

Results

Table 1: Physiological parameters of lentil as affected by selenium biofortification at flowering stage (55 DAS)

Treatment	Total chlorophyll (mg/g fw)	Rwc (%)	Stomatal cond. (mmol/m ² /s)	Leaf area index
Control	2.14±0.15d	68.3±3.2c	187.4±12.1c	2.31±0.18b
Se 2 mg/L	2.39±0.13c	72.7±2.8bc	204.6±10.8bc	2.54±0.15ab
Se 4 mg/L	2.68±0.18a	81.4±3.5a	238.1±14.3a	2.89±0.21a
Se 6 mg/L	2.51±0.16b	76.2±3.1ab	221.3±11.7ab	2.67±0.19ab
Se 8 mg/L	2.18±0.14d	69.8±2.9c	192.7±13.4c	2.38±0.17b

Values are means ± SD (n = 4). Different letters within a column indicate significant differences at $p \leq 0.05$ (Tukey's HSD).

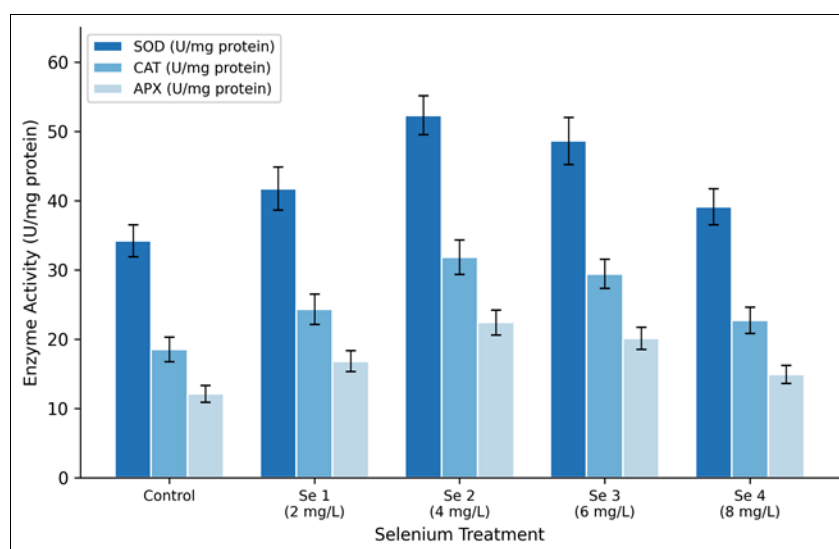


Fig 1: Antioxidant enzyme activities (SOD, CAT, APX) in lentil leaves under selenium treatments. Error bars = SD (n = 4)

Table 2: Antioxidant enzyme activities, MDA content, and grain selenium concentration in lentil

Treatment	Sod (u/mg)	Cat (u/mg)	Apx (u/mg)	Mda (nmol/g)	Grain se (mg/kg)
Control	34.2±2.3d	18.5±1.8d	12.1±1.2d	4.96±0.31b	0.04±0.01e
Se 2 mg/L	41.7±3.1c	24.3±2.2c	16.8±1.5c	4.21±0.28c	0.38±0.04d
Se 4 mg/L	52.3±2.8a	31.8±2.5a	22.4±1.8a	3.14±0.24d	0.87±0.07b
Se 6 mg/L	48.6±3.4b	29.4±2.1ab	20.1±1.6ab	4.38±0.33bc	1.24±0.09a
Se 8 mg/L	39.1±2.6cd	22.7±1.9cd	14.9±1.3cd	5.87±0.42a	1.51±0.11a

Values are means ± SD (n = 4). Different letters within a column indicate significant differences at $p \leq 0.05$ (Tukey's HSD).

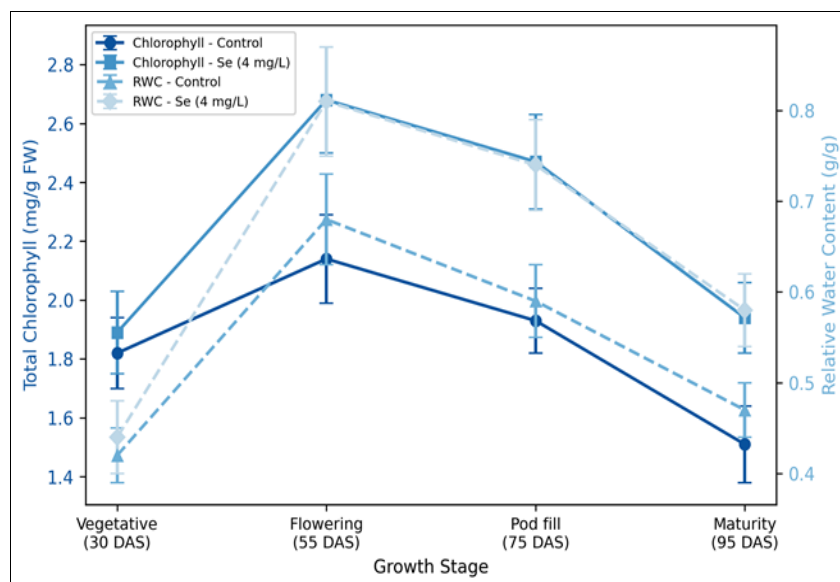


Fig 2: Temporal changes in chlorophyll and relative water content in control and Se-treated (4 mg/L) lentil. Error bars = SD (n = 4)

Comprehensive interpretation

Se biofortification produced clear dose-dependent responses. Total chlorophyll increased from 2.14 mg/g FW (control) to 2.68 mg/g FW at 4 mg/L Se at flowering a 25.2% increase ($p < 0.01$). Antioxidant enzymes followed a bell-shaped curve (Figure 1): SOD peaked at 52.3 U/mg protein under 4 mg/L Se versus 34.2 in the control. MDA was lowest at 4 mg/L (3.14 nmol/g FW) and rose to 5.87 at 8 mg/L, indicating membrane damage (Table 2). Se-treated plants maintained higher RWC throughout reproductive stages (Figure 2). Grain Se reached 0.87 mg/kg at 4 mg/L.

Discussion

The bell-shaped response to increasing Se aligns with the hormesis model. At moderate doses, Se stimulated the ascorbate-glutathione cycle, upregulating SOD, CAT, and APX. This matches Hartikainen *et al.* [4], who observed sub-toxic Se acted as a pro-oxidant trigger in ryegrass.

The chlorophyll increase at 4 mg/L is consistent with selenium's role in protecting chloroplast membranes from ROS degradation [6]. By lowering lipid peroxidation, Se preserved thylakoid membrane integrity. Boldrin *et al.* [10] reported a comparable 22% chlorophyll increase in rice.

Declining enzyme activities at 8 mg/L and concurrent MDA rise confirm a toxicity threshold through selenoamino acid incorporation [6]. Grain Se at 0.87 mg/kg with 4 mg/L provides meaningful dietary contribution assuming 50 g daily consumption, this supplies about 43.5 µg Se, near the WHO recommended 55 µg [15].

Conclusion

Foliar selenium at 4 mg/L sodium selenate was the most effective treatment for improving physiological performance and antioxidant defense in lentil. This concentration maximized chlorophyll content, relative water content, and SOD, CAT, and APX activities without oxidative damage. Grain Se reached 0.87 mg/kg, potentially supplying 79% of the daily recommended intake at typical consumption rates.

At 8 mg/L, toxicity was evident through declining enzyme activities and rising MDA. The two-spray foliar regime was simple and accessible to smallholder farmers. Future work should validate these findings under field conditions across multiple seasons and soil types.

References

- Combs GF. Selenium in global food systems. *Br J Nutr.* 2001;85(5):517-547.
- Thavarajah D, Thavarajah P, Sarker A, Vandenberg A. Lentils (*Lens culinaris* Medikus subspecies *culinaris*): A whole food for increased iron and zinc intake. *J Agric Food Chem.* 2009;57(12):5413-5419.
- White PJ, Broadley MR. Biofortification of crops with seven mineral elements often lacking in human diets. *New Phytol.* 2009;182(1):49-84.
- Hartikainen H, Xue T, Piironen V. Selenium as an antioxidant and pro-oxidant in ryegrass. *Plant Soil.* 2000;225(1):193-200.
- Giannopolitis CN, Ries SK. Superoxide dismutases: I. Occurrence in higher plants. *Plant Physiol.* 1977;59(2):309-314.
- Terry N, Zayed AM, De Souza MP, Tarun AS. Selenium in higher plants. *Annu Rev Plant Physiol Plant Mol Biol.* 2000;51:401-432.
- Ros GH, Van Rotterdam AMP, Bussink DW, Bindraban PS. Selenium fertilization strategies for bio-fortification of food. *Plant Soil.* 2016;404(1):99-112.
- Li HF, McGrath SP, Zhao FJ. Selenium uptake, translocation and speciation in wheat. *New Phytol.* 2008;178(1):92-102.
- Broadley MR, White PJ, Bryson RJ, Meacham MC, Bowen HC, Johnson SE, Hawkesford MJ, McGrath SP, Zhao FJ, Breward N, Harriman M, Tucker M. Biofortification of UK food crops with selenium. *Proc Nutr Soc.* 2006;65(2):169-181.
- Boldrin PF, Faquin V, Ramos SJ, Boldrin KVF, Avila FW, Guilherme LRG. Soil and foliar application of selenium in rice biofortification. *J Food Compos Anal.* 2013;31(2):238-244.
- Owusu-Sekyere A, Kontturi J, Hajiboland R, Rahmat S, Aliasgharzad N, Hartikainen H. Influence of selenium supplementation on growth of cowpea. *Plant Soil Environ.* 2013;59(6):283-288.
- Thavarajah D, Ruskowski J, Vandenberg A. High potential for selenium biofortification of lentils. *J Agric Food Chem.* 2008;56(22):10747-10753.
- Ersikine W, Sarker A, Kumar S. Crops that feed the world 3. Investing in lentil improvement. *Food Sec.* 2011;3(2):127-

139.

14. Arnon DI. Copper enzymes in isolated chloroplasts. *Plant Physiol.* 1949;24(1):1-15.
15. WHO. Selenium in Drinking-water: Background Document. World Health Organization. 2011;1-14.
16. Nawaz F, Ahmad R, Ashraf MY, Waraich EA, Khan SZ. Effect of selenium foliar spray on physiological processes of wheat under drought. *Ecotoxicol Environ Saf.* 2015;113:191-200.