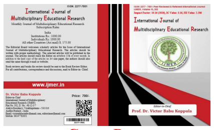

Cover Page

INTERNATIONAL JOURNAL OF MULTIDISCIPLINARY EDUCATIONAL RESEARCH
ISSN:2277-7881(Print); IMPACT FACTOR :10.16(2026); IC VALUE:5.16; ISI VALUE:2.286
PEER REVIEWED AND REFEREED INTERNATIONAL JOURNAL
(Fulfilled Suggests Parameters of UGC by IJMER)
Volume:15, Issue:3(5), March 2026
Scopus Review ID: A2B96D3ACF3FEA2A
Article Received: Reviewed: Accepted
Publisher: Sucharitha Publication, India
Online Copy of Article Publication Available: www.ijmer.in

Zinc-Induced Oxidative Stress And Nitrogen Metabolism Alterations In *Cajanus Cajan*.L And *Sorghum Bicolor*.L

[1]A.Satya Gowri Parvathi and [2]B.Sujathi

[1]Lecturers In Botany, SKR Government Degree College, Rajamahendravaram, Andhra Pradesh, India

[2]Professor, Department of Botany, Andhra University, Visakhapatnam, Pradesh, India

Abstract

This study investigates the impact of zinc stress on lipid peroxidation and nitrogen metabolism in red gram (Cajanus cajan) and Jowar (Sorghum bicolor) plants. Results show that proline accumulation increases with rising zinc concentrations, with the highest values recorded in Cajanus leaves. The findings suggest that proline plays a crucial role in plant defense mechanisms under zinc stress. The study provides insights into the physiological responses of these plants to zinc stress, highlighting potential strategies for improving crop tolerance to environmental contamination. The results have implications for agricultural practices and crop management in zinc-contaminated soils.

Key words: Accumulation, lipid peroxidation, proline, contamination, Zinc

Introduction

Zinc (Zn) is an essential micronutrient required for normal plant growth, functioning as a structural and catalytic component of numerous enzymes and proteins and playing a key role in gene expression and nitrogen metabolism (Alloway, 2008; Cakmak, 2000). However, excessive Zn accumulation in agricultural soils has emerged as a serious environmental concern due to industrial activities, mining, sewage sludge application, wastewater irrigation and intensive fertilizer use (Khan et al., 2008; Zhang et al., 2010). Elevated Zn levels disrupt cellular homeostasis and induce oxidative stress through enhanced production of reactive oxygen species (ROS), resulting in membrane damage and metabolic dysfunction. Lipid peroxidation is a major consequence of Zn toxicity and serves as a reliable indicator of oxidative damage in plants. Excess Zn also interferes with protein synthesis and accelerates protein degradation, thereby affecting plant growth and productivity (Palma et al., 2002). Recent studies have further highlighted that Zn-induced stress alters antioxidant metabolism and cellular redox balance, leading to species- and organ-specific physiological responses (Othman, 2024; Singh et al., 2025). Proline accumulation is a common adaptive response of plants to heavy metal stress and plays a crucial role in osmotic adjustment, membrane stabilization and ROS detoxification (Saradhi et al., 1993; Sharma & Dietz, 2006). Enhanced proline accumulation under Zn and other heavy metal stresses has been widely reported as a protective mechanism (Ahsan et al., 2025). Understanding such biochemical responses is essential for improving crop tolerance in metal-contaminated soils. Therefore, the present study evaluates Zn-induced changes in lipid peroxidation, protein metabolism and proline accumulation in *Cajanus cajan* and *Sorghum bicolor*.

Materials and methods

Field preparation and experimental design

The district regional climate is Agro-Ecological Sub Region on Eastern Coastal plain, hot sub-humid to the semi-arid ecoregion (12.1, 18.4). Agro-Climatic Region East Coast plain and hill region XI). Geographic coordinates of district latitude 16o 58' 60"N longitude 18o 46' 60" E altitude 13m AMSL. Rajahmundry is an industrial area surrounding villages that mainly depend on agriculture. The stone crushers and smaller industries caused heavy metal pollution along with automobile exhausts and affect the agricultural crops. In the East Godavari district, mainly cultivated crops are Rice, Maize, Red gram *Sorghum*, Sugar cane, and some other vegetable crops.

Six fields (three fields for *Cajanus* and three for *Sorghum*) were taken based on the Zn concentration. Out of the six fields, two fields had high Zn concentrations belong to Area-1 (10 km distance from the industries), having ~50 ppm(50ppm) of Zn, and two fields belong to Area-2 (5 km distance from the industries), having ~100ppm of Zn. Reaining two fields were normal area (30-50 km from industries.), having ~ 2ppm Zn which is located in Rampachodavaram rural areas, is considered as ontrolled field. Remaining elements all are adequate in range in each field, but only Zn was high level in those four fields. Based on this reason further research was conducted for Zn effect on plant growth and development. Experimental field was ploughed with a tractor and harrowed before seed sowing.

Plant material

Two types of plants Pigeonpea (*Cajanus cajan* (L.) Millspaugh) and *Sorghum bicolour* (L) were selected for the investigation. The seeds were obtained from agriculture research stations Rampachodavaram and Rajahmundry east Godavari district Andhra Pradesh. The seeds are placed into the furrows at a specific distance manually by a man working behind a plough. In present experimental fields depth of seed sowing is 25mm in *Sorghum* and 5cm in *Cajanus*.

1. Lipid peroxidation

The lipid peroxidation of *Cajanus* and *Sorghum* plants of control and contaminated areas plant material were determined as a measure of malondialdehyde formation which is a product of lipid peroxidation. The procedure was a minor modification of the method adopted by Heath and Packer (1968). One g of plant material was macerated in 5 ml of 0.1% TCA. The homogenate was centrifuged at 10,000 x g for 5 min. To one ml of the aliquot of the supernatant 4 ml of 20% TCA containing 0.5%, thiobarbituric acid was added. The mixture was heated at 950C for 30 min and then quickly cooled on an ice bath. The mixture was centrifuged at 10,000 x g for 15 min and the absorbance of the supernatant was measured at 532 nm and the value for the non-specific absorption at 600 nm was subtracted.

The concentration of malondialdehyde was calculated using an extinction coefficient of ΣM = 155mM -1cm-1µ moles.

1.1. Total Lipids

Extraction and estimation: Total lipid extraction was carried out according to the method of Bligh and Dyer (1959).

The total lipid of each sample was dissolved in chloroform, made up to 5 ml, and was preserved at 00C in a tightly stoppered standard flask until further analysis. The total lipid content was expressed as mg per gram dry weight.

2. **Nitrogen Metabolism**

2.1. Total Proteins

Total protein content was estimated by the method of Lowry *et al.,* (1951).

2.2 Proline

Proline content was estimated by the acid ninhydrin method of Troll and Lindsley (1955) as modified by Tully *et al.,* (1979).

Statistical Data Analysis

The physiological, biochemical parameters, antioxidant metabolism and protein contents were the mean of independent experimental replicates (n =3). Means and Standard errors were carried out for each treatment. All the results furnished have been critically analyzed on SPSS statistical tool, which thoroughly calculated Means followed by standard error.

Cover Page

INTERNATIONAL JOURNAL OF MULTIDISCIPLINARY EDUCATIONAL RESEARCH
ISSN:2277-7881(Print); IMPACT FACTOR :10.16(2026); IC VALUE:5.16; ISI VALUE:2.286
PEER REVIEWED AND REFEREED INTERNATIONAL JOURNAL
(Fulfilled Suggests Parameters of UGC by IJMER)
Volume:15, Issue:3(5), March 2026
Scopus Review ID: A2B96D3ACF3FEA2A
Article Received: Reviewed: Accepted
Publisher: Sucharitha Publication, India
Online Copy of Article Publication Available: www.ijmer.in

Results

Lipid Peroxidation

The formation of malondialdehyde is a measure of lipid peroxidation. Therefore lipid peroxidation was determined in relation to the amount of malondialdehyde formed in all the parts of *Cajanus* and *Sorghum* grown under Zn stress. The enhancement in the levels of malondialdehyde content was observed in the roots of two plants with increasing age of the plants as well as with increasing Zn concentrations. Moreover, malondialdehyde content of the roots of the two plants registered higher values at 100ppm Zn when compared to their controls. The changes in the levels of lipid peroxidation product of the stems and leaves Zn stressed plants showed a trend similar to the roots both increasing age and with increasing Zn concentrations (table 1).

The malondialdehyde content of the *Cajanus* leaf part an increase was recorded as 1.58,2.27 folds in pre flowering stage, 1.72, 1.85 folds in flowering stage and 1.97,2.59 in post flowering stage 50 and 100ppm Zn concentrations respectively it was Significant at $p \leq 0.01$. In *Cajanus* stem an increase was recorded as 1.02, 1.25 folds in pre flowering stage, 1.16, 1.61 folds in flowering stage and 1.55, 3.52 in post flowering stage 50 and 100ppm Zn concentrations respectively. The same pattern was followed in *Cajanus* root also, here an increase was recorded as 1.49, 1.64 folds in pre flowering stage, 1.24, 1.46 folds in flowering stage and 1.12, 1.16 in post flowering stage 50 and 100ppm Zn concentrations respectively.

In *Sorghum* leaf part an increase was recorded as 1.19, 1.29 folds in pre flowering stage, 1.25, 1.38 folds in flowering stage and 1.61, 1.71 folds in post flowering stage 50 and 100ppm Zn concentrations respectively it was Significant at $p < 0.05$. In *Sorghum* stem an increase was recorded as 1.18, 1.84 folds in pre flowering stage, 1.18, 1.63 folds in flowering stage and 1.10, 1.79 folds in post flowering stage 50 and 100ppm Zn concentrations respectively. The same pattern was followed in *Sorghum* root also, here an increase was recorded as 1.11, 1.25 folds in pre flowering stage, 1.06, 1.10 folds in flowering stage and 1.01, 1.70 folds in post flowering stage 50 and 100ppm Zn concentrations respectively.

In between the two plants, the lipid peroxidation of membrane was more active in *Cajanus* than *Sorghum* in the response to Zn stress.

Total Lipids

Total lipids of *Cajanus* and *Sorghum* plants were recorded in three plant stages (pre flowering, flowering and post flowering) and three different contaminated areas (control, 50ppm and 100ppm). Lipids content was gradually increased from 2ppm to 100ppm in both *Cajanus* and *Sorghum* plants. This trend was followed in three plant stages. The total lipid content of two plant parts leaf, stem and root increasing slightly with increasing age of plants and as well as increasing Zn concentrations. However, the lipid content in roots registered lower values when compare to leaf and stem.

In *Cajanus* leaf part an increase was recorded as 1.06, 1.31 folds in pre flowering stage, 1.04, 1.24 folds in flowering stage and 1.08, 1.10 folds in post flowering stage 50 and 100ppm Zn concentrations respectively. In *Cajanus* stem an increase was recorded as 1.24, 1.33 folds in pre flowering stage, 1.09, 1.15 folds in flowering stage and 1.03, 1.09 in post flowering stage 50 and 100ppm Zn concentrations respectively. The same pattern was followed in *Cajanus* root also, here an increase was recorded as 1.05, 1.12 folds in pre flowering stage, 1.12, 1.22 folds in flowering stage and 1.16, 1.18 in post flowering stage 50 and 100ppm Zn concentrations respectively (table.2).

On the other hand in *Sorghum* also followed the same pattern for lipids. In *Sorghum* leaf part an increase was recorded as 1.27, 1.30 folds in pre flowering stage, 1.27, 1.40 folds in flowering stage and 1.21, 1.53 in post flowering stage 50 and 100ppm Zn concentrations respectively. In *Sorghum* stem an increase was recorded as 1.05, 1.15 folds in pre flowering stage, 1.08, 1.09 folds in flowering stage and 1.06, 1.10 in post flowering stage 50 and 100ppm Zn concentrations respectively. The same pattern was followed in *Sorghum* root also, here an increase was recorded as 1.03, 1.05 folds in pre flowering stage, 1.03, 1.09 folds in flowering stage and 1.05, 1.07 folds in post flowering stage 50 and 100ppm Zn concentrations respectively (table 2).

Cover Page

INTERNATIONAL JOURNAL OF MULTIDISCIPLINARY EDUCATIONAL RESEARCH
ISSN:2277-7881(Print); IMPACT FACTOR :10.16(2026); IC VALUE:5.16; ISI VALUE:2.286
PEER REVIEWED AND REFEREED INTERNATIONAL JOURNAL
(Fulfilled Suggests Parameters of UGC by IJMER)
Volume:15, Issue:3(5), March 2026
Scopus Review ID: A2B96D3ACF3FEA2A
Article Received: Reviewed: Accepted
Publisher: Sucharitha Publication, India
Online Copy of Article Publication Available: www.ijmer.in

Nitrogen metabolism

Total Proteins

Total proteins of *Cajanus* and *Sorghum* plants were recorded in three plant stages (pre flowering, flowering and post flowering) and three different contaminated areas 2, 50 and 100ppm Zn concentrations. Total protein content was gradually decreased from 50 to100ppm Zn concentrations when compare to their respective their controls in both of the plants *Cajanus* and *Sorghum*. This trend was followed in three plant stages.

In *Cajanus* leaf part the decrease was recorded as 1.03and 1.31folds in pre flowering stage, 1.14 and 1.52 folds in flowering stage and folds in post flowering stage 50 and 100ppm Zn concentrations respectively. In *Cajanus* stem the decrease was recorded as 1.26,1.72 folds in pre flowering stage 1.12,1.32 folds in flowering stage and 1.13,1.44 folds in post flowering stage 50 and 100ppm Zn concentrations respectively. The same pattern was followed in *Cajanus* root also, here the decrease was recorded as 3.24,4.69 folds in pre flowering stage, 1.67,1.96 in flowering stage and 1.30,1.74 in post flowering stage 50 and 100ppm Zn concentrations respectively (table.3)

On the other hand in *Sorghum* also followed the same pattern for total proteins. In *Sorghum* leaf part the decrease was recorded as 1.15,1.32 folds in pre flowering stage, 1.19,1.7 folds in flowering stage and 1.84,3.08 in post flowering stage 50 and 100ppm Zn concentrations respectively. In *Sorghum* stem the decrease was recorded as 1.15,1.60 folds in pre flowering stage, 1.24,1.79 folds in flowering stage and 1.11,1.21 folds in post flowering stage 50 and 100ppm Zn concentrations respectively. The same pattern was root also here the decrease was recorded 1.53,2.54 folds in pre flowering stage, 1.47,2.11 folds in flowering stage and 1.28,1.34 folds in post flowering stage 50 and 100ppm Zn concentrations respectively (table.3).

The protein content was registered *Cajanus* root when compared to *Sorghum*. Total protein contents was more affect at 100ppm Zn than 2 and 50ppm.

Proline content

Proline content of *Sorghum* and *Cajanus* plants were recorded in three plant stages (pre flowering, flowering and post flowering) and three different contaminated areas (control, 50ppm and 100ppm). Proline content was gradually increased from 2ppm to 100ppm fields. This trend was followed in three plant stages in both *Cajanus* and *Sorghum* plants.

In *Cajanus* leaf part an increase was recorded as 1.10, 1.11 folds in pre flowering stage, 1.00, 1.01 folds in flowering stage and 1.06, 1.07 in post flowering stage 50 and 100ppm Zn concentrations respectively. In *Cajanus* stem an increase was recorded as 1.02, 1.07 folds in pre flowering stage, 1.02, 1.05 folds in flowering stage and 1.02, 1.13 in post flowering stage 50 and 100ppm Zn concentrations respectively. The same pattern was followed in *Cajanus* root also, here an increase was recorded as 1.00, 1.07 folds in pre flowering stage, 1.09, 1.18 folds in flowering stage and 1.17, 1.27 in post flowering stage 50 and 100ppm Zn concentrations respectively (table.4**).**

On the other hand in *Sorghum* also followed the same pattern for proline content. The part of increase was recorded as 1.17, 1.25 folds in pre flowering stage, 1.06, 1.08 folds in flowering stage and 1.05, 1.08 in pre flowering stage 50 and 100ppm Zn concentrations respectively. In *Sorghum* stem an increase was recorded as 1.23, 1.35 folds in pre flowering stage, 1.02, 1.07 folds in flowering stage and 1.16, 1.32 in post flowering stage 50 and 100ppm Zn concentrations respectively. The same pattern was followed in *Sorghum* root also, here an increase was recorded as 1.03, 1.01 folds in pre flowering stage, 1.02, 1.06 folds in flowering stage and 1.09, 1.11 in post flowering stage 50 and 100ppm Zn concentrations respectively (table.4).

The Correlation coefficients *Cajanus* and *Sorghum* plants was depended on the Zn concentration. The three stages significantly varied with increasing Zn concentration on Proline, Lipid peroxidase and Total lipid shown strong positive correlation with increasing Zn concentration, whereas Total protein shown strong negative correlation. The results were shown in (Table: 5-10)

Cover Page

INTERNATIONAL JOURNAL OF MULTIDISCIPLINARY EDUCATIONAL RESEARCH
ISSN:2277-7881(Print); IMPACT FACTOR :10.16(2026); IC VALUE:5.16; ISI VALUE:2.286
PEER REVIEWED AND REFEREED INTERNATIONAL JOURNAL
(Fulfilled Suggests Parameters of UGC by IJMER)
Volume:15, Issue:3(5), March 2026
Scopus Review ID: A2B96D3ACF3FEA2A
Article Received: Reviewed: Accepted
Publisher: Sucharitha Publication, India
Online Copy of Article Publication Available: www.ijmer.in

Discussion

Lipid peroxidation

Lipid peroxidation is a reliable indicator of oxidative damage under heavy metal stress. In the present investigation, malondialdehyde (MDA) content increased significantly in the roots, stems and leaves of both *Cajanus cajan* and *Sorghum bicolor* with increasing plant age and zinc (Zn) concentrations. Among the two species, *Cajanus* exhibited comparatively higher membrane lipid peroxidation than *Sorghum*, indicating greater sensitivity to Zn stress. Roots were the most affected organs, particularly at 100 ppm Zn, suggesting direct metal accumulation and enhanced oxidative stress at the site of uptake.

Heavy metals, although essential in trace amounts, induce excessive production of reactive oxygen species (ROS) at higher concentrations, leading to membrane damage and cellular dysfunction (Kyunghee & Junghoon, 2001). Similar increases in membrane permeability and oxidative injury were reported in Zn-stressed cowpea leaves exposed to high irradiance conditions, confirming the combined effect of Zn toxicity and environmental stress on membrane stability (Michael et al., 2014). Further demonstrated that Zn-induced oxidative stress elevates MDA levels and disrupts membrane integrity in crop plants, highlighting the role of Zn in altering cellular redox balance (Singh et al., 2025).

Lipid peroxidation was highest during the flowering stage in both plants and gradually declined at the post-flowering stage, indicating a stage-specific vulnerability to Zn stress. Comparable results were observed under Pb and Cd stress in *Phaseolus vulgaris*, where excess metal exposure enhanced MDA production in a concentration-dependent manner (Bharadwaj et al., 2009). The enhanced membrane lipid peroxidation under heavy metal stress has been attributed to oxidative degradation of membrane lipids, as reported earlier for Pb- and Cd-induced toxicity (Gallego et al., 2012). Recent reports also confirm that Zn-mediated oxidative damage is closely associated with increased ROS generation and lipid peroxidation in plants exposed to metal stress (Ahsan et al., 2025).

Lipids

The total lipid content in leaves, stems and roots of both plants showed a slight increase with advancing plant age and increasing Zn concentration. However, roots consistently recorded lower lipid levels compared to aerial parts, reflecting enhanced lipid degradation under Zn-induced oxidative stress. Alterations in lipid metabolism under heavy metal stress have been widely reported, with reductions in total lipids, phospholipids and sterols observed under Cu and Cd toxicity in pea, pepper and cucumber plants (Quartacci et al., 2000; Jemal et al., 2000; Janicka et al., 2008).

Recent studies indicate that Zn stress alters membrane lipid composition and fluidity, thereby affecting membrane-bound processes and cellular signaling (Zinc toxicity in plants, 2024). The observed variation in lipid content in the present study suggests that Zn exposure modulates lipid biosynthesis and degradation depending on plant species, organ type and developmental stage.

Proteins

Protein content exhibited a declining trend with increasing Zn concentration in both plant species. Higher protein levels were recorded at lower Zn concentrations, while significant reductions were observed at 50 and 100 ppm Zn, particularly in roots. This decrease may be attributed to the binding of Zn ions to sulfhydryl groups of proteins, resulting in structural modifications and enzyme inactivation (Zengin & Kirbag, 2007).

Maximum protein content was observed during the flowering stage in control plants, whereas Zn-treated plants consistently showed lower protein levels. The reduction was more pronounced in *Cajanus* than in *Sorghum*, indicating species-specific

Cover Page

INTERNATIONAL JOURNAL OF MULTIDISCIPLINARY EDUCATIONAL RESEARCH
ISSN:2277-7881(Print); IMPACT FACTOR :10.16(2026); IC VALUE:5.16; ISI VALUE:2.286
PEER REVIEWED AND REFEREED INTERNATIONAL JOURNAL
(Fulfilled Suggests Parameters of UGC by IJMER)
Volume:15, Issue:3(5), March 2026
Scopus Review ID: A2B96D3ACF3FEA2A
Article Received: Reviewed: Accepted
Publisher: Sucharitha Publication, India
Online Copy of Article Publication Available: www.ijmer.in

tolerance mechanisms. Similar decreases in protein content under Zn and Cu stress have been reported in *Vigna radiata* (Mittal et al., 2009). Some investigations suggest that excess Zn disrupts protein synthesis and accelerates proteolytic activity under oxidative stress conditions (Othman, 2025).

Proline

Proline accumulation increased progressively with rising Zn concentrations from 2 ppm to 100 ppm in both *Cajanus* and *Sorghum*. The highest proline levels were recorded in leaves, followed by stems and roots, with maximum accumulation during the flowering stage. Proline content declined at the post-flowering stage, indicating its role as a stress-responsive osmoprotectant during active growth phases. Enhanced proline accumulation under environmental contamination has been reported in *Philadelphus coronarius* leaves (Kafel et al., 2010). Proline acts as an osmolyte, ROS scavenger and stabilizer of proteins and membranes under metal stress. Zn stress significantly enhances proline biosynthesis as part of the plant's adaptive defense mechanism (Parlak & Yilmaz, 2012; Singh et al., 2025). Increased proline accumulation under heavy metal stress has been widely reported across diverse plant species, reinforcing its role as a universal stress marker (Tripathi & Gaur, 2004; Balestrasse et al., 2005; Sun et al., 2007).

Conclusions.

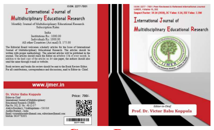
The present study demonstrated that zinc (Zn) stress significantly influenced lipid peroxidation, total lipid content and proline accumulation in *Cajanus cajan* and *Sorghum bicolor*. Lipid peroxidation and total lipid activity were more pronounced in *Cajanus* than in *Sorghum*, indicating greater sensitivity of *Cajanus* to Zn-induced oxidative stress. Among the plant organs, roots were the most severely affected, particularly at 100 ppm Zn, highlighting their primary role in metal uptake and stress response. Lipid peroxidation reached its maximum during the flowering stage in both plant species and gradually declined at the post-flowering stage, suggesting stage-dependent susceptibility to Zn toxicity. Proline accumulation was markedly higher at 100 ppm Zn during the flowering stage in both plants, with increased levels observed in stems and roots; however, leaves consistently exhibited the highest proline content. These findings indicate that proline plays a protective role in mitigating Zn-induced stress, particularly during critical growth stages.

Overall, the results emphasize species- growth stage-specific responses to Zn stress and provide insights into the physiological and biochemical mechanisms underlying Zn tolerance in agricultural plants

Table.1

Lipid peroxidation of *Cajanus* and *Sorghum* Leaf, Stem and Root at three stages
(MEAN OF 3 REPLICATIONS ± S.E)

	Zn concentration ppm	Lipid peroxidation (µ moles MDA/gm FW)		
Leaf		**Pre flowering**	**flowering**	**Post flowering**
Cajanus	**2**	15.29±0.05	19.61±0.27	17.42±0.21
	50	24.19±0.18	33.87±0.83	32.45±0.46
	100	34.84±0.13	36.32±0.28	45.16±0.41
Sorghum	**2**	15.35±0.06	19.10±0.36	25.23±0.27
	50	18.39±0.09	23.94±0.21	40.65±0.35
	100	19.81±0.15	26.52±0.19	43.23±0.28
Stem				


Cover Page

INTERNATIONAL JOURNAL OF MULTIDISCIPLINARY EDUCATIONAL RESEARCH
ISSN:2277-7881(Print); IMPACT FACTOR :10.16(2026); IC VALUE:5.16; ISI VALUE:2.286
PEER REVIEWED AND REFEREED INTERNATIONAL JOURNAL
(Fulfilled Suggests Parameters of UGC by IJMER)
Volume:15, Issue:3(5), March 2026
Scopus Review ID: A2B96D3ACF3FEA2A
Article Received: Reviewed: Accepted
Publisher: Sucharitha Publication, India
Online Copy of Article Publication Available: www.ijmer.in

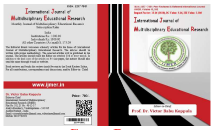
Cajanus	**2**	33.81±0.26	27.61±0.26	10.84±0.31
	50	34.77±0.33	32.26±0.32	16.84±0.27
	100	42.32±0.13	44.52±0.84	41.42±0.28
Sorghum	**2**	25.94±0.15	32.19±0.28	35.74±0.64
	50	30.71±0.09	38.26±0.31	39.48±0.25
	100	47.81±0.18	52.52±0.34	64.13±0.22
Root				
Cajanus	**2**	43.10±0.19	58.84±0.28	55.23±0.18
	50	64.45±0.18	73.16±0.25	62.39±0.82
	100	70.97±0.15	86.39±0.19	64.13±0.17
Sorghum	**2**	47.74±0.45	51.87±0.42	56.19±0.26
	50	53.16±0.49	55.42±0.18	57.16±0.73
	100	60.13±0.29	57.10±0.73	69.03±0.17

Table.2

Total Lipids of *Cajanus* and *Sorghum* Leaf, Stem and Root at three stages

(MEAN OF 3 REPLICATIONS ± S.E)

		Total Lipid (% / dw)		
Leaf	Zn concentration ppm	**pre flowering**	**Flowering**	**post flowering**
Cajanus	**2**	2.61±0.03	2.95±0.07	2.73±0.02
	50	2.79±0.02	3.08±0.03	2.96±0.06
	100	3.43±0.02	3.67±0.05	3.03±0.02
Sorghum	**2**	1.85±0.02	1.93±0.07	1.92±0.06
	50	2.36±0.01	2.47±0.02	2.34±0.06
	100	2.41±0.05	2.72±0.02	2.95±0.03
Stem				
Cajanus	**2**	2.91±0.05	3.73±0.06	3.81±0.04
	50	3.63±0.07	4.07±0.03	3.94±0.02
	100	3.88±0.02	4.29±0.05	4.16±0.04
Sorghum	**2**	2.5±0.02	3.15±0.02	3.19±0.06
	50	2.64±0.02	3.42±0.02	3.4±0.08
	100	2.89±0.00	3.45±0.04	3.52±0.02
Root				
Cajanus	**2**	2.32±0.2	2.49±0.07	2.58±0.02


Cover Page

INTERNATIONAL JOURNAL OF MULTIDISCIPLINARY EDUCATIONAL RESEARCH
ISSN:2277-7881(Print); IMPACT FACTOR :10.16(2026); IC VALUE:5.16; ISI VALUE:2.286
PEER REVIEWED AND REFEREED INTERNATIONAL JOURNAL
(Fulfilled Suggests Parameters of UGC by IJMER)
Volume:15, Issue:3(5), March 2026
Scopus Review ID: A2B96D3ACF3FEA2A
Article Received: Reviewed: Accepted
Publisher: Sucharitha Publication, India
Online Copy of Article Publication Available: www.ijmer.in

	50	2.45±0.09	2.81±0.04	2.9±0.02
	100	2.62±0.05	2.95±0.02	3.05±0.07
Sorghum	**2**	1.88±0.03	1.99±0.06	2.84±0.05
	50	1.94±0.7	2.05±0.04	2.98±0.03
	100	1.99±0.08	2.17±0.07	3.05±0.00

Table.3
Total protein content of *Cajanus* and *Sorghum* Leaf, Stem and Root at three stages
(MEAN OF 3 REPLICATIONS ± S.E)

Leaf	Zn concentration ppm	**Protein concentration (µg/ml)**	**Protein concentration (µg/ml)**	**Protein concentration (µg/ml)**
Cajanus	**2**	597±2.34	819±1.83	435±1.91
	50	579±1.86	713±2.85	383±1.53
	100	453±1.37	536±1.74	315±1.84
Sorghum	**2**	471±1.76	640±1.93	383±1.01
	50	408±1.98	534±1.52	208±0.93
	100	355±2.15	435±1.83	124±0.84
Stem				
Cajanus	**2**	539±2.16	669±2.81	606±2.09
	50	425±2.18	597±2.01	532±1.93
	100	312±1.58	506±1.28	418±1.30
Sorghum	**2**	348±1.44	374±1.24	304±0.95
	50	302±1.35	300±0.98	273±0.89
	100	217±1.45	215±0.95	215±1.94
Root				
Cajanus	**Control**	380±1.33	534±1.62	568±1.93
	2	117±1.33	318±1.17	435±1.29
	50	81±1.85	272±2.81	326±1.05
Sorghum	**100**	430±1.84	640±2.91	675±2.08
	2	281±1.72	435±1.50	524±2.11
	50	169±0.94	303±1.81	502±1.95

Table.4

Proline concentration of *Cajanus* and *Sorghum* Leaf, Stem and Root at three stages
(Mean of 3 replications ± S.E)

		Proline content (µg/g FW)		
Leaf	Zn concentration ppm	**Pre Flowering**	**Flowering**	**Post flowering**
Cajanus	**2**	88.1±2.09	121.8±2.90	77.3±0.98
	50	97.3±2.01	123.1±3.01	82.5±1.06
	10	98.6±1.08	123.8±2.19	83.2±1.07
Sorghum	**2**	54.3±1.06	73.3±1.08	75.1±0.91
	50	63.8±1.05	78.3±1.07	79.3±0.89
	100	68.2±0.97	79.2±1.40	81.4±1.07
Stem				
Cajanus	**2**	32.8±0.98	52.7±1.09	28.8±0.98
	50	33.7±093	54.1±0.98	29.4±0.27
	10	35.2±0.33	55.6±1.00	32.7±0.31
Sorghum	**2**	29.8±0.45	48.3±1.09	22.4±0.48
	50	32.6±0.43	49.5±2.07	26.1±0.29
	100	24.1±0.08	52.1±1.08	29.6±0.89
Root				
Cajanus	**2**	38.6±0.94	49.2±0.98	33.6±0.91
	50	38.9±0.09	53.7±0.79	39.6±0.33
	10	41.4±0.19	58.1±0.32	42.8±0.36
Sorghum	**2**	42.5±1.09	55.4±0.19	44.2±0.45
	50	43.8±0.94	56.8±0.64	48.5±0.22
	100	43.9±0.91	59.2±0.26	49.2±0.74

Table. 5 Correlation coefficients between nitrogen metabolism and lipid peroxidation with increasing concentration of Zn in the leaf tissue of pre flowering stage *Cajanus* plant.

	Zn Conc	*Total Protein*	*Free Proline*	*Lipid Peroxidase*	*Total Lipid*
Zn Conc	1				
Total Protein	-0.917*	1			
Proline	0.907**	-0.666**	1		
Lipid Peroxidase	0.998*	-0.936**	0.884**	1	
Total Lipid	0.951*	-0.995	0.734*	0.965*	1

*Significant at p≤ 0.05
**Significant at p≤ 0.01

Table. 6 Correlation coefficients between nitrogen metabolism and lipid peroxidation with increasing concentration of Zn in the leaf tissue of pre flowering stage *Sorghum* plant.

	Zn Conc	*Total Protein*	*Free Proline*	*Lipid Peroxidase*	*Total Lipid*
Zn Conc	1				
Total Protein	-0.999*	1			
Free Proline	0.978**	-0.987**	1		
Lipid Peroxidase	0.979**	-0.987*	0.999**	1	
Total Lipid	0.904*	-0.924*	0.973*	0.972*	1

*Significant at p≤ 0.05
**Significant at p≤ 0.01

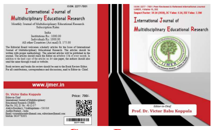
Table. 7 Correlation coefficients between nitrogen metabolism and lipid peroxidation with increasing concentration of Zn in the leaf tissue of flowering stage *Cajanus* plant.

	Zn Conc	*Total Protein*	*Free Proline*	*Lipid Peroxidase*	*Total Lipid*
Zn Conc	1				
Total Protein	-0.989*	1			
Proline	-0.804**	0.711	1		
Lipid Peroxidase	0.925**	-0.862**	-0.969**	1	
Total Lipid	0.938*	-0.978*	-0.549*	0.738*	1

*Significant at p≤ 0.05
**Significant at p≤ 0.01

Table .8 Correlation coefficients between nitrogen metabolism and lipid peroxidation with increasing concentration of Zn in the leaf tissue of flowering stage *Sorghum* plant.

	Zn Conc	*Total Protein*	*Free Proline*	*Lipid Peroxidase*	*Total Lipid*
Zn Conc	1				
Total Protein	-0.999**	1			
Free Proline	0.928*	-0.935**	1		


Cover Page

INTERNATIONAL JOURNAL OF MULTIDISCIPLINARY EDUCATIONAL RESEARCH
ISSN:2277-7881(Print); IMPACT FACTOR :10.16(2026); IC VALUE:5.16; ISI VALUE:2.286
PEER REVIEWED AND REFEREED INTERNATIONAL JOURNAL
(Fulfilled Suggests Parameters of UGC by IJMER)
Volume:15, Issue:3(5), March 2026
Scopus Review ID: A2B96D3ACF3FEA2A
Article Received: Reviewed: Accepted
Publisher: Sucharitha Publication, India
Online Copy of Article Publication Available: www.ijmer.in

Lipid Peroxidase	0.985*	-0.988**	0.978	1	
Total Lipid	0.978*	-0.982*	0.985*	0.999*	1

*Significant at p≤ 0.05
**Significant at p≤ 0.01

Table. 9 Correlation coefficients between nitrogen metabolism and lipid peroxidation with increasing concentration of Zn in the leaf tissue of post flowering stage *Cajanus* plant.

	Zn Conc	*Total Protein*	*Free Proline*	*Lipid Peroxidase*	*Total Lipid*
Zn Conc	1				
Total Protein	-0.997**	1			
Proline	0.915**	-0.882*	1		
Lipid Peroxidase	0.999*	-0.992*	0.934**	1	
Total Lipid	0.956*	-0.931	0.993*	0.969*	1

*Significant at p≤ 0.05
**Significant at p≤ 0.01

Table. 10 Correlation coefficients between nitrogen metabolism and lipid peroxidation with increasing concentration of Zn in the leaf tissue of post flowering stage *Sorghum* plant.

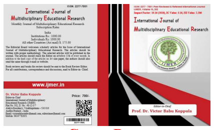
	Zn Conc	*Total Protein*	*Free Proline*	*Lipid Peroxidase*	*Total Lipid*
Zn Conc	1				
Total Protein	-0.98**	1			
Free Proline	0.982**	-0.999**	1		
Lipid Peroxidase	0.925**	-0.982*	0.979**	1	
Total Lipid	0.994*	-0.953*	0.956*	0.879*	1

*Significant at p≤ 0.05
**Significant at p≤ 0

References

1. Ahmad, P., & Jhon, R. (2009). Proline accumulation and antioxidative defense system in plants under stress. Botanical Research International, 2(4), 251–257.
2. Ahsan, M., Radicetti, E., Mancinelli, R., et al. (2025). *Alleviation of cadmium stress and improved growth performance of Catharanthus roseus by foliar application of zinc oxide nanoparticles.* South African Journal of Botany, 176, 129–140. https://doi.org/10.1016/j.sajb.2024.10.057.
3. Ahsan, M., Radicetti, E., Mancinelli, R., et al. (2025). Zinc-mediated alleviation of oxidative damage and proline metabolism under metal stress. South African Journal of Botany, 176, 129–140. https://doi.org/10.1016/j.sajb.2024.10.057
4. Alloway, B. J. (2008). Zinc in soils and crop nutrition (2nd ed.). International Zinc Association.
5. antioxidative enzymes. *Plant Growth Regulation, 55*(1), 1–10. https://doi.org/10.1007/s10725-008-9258-7.
6. Balestrasse, K. B., Gallego, S. M., & Tomaro, M. L. (2005). Cadmium-induced proline accumulation in nodules of soybean plants. *Plant Physiology and Biochemistry, 43*(7), 603–609. https://doi.org/10.1016/j.plaphy.2005.03.007.

7. Bharadwaj, L., Singh, S., & Sharma, R. K. (2009). Effect of lead and cadmium on lipid peroxidation and antioxidative enzymes in *Phaseolus vulgaris* L. *Journal of Environmental Biology, 30*(3), 415–420.
8. Cakmak, I. (2000). Possible roles of zinc in protecting plant cells from damage by reactive oxygen species. New Phytologist*, 146*(2), 185–205. https://doi.org/10.1046/j.1469-8137.2000.00630.x
9. Chen, C. T., Chen, T. H., & Lin, C. H. (2004). Proline accumulation and oxidative stress under heavy metal exposure. *Plant Science, 166*(3), 697–705. https://doi.org/10.1016/j.plantsci.2003.11.005
10. Dhir, B., Nasim, S. A., & Khan, M. I. R. (2014). Proline metabolism and metal tolerance in plants under cadmium stress. Environmental Science and Pollution Research*, 21*(18), 10740–10748. https://doi.org/10.1007/s11356-014-3094-6
11. Gallego, S. M., Pena, L. B., Barcia, R. A., Azpilicueta, C. E., Iannone, M. F., Rosales, E. P., Zawoznik, M. S., Groppa, M. D., & Benavides, M. P. (2012). Unravelling cadmium toxicity and tolerance in plants: Insight into regulatory mechanisms. *Environmental and Experimental Botany, 83*, 33–46. https://doi.org/10.1016/j.envexpbot.2012.04.006
12. Janicka, M., Reda, M., & Kabała, C. (2008). Effect of cadmium and copper stress on lipid composition in cucumber seedlings. *Biologia Plantarum, 52*(4), 703–708. https://doi.org/10.1007/s10535-008-0142-9
13. Jemal, F., Zarrouk, M., Ghorbal, M. H., & Guerfel, M. (2000). Changes in lipid composition of pepper plants in response to cadmium stress. *Plant Science, 152*(1), 47–53. https://doi.org/10.1016/S0168-9452(99)00207-9
14. Kafel, A., Bednarska, K., Augustyniak, M., & Witas, I. (2010). Proline accumulation as a response to environmental contamination in *Philadelphus coronarius* leaves. *EnvironmentalPollution,158*(10),3099–3104. https://doi.org/10.1016/j.envpol.2010.06.017
15. Karimi, P., Khavari-Nejad, R. A., Niknam, V., Ghahremaninejad, F., & Najafi, F. (2012). The effects of excess copper on proline, protein and lipid peroxidation in plants. *Plant Omics Journal, 5*(5), 455–460.
16. Kasai, K., Mori, N., & Nakamura, C. (1998). Changes in proline content and related enzyme activities under stress conditions. Plant Cell Physiology, *39*(5), 556–559. https://doi.org/10.1093/oxfordjournals.pcp.a029401
17. Khan, S., Cao, Q., Zheng, Y. M., Huang, Y. Z., & Zhu, Y. G. (2008). Health risks of heavy metals in contaminated soils and food crops irrigated with wastewater in Beijing, China. Environmental Pollution*, 152*(3), 686–692. https://doi.org/10.1016/j.envpol.2007.06.056
18. Kyunghee, J., & Junghoon, K. (2001). Heavy metal stress and antioxidative responses in plants. *Journal of Plant Biology, 44*(3), 193–200. https://doi.org/10.1007/BF03030320.
19. Michael, P. I., Inbaraj, M. P., & Selvaraj, T. (2014). Zinc-induced oxidative stress in cowpea (*Vigna unguiculata*) under high irradiance conditions. *Environmental and Experimental Botany, 100*, 54–62. https://doi.org/10.1016/j.envexpbot.2013.12.002.
20. Mittal, N., Thakur, S., & Verma, R. (2009). Effect of heavy metals on protein content in *Vigna radiata. Indian Journal of Plant Physiology, 14*(4), 395–399.
21. Nikolić, N., Kojić, D., Pilipović, A., Pajević, S., Krstić, B., Borišev, M., & Orlović, S. (2008). Responses of hybrid poplar to cadmium stress: Proline accumulation and
22. Noor, S., Ali, S., Abbas, F., et al. (2022). Heavy metal-induced toxicity and oxidative stress in plants: A critical review. Environmental Pollution*, 308*, 119632. https://doi.org/10.1016/j.envpol.2022.119632
23. Othman, E. Z. (2024). Zinc-induced regulation of antioxidant metabolism and stress tolerance in plants. Journal of Environmental Management, *349*, 119245. https://doi.org/10.1016/j.jenvman.2024.119245
24. Othman, E. Z. (2025). *Novel synergistic effect of zinc and manganese oxide nanoparticles on plant biochemical responses under stress.* Journal of Environmental Management, 355, 117696. https://doi.org/10.1016/j.jenvman.2025.117696.


Cover Page

INTERNATIONAL JOURNAL OF MULTIDISCIPLINARY EDUCATIONAL RESEARCH
ISSN:2277-7881(Print); IMPACT FACTOR :10.16(2026); IC VALUE:5.16; ISI VALUE:2.286
PEER REVIEWED AND REFEREED INTERNATIONAL JOURNAL
(Fulfilled Suggests Parameters of UGC by IJMER)
Volume:15, Issue:3(5), March 2026
Scopus Review ID: A2B96D3ACF3FEA2A
Article Received: Reviewed: Accepted
Publisher: Sucharitha Publication, India
Online Copy of Article Publication Available: www.ijmer.in

25. Palma, J. M., Sandalio, L. M., Corpas, F. J., Romero-Puertas, M. C., McCarthy, I., & del Río, L. A. (2002). Plant proteases, protein degradation and oxidative stress: Role of peroxisomes. Plant Physiology and Biochemistry*, 40*(6–8), 521–530. https://doi.org/10. 1016/S0981-9428(02)01405-6.
26. Parlak, K. U., & Yilmaz, D. D. (2012). Response of proline content in aquatic plants under zinc stress. *Biological Trace Element Research, 145*(3), 307–313. https://doi.org/10.1007/s12011-011-9193-3.
27. Quartacci, M. F., Pinzino, C., Sgherri, C. L. M., & Navari-Izzo, F. (2000). Lipid composition and antioxidative response of pea plants subjected to copper stress. *Plant Physiology, 123*(1), 109–116. https://doi.org/10.1104/pp.123.1.109.
28. Saradhi, P. P., Alia, Arora, S., & Prasad, K. V. S. K. (1993). Proline accumulation under heavy metal stress. Journal of Plant Physiology*, 141*(5), 617–620. https://doi.org/10.1016/S0176-1617(11)80494-3.
29. Schat, H., Sharma, S. S., & Vooijs, R. (1997). Heavy metal–induced accumulation of free proline in metal-tolerant and non-tolerant ecotypes. Physiologia Plantarum, *101*(3), 477–482. https://doi.org/10.1111/j.1399-3054.1997.tb01032.x.
30. Sharma, S. S., & Dietz, K. J. (2006). The significance of amino acids and amino acid-derived molecules in plant responses and adaptation to heavy metal stress. Journal of Experimental Botany*, 57*(4), 711–726. https://doi.org/10.1093/jxb/erj073.
31. Singh, T., Saffeullah, P., & Umar, S. (2025). *Foliar application of zinc oxide (ZnO) nanoparticles ameliorates growth, yield traits, osmolytes, cell viability, and antioxidant system of Brassica juncea under lead stress.* Chemosphere, 370, 143950. https://doi.org/10.1016/j.chemosphere. 2024.143950.
32. Singh, T., Saffeullah, P., & Umar, S. (2025). Foliar application of zinc oxide nanoparticles ameliorates oxidative stress and osmolyte accumulation in plants under heavy metal stress. Chemosphere, *370*, 143950. https://doi.org/10.1016/j.chemosphere. 2024.143950
33. Sun, R. L., Zhou, Q. X., Sun, F. H., & Jin, C. X. (2007). Antioxidative defense and proline accumulation in plants under heavy metal stress. *Chemosphere, 68*(5), 848–856. https://doi.org/10.1016/j.chemosphere.2007.01.013.
34. Tripathi, B. N., & Gaur, J. P. (2004). Relationship between copper- and zinc-induced oxidative stress and proline accumulation in plants*. Plant Physiology and Biochemistry, 42*(11), 925–932. https://doi.org/10.1016/j.plaphy.2004.09.007.
35. Zengin, F. K., & Kirbag, S. (2007). Effects of copper and zinc on protein content and antioxidative enzyme activities in *Phaseolus vulgaris* seedlings. *Journal of Environmental Biology, 28*(3), 451–456.
36. Zhang, X., Zhong, T., Liu, L., & Ouyang, X. (2010). Impact of soil heavy metal pollution on food safety in China. *PLoS ONE, 5*(8), e11822.https://doi.org/10.1371/journal.pone. 0011822.
37. *Zinc toxicity in plants: a brief overview on recent developments* (2024). In Zinc in Plants: Current Knowledge and Recent Advances (pp. 77-93). Elsevier.
38. Zinc toxicity in plants: Recent advances and future perspectives. (2024). In *Zinc in* plants: Current knowledge and recent advance*s* (pp. 77–95). Elsevier. https://doi.org/10.1016/B978-0-12-824448-1.00005-4.