FEDERAL UNIVERSITY WUKARI INTERNATIONAL JOURNAL OF BIOSCIENCES (FUWIJOB)
pISSN: 3121-9799, eISSN: 3121-973x; Volume 2, No. 2, June, 2026
https://fuwijob.com.ng

Enhancing Water Use Efficiency and Stress Tolerance in Roma VF Tomato Through Exogenous Proline Application

[1]Adelowo, J. M.*, [2]Olayinka, B. U., [3]Odoemene, S. N., [3]Josiah, O. M., [4]Morakinyo, A. E. & [2]Etejere, E. O.

[1]Department of Biotechnology and Molecular Biology, Federal University of Health Sciences, Ila Orangun

[2]Department of Plant Biology, University of Ilorin, Ilorin

[3]Department of Biological Sciences, Adeleke University, Ede, Osun State, Nigeria

[4]Department of Biochemistry, Adeleke University, Ede, Osun State, Nigeria

***Corresponding author**: joy.adelowo@fuhsi.edu.ng

ABSTRACT

Water scarcity during the dry season limits the continuous supply of fresh tomatoes. This study aimed to determine the optimal proline concentration for mitigating water stress in tomato plants while reducing irrigation water usage. Field experiments were conducted in two seasons, where tomato plants were subjected to water stress two weeks after transplanting and treated with foliar-applied proline at 0, 5, 10, and 15 mg/L. Unstressed treatment was included as a positive control. Field capacity was determined using the gravimetric method in order to accurately calculate the water required per plant viz: 2 L of water per plant was applied during the vegetative stage, while 3 L was applied during flowering and fruiting stages. The experiment followed a randomized complete block design with three replications. Results showed that water stress increased antioxidant enzyme activity, including glutathione reductase, catalase, and ascorbate peroxidase. Lycopene content was lowest in stressed plants without proline, while β-carotene was highest in stressed plants treated with 5 mg/L proline. The highest antioxidant enzyme activities were recorded in stressed plants treated with 15 mg/L proline. Although marketable yield was highest in unstressed plants, it was statistically comparable to that of stressed plants treated with 15 mg/L proline. This treatment also improved leaf relative water content, chlorophyll production, and lipid peroxidation control. These findings suggest that exogenous proline at 15 mg/L enhances tomato resilience to water stress and offers a sustainable approach to reducing irrigation water requirements while maintaining yield during dry seasons.

Keywords: proline, water stress, tomato, antioxidant enzyme, yield

INTRODUCTION

Tomato (*Solanum lycopersicum*) is one of the world's most vital crops widely recognized for its nutritional contributions to health (Waheed *et al*, 2020; Collins *et al*., 2022). It is rich in essential nutrients such as mineral, dietary fibres, non-enzymatic antioxidant including lycopene and beta-carotene, vitamins A, B and C thus making it a component of a healthy diet (Akinboye *et al*., 2024; Wang *et al*., 2023). *S. lycopersicum* can be processed into diverse products including soap, sauces, juice, and puree thereby enhancing its culinary versatility. There are several varieties of tomato, one of such varieties is Roma VF which is a determinate type that is characterized by its distinct pear shaped fruit (Kathimba *et al.*, 2021).

Tomato cultivation is significantly influenced by water availability as the crop requires substantial irrigation and is venerable to water stress (Alordzinu *et al.*, 2021, Alshami *et al.*, 2023). Water deficit is increasingly recognized as a critical factor affecting agricultural productivity as a result of inadequate rainfall and limited soil moisture during dry season (Bhattacharya, 2021). Due to increasing demand of water for household and industrial sector, the efficient management of irrigation water becomes essential to sustain crop yield (Ungureanu *et al.*, 2020)

Several researchers suggested that metabolic responses to water stress involve generation of by products such as malondialdehyde and various from of reactive oxygen species (ROS) (Hasanuzzaman et al., 2020; García-Caparrós *et al.*, 2021). These ROS can cause cellular damage thereby necessitating the presence of enzymatic antioxidant such as catalase (CAT), ascorbate peroxidase (APX), and glutathione (GR) which can combat the oxidative stress (Zandi and Schnug, 2022).

Growth regulators such as proline have emerged as vital component in modulating the plant defense response against oxidative stress (Zulfiqar and Ashraf, 2023). The interplay between the ascorbate-glutathione pathway and proline metabolism has been shown to enhanced tolerance to abiotic stress (Khalid *et al.*, 2022). When the activity of enzymatic antioxidant is increase, it can effectively prevent oxidative damage to plant during water deficit condition (Zulfiqar and Ashraf, 2023). Therefore, the aim of this research was to explore the potential of exogenously applied proline to improve the yield of tomato subjected to water deficit.

MATERIALS AND METHODS

The study was conducted during the dry seasons at the experimental farmland of Adeleke University, Ede, Osun State, Nigeria (Lat. 7°46'N, Long. 4°30'E; Alt. 275 ± 48 m). Roma VF tomato seeds were sourced from the National Horticultural Research Institute, Ibadan, and were sown in a nursery for four weeks. At transplanting, each seedling received 240 mL of a starter solution.

Before transplanting of tomato seedlings to plots, topsoil (0–15 cm) samples were collected, air-dried, sieved (2 mm), and analyzed for pH (1:2.5 soil:water), organic carbon (Hernandez *et al,* 2023), total nitrogen (Rutherford *et al.*, 2007), available phosphorus (Koralage *et al.*, 2015) , and texture (hydrometer method). Plots (2 m × 1.5 m) were prepared by mechanical hoeing with 0.5 m intra-row spacing and 1 m between treatments. The experiment followed a randomized block design with three replications.

Plants were subjected to water stress two weeks post-transplanting from nursery and irrigated at 7-day intervals, while unstressed plants received water every 2 days. There were five treatments which include: T1: Stressed + 5 mg/L proline; T2: Stressed + 10 mg/L proline; T3: Stressed + 15 mg/L proline; T4: Unstressed control and T5: Stressed + distilled water. Proline (J.T. Baker™) was foliar-sprayed (10 mL/plant) using a hand-held aerosol sprayer on 0, 14 and 28 after stress began.

Standard agronomic practices were uniformly applied. Weed control involved hoeing and hand-pulling. Irrigation was based on field capacity by gravimetric method (IAEA, 2008): 2 L/plant during vegetative stage, 3 L during flowering/fruiting.

Measurement of Parameter

Leaf relative water content (RWC) was measured using Barrs and Weatherley (1962) method.

Enzymatic Antioxidant Activities (EAA): Catalase (Aebi, 1984), ascorbate peroxidase (Nakano and Asada, 1981), and glutathione reductase (Massey and Williams, 1965) were measured at 14, 28, and 42 days post-stress.

Malondialdehyde (MDA): MDA was determined following the method of Hodges et al. (1999).

Beta-Carotene and Lycopene: The two carotenoids were quantified from mature fruits using Nagata and Yamashita (1992) method.

Fruit Yield: When all the treatments had one or more ripe fruit, yield per plant was weighed and converted to t/ha using**:**

$$\text{Fruit yield (t/ ha)} = \frac{Fruit\ weight\ persubplot(kg)\ X\ 10{,}000}{Size\ of\ subplot\ X\ 1000}$$

Where 10, 000 = size of hectare in m^2; 1,000 = conversion of kg to tonne

Data Analysis: The data were subjected to analysis of variance using SPSS-16.0 statistical analysis software (SPSS Inc., Chicago, IL, USA). Means were separated using Duncan's multiple range test (DMRT) at P <0.05%.

RESULTS AND DISCUSSION

The study investigated the effects of exogenous proline on tomato plants exposed to water deficit stress over two seasons. The experimental soil was a sandy loam type with slightly acidic pH, showed stable physical and chemical properties between seasons (Table 1), offering a consistent basis for treatment comparisons.

Table 1: Physico-chemical properties of the experimental surface of soil before cropping in 2018/2019 and 2019/2020 seasons

Season	Physical properties				Chemical properties				
	Sand (%)	Silt (%)	Clay (%)	Textural class	Organic content	pH	Nitrogen (%)	Phosphorus (mg/Kg)	Potassium (cmol/Kg)
1	65.00	25.00	10.00	Sandy loam	1.34	5.72	1.25	2.29	1.15
2	68.00	23.00	9.00	Sandy loam	1.58	5.66	1.32	2.27	1.17

Effect of Proline on Relative Water Content

Proline-treated plants, particularly at 15 mg/L (T3), consistently maintained higher RWC values across all in both seasons compared to other treatments (Table 2). While the stressed untreated plants (T5) had the lowest RWC, the highest was observed in unstressed plants (T4) and T3. The significance levels ($p < 0.05$) confirm that these differences are not due to random variation. The significant improvement in RWC across all developmental stages in proline-treated plants, particularly at 15 mg/L (T3), suggests that exogenous proline facilitates osmotic adjustment and water conservation under stress.

Table 2: Effects of application of exogenous proline on relative water content (%) of tomato plants during seasons 1 and 2

	Days after application of proline					
	Season 1			Season 2		
Treatments	14	28	42	14	28	42
T1	71.86±1.87^{c}	70.08±4.46^{b}	66.13±2.52^{b}	77.25±1.22^{c}	76.24±3.33^{b}	70.48±3.34^{ab}
T2	78.71±4.81^{bc}	76.88±2.16^{ab}	70.50±1.09^{a}	83.7±90.68^{b}	80.94±3.68^{ab}	74.97±3.54^{a}
T3	86.13±1.77^{ab}	83.72±3.51^{a}	72.18±3.21^{a}	87.31±1.06^{a}	86..70±0.67^{ab}	77.24±3.43^{a}
T4	89.25±2,03^{a}	84.28±2.42^{a}	72.50±1.43^{a}	89.58±0.21^{a}	87.42±3.96^{a}	79.76±3.39^{a}
T5	60.18±0.69^{d}	57.49±4.28^{c}	55.89±3.71^{b}	61.84±0.62^{d}	62.34±3.38^{c}	60.53±4.26^{b}
p-level	0.000	0.001	0.000	0.000	0.002	0.019

Each value represented Mean ± Standard error of mean. Similar letters are not statistically different at $p<0.05$ probability according to Duncan Multiple Range Test. (T_1= stressed plants that were sprayed 5 mg/L; T_2= stressed plants that were sprayed 10 mg/L of proline; T_3= stressed plants that were sprayed 15 mg/L of proline; T_4= unstressed plants (positive control) and T_5= stressed plants (negative control)

This is consistent with previous reports where proline accumulation was associated with enhanced cellular hydration, maintenance of turgor pressure, and reduced water loss during dehydration (Patanè et al., 2022). In contrast, untreated stressed plants (T5) exhibited the lowest RWC values, indicating severe water deficit stress and impaired water balance mechanisms (Bhattacharya and Bhattacharya, 2021).

Effect of Proline on Enzymatic Activities

From Table 3, T3 consistently exhibited the highest GR activity throughout all growth stages in both seasons, suggesting that higher concentrations of exogenous proline enhance the anti-oxidative defense system. Compared to T5, which showed moderate GR activity under stress, T3 indicates a stronger capacity to mitigate oxidative damage. Interestingly, T4 (unstressed control) exhibited lower GR activity. Results showed that T3 had significantly higher CAT activity, especially during the vegetative and flowering stages in Season 1, with diminishing significance in Season 2. This suggests that proline improves H_2O_2 detoxification, protecting plant cells from oxidative damage. The low CAT activity in T4 aligns with minimal oxidative stress in non-stressed plants, whereas T5 had moderately elevated CAT activity, insufficient to prevent oxidative damage, as evidenced by other stress markers. APX is another key enzyme in scavenging hydrogen peroxide. T3 plants displayed significantly elevated APX activity across all stages and seasons, supporting the notion that proline augments the enzymatic antioxidant system. The APX activity was markedly lower in T4 and T5, confirming that proline actively contributes to upregulating defense mechanisms against oxidative stress rather than stress alone triggering the response.

The high MDA in T1 and T2, though lower than T5, further demonstrates a dose-dependent protective role of proline, with 15 mg/L offering the most effective protection. The lowest MDA levels in T3-treated plants suggest that proline confers protection against membrane damage by enhancing antioxidant capacity and limiting ROS-induced peroxidation

Table 3: Effects of application of exogenous roline on glutathione reductase activity, catalase activity and ascorbate peroxidase activity of tomato plants during Seasons 1 and 2

	Days After application of proline					
	Season 1			Season 2		
Treatments	14	28	42	14	28	42
	Glutathione reductase activity (10^{-2} μ mol. mg^{-1} min^{-1})					
T1	0.27±0.03^{c}	0.25±0.23^{bc}	0.18±0.02^{c}	0.50±0.04^{ab}	0.46± 0.10^{a}	0.48±0.11^{ab}
T2	0.48±0.30^{b}	0.39±0.03^{b}	0.36± 0.06^{b}	0.56±0.06^{ab}	0.52±0.17^{a}	0.51±0.53^{ab}
T3	0.82±0.11^{a}	0.74±0.04^{a}	0.73±0.04^{a}	0.64±0.09^{a}	0.59±0.04^{a}	0.58±0.04^{a}
T4	0.17±0.03^{c}	0.14±0.01^{d}	0.13±0.02^{c}	0.12±0.01^{c}	0.13±0.02^{b}	0.11±0.02^{c}
T5	0.22±0.04^{c}	0.34±0.03^{c}	0.16±0.04^{c}	0.43±0.05^{b}	0.41±0.03^{ab}	0.34±0.02^{b}
p-level	<0.000	<0.000	<0.000	<0.000	0.044	0.003
	Catalase activity (10^{-2} μ mol. mg^{-1} min^{-1})					
T1	0.23± 0.08^{c}	0.27±0.01^{bc}	0.24±0.02^{b}	0.13±0.01^{ab}	0.10± 0.01^{ab}	0.10±0.01^{a}
T2	0.41±0.03^{b}	0.36± 0.02^{b}	0.34±0.02^{ab}	0.14±0.01^{ab}	0.12±0.01^{ab}	0.11±0.01^{a}
T3	0.58± 0.05^{a}	0.57±0.05^{a}	0.44±0.11^{a}	0.16±0.02^{a}	0.14±0.02^{a}	0.12± 0.01^{a}
T4	0.09±0.01^{d}	0.05± 0.02^{d}	0.04±0.02^{c}	0.09±0.19^{b}	0.08±0.02^{b}	0.07±0.01^{b}
T5	0.16±0.03^{cd}	0.18±0.02^{c}	0.16±0.02^{bc}	0.12± 0.02^{ab}	0.11± 0.02^{ab}	0.09±0.01^{ab}
p-level	<0.000	<0.000	0.004	0.180	0.129	0.034
	Ascorbate peroxidase activity (μ mol. $^{-}mg^{-1}$ min^{-1})					
T1	0.13± 0.03^{b}	0.08±0.01^{c}	0.08±0.02^{bc}	0.15±0.02^{b}	0.09±0.01^{c}	0.08±0.97^{bc}
T2	0.63± 0.08^{a}	0.42± 0.04^{b}	0.09±0.01^{b}	0.68± 0.05^{a}	0.45±0.02^{b}	0.16± 0.02^{b}
T3	0.64±0.09^{a}	0.64±0.12^{a}	0.62±0.04^{a}	0.70±0.06^{a}	0.69±0.11^{a}	0.67±0.07^{a}

T4	0.02±0.00^{b}	0.02± 0.01^{c}	0.02±0.01c	0.05 0.00^{b}	0.03±0.00^{c}	0.03± 0.00^{c}
T5	0.02±0.003^{b}	0.06± 0.01^{c}	0.04±0.01^{bc}	0.08± 0.00^{b}	0.07±0.00^{c}	0.09± 0.00^{bc}
p-level	<0.000	<0.000	<0.000	<0.000	<0.000	<0.000

Each value represented Mean ± Standard error of mean. Similar letters are not statistically different at $p<0.05$ probability according to Duncan Multiple Range Test. (T_1= stressed plants that were sprayed 5 mg/L; T_2= stressed plants that were sprayed 10 mg/L of proline; T_3= stressed plants that were sprayed 15 mg/L of proline; T_4= unstressed plants (positive control) and T_5= stressed plants (negative control)

The significant upregulation of key antioxidant enzymes GR, CAT, and APX in proline-treated plants supports the role of proline in activating the antioxidant defense system (Sachdev *et al.*, 2023). The observed increase in GR activity in T3 plants enhances the cellular glutathione pool, thereby maintaining redox homeostasis and facilitating ROS detoxification (Khalid *et al.*, 2022). Similarly, elevated CAT and APX activities aid in the breakdown of hydrogen peroxide which is a major ROS that accumulates under stress (Fujita and Hasanuzzaman, 2022). These findings are in line with prior studies showing that exogenous proline enhances the activity of enzymatic antioxidants, thereby reducing oxidative damage and improving stress resilience (Fujita and Hasanuzzaman, 2022).

Oxidative Stress and Antioxidant Enzymes

Table 4 showed that T3 treatment consistently had the lowest MDA values across all stages and seasons. This suggests that proline reduces membrane damage by quenching ROS. In contrast, T5 recorded the highest MDA, revealing severe oxidative damage in stressed, untreated plants.

Table 4: Effects of application of exogenous proline on MDA content (μmol./g fresh weight) of tomato plants during seasons 1 and 2

	Days after application of proline					
Treatments	Season 1			Season 2		
	14	28	42	14	28	42
T_1	0.47±0.00	0.36±0.01	0.22±0.01	0.47±0.01	0.37±0.00	0.24±0.01
T_2	0.48±0.01	0.35±0.01	0.22±0.01	0.48±0.02	0.37±0.00	0.23±0.01
T_3	0.03±0.01	0.06±0.02	0.02±0.00	0.03±0.02	0.03±0.01	0.02±0.00
T_4	0.14±0.02	0.12±0.01	0.10±0.02	0.13±0.01	0.14±0.03	0.14±0.03
T_5	0.57± 0.09	0.35 ±0.04	0.24 ±0.02	0.57 ±0.01	0.35 ±0.12	0.24 ±0.02
p-level	0.002	0.000	0.000	0.001	0.002	0.002

Each value represented Mean ± Standard error of mean. Similar letters are not statistically different at $p<0.05$ probability according to Duncan Multiple Range Test. (T_1= stressed plants that were sprayed 5 mg/L; T_2= stressed plants that were sprayed 10 mg/L of proline; T_3= stressed plants that were sprayed 15 mg/L of proline; T_4= unstressed plants (positive control) and T_5= stressed plants (negative control)

In contrast, the high MDA accumulation in T5 plants highlights the extent of cellular injury under unmanaged stress conditions (Atta *et al*., 2024).

T3-treated plants demonstrated increased activity of antioxidant enzymes including GR, CAT, and APX, contributing to enhanced ROS scavenging and cellular protection. These findings support earlier research that identified proline as a modulator of redox homeostasis and enzymatic defense (Fujita & Hasanuzzaman, 2022; Khalid et al., 2022; Sachdev et al., 2023). In contrast, stressed untreated plants (T5) showed insufficient enzymatic responses, leading to higher oxidative damage. A reduction in lipid peroxidation was evidenced by lower MDA levels in T3 plants, indicating effective protection against oxidative membrane damage (Atta et al., 2024). T5 plants recorded the highest MDA levels, reflecting pronounced cellular injury under stress.

Effect of Proline on Yield

Carotenoids such as lycopene and beta-carotene serve as antioxidants and are keys to fruit quality. No statistically significant difference was observed among treatments for lycopene and beta-carotene in Season 2, indicating that proline may not significantly alter these metabolites under field conditions. However, yield data show that T3 plants significantly outperformed other stressed groups (T1, T2, and T5), with yields approaching that of unstressed T4. This indicates that although proline may not enhance carotenoid biosynthesis, it significantly boosts yield under stress by improving physiological resilience and reducing oxidative damage.

Table 5: Effects of application of exogenous proline on carotenoid content (µmol./g fresh weigh) and yield (t/ha) of fruit of tomato plants during seasons 1 and 2

Treatments	Season 1			Season 2		
	Lycopene	Beta-carotene	Fruit yield	Lycopene	Beta-carotene	Fruit yield
T1	23.41 ± 0.14^{a}	2.56 ± 0.41^{a}	2.12 ± 0.58^{cd}	24.15 ± 0.40^{a}	2.63 ± 0.42^{a}	2.90 ± 0.52^{bc}
T2	25.27 ± 0.35^{a}	2.27 ± 0.09^{a}	3.99 ± 0.93^{bc}	25.41 ± 0.36^{a}	2.34 ± 0.28^{a}	5.31 ± 1.14^{ab}
T3	25.29 ± 0.13^{a}	2.19 ± 0.15^{a}	6.41 ± 0.72^{ab}	26.07 ± 0.50^{a}	2.26 ± 0.06^{a}	7.87 ± 1.37^{a}
T4	19.33 ± -0.52^{a}	2.10 ± 0.14^{a}	8.24 ± 1.49^{a}	19.99 ± 0.53^{a}	2.16 ± 0.11^{a}	8.36 ± 0.46^{a}
T5	27.87 ± 1.42^{a}	2.20 ± 0.29^{a}	0.85 ± 0.29^{d}	28.75 ± 1.62^{a}	2.27 ± 0.14^{a}	1.06 ± 0.22^{c}
p-level	0.000	0.000	0.001	0.728	0.695	0.001

Each value represented Mean ± Standard error of mean. Similar letters are not statistically different at $p<0.05$ probability according to Duncan Multiple Range Test. (T_1= stressed plants that were sprayed 5 mg/L; T_2= stressed plants that were sprayed 10 mg/L of proline; T_3= stressed plants that were sprayed 15 mg/L of proline; T_4= unstressed plants (positive control) and T_5= stressed plants (negative control)

Carotenoids, particularly lycopene and beta-carotene, are important antioxidants and indicators of fruit nutritional quality (Tufail *et al.*, 2024). While the differences in carotenoid content among treatments were not statistically significant in Season 2, proline-treated plants maintained higher carotenoid levels comparable to unstressed controls. This supported by the work of Xi *et al.* (2021) that stress causes an increase in carotenoids.

CONCLUSION

Exogenous application of proline, particularly at 15 mg/L, significantly enhances drought tolerance in tomato plants by improving water retention, augmenting antioxidant enzyme activities, reducing oxidative membrane damage, preserving photosynthetic pigments, and enhancing yield under stress conditions. These findings provide strong empirical support for the use of proline as a low-cost, effective strategy for enhancing abiotic stress tolerance in tomato production systems. Future studies should explore the molecular signaling pathways underlying proline-mediated stress responses and evaluate its long-term agronomic feasibility across different environmental conditions.

CONFLICT OF INTEREST

The authors declare that there was no known conflict of interest

REFERENCES

Aebi, H. (1984). *Catalase in vitro. In Methods in enzymology*. Academic press, pp 121-126. doi:10.1016/S0076-6879(84)05016-3

Akinboye, O. E., Afodu, O. J., Oyekale, O. O., Awoyinka, R. I. and Abolaji, A. O. (2024). Effects of four different processing methods on nutrients and bioactive profiles of tomato (Solanum lycopersicum L.). *Nigerian Journal of Agriculture and Agricultural Technology*, 4(3), 1–11. https://doi.org/10.59331/njaat.v4i3.770

Alordzinu, K. E., Jiuhao, L., Appiah, S. A., Aasmi, A. A. L., Blege, P. K. and Afful, E. A. (2021). Water stress affects the physio-morphological development of tomato growth. *African Journal of Agricultural Research*, 17(5), 733–742. https://doi.org/10.5897/AJAR2021.15450

Alshami, A. K., El-Shafei, A., Al-Omran, A. M., Alghamdi, A. G., Louki, I. and Alkhasha, A. (2023). Responses of tomato crop and water productivity to deficit irrigation strategies and salinity stress in greenhouse. *Agronomy*, 13(12), 3016. https://doi.org/10.3390/agronomy13123016

Atta, N., Shahbaz, M., Farhat, F., Maqsood, M. F., Zulfiqar, U., Naz, N., Ahmed, M. M., Hassan, N. U., Mujahid, N., Mustafa, A. E. Z. M. A., Elshikh, M. S. and Chaudhary, T. (2024). Proline-mediated redox regulation in wheat for mitigating nickel-induced stress and soil decontamination. *Scientific Reports*, 14(1), 456. https://doi.org/10.1038/s41598-024-51984-7

Barrs, H. D. and Weatherley, P.E. (1962) A Re-Examination of the Relative Turgidity Techniques for Estimating Water Deficits in Leaves. *Australian Journal of Biological Sciences*, 15, 413-428. http://dx.doi.org/10.1071/BI9620413

Bhattacharya, A. (2021). Effect of soil water deficit on growth and development of plants: A review. In Soil water deficit and physiological issues in plants. Springer. https://doi.org/10.1007/978-981-33-6276-5_5

Bhattacharya, A. and Bhattacharya, A. (2021). Effect of soil water deficit on growth and development of plants: A review. In Soil water deficit and physiological issues in plants (pp. 393–488). Springer. https://doi.org/10.1007/978-981-33-6441-0_14

Collins, E. J., Bowyer, C., Tsouza, A. and Chopra, M. (2022). Tomatoes: An extensive review of the associated health impacts of tomatoes and factors that can affect their cultivation. *Biology*, 11(2), 239. https://doi.org/10.3390/biology11020239

Fujita, M., and Hasanuzzaman, M. (2022). Approaches to enhancing antioxidant defense in plants. *Antioxidants*, 11(5), 925. https://doi.org/10.3390/antiox11050925

García-Caparrós, P., De Filippis, L., Gul, A., Hasanuzzaman, M., Ozturk, M., Altay, V. and Lao, M. T. (2021). Oxidative stress and antioxidant metabolism under adverse environmental conditions: A review. *Botanical Review*, 87, 421–466. https://doi.org/10.1007/s12229-020-09231-1

Hasanuzzaman, M., Bhuyan, M. H. M. B., Parvin, K., Bhuiyan, T. F., Anee, T. I., Nahar, K., Hossen, M. S., Zulfiqar, F., Alam, M. M. and Fujita, M. (2020). Regulation of ROS metabolism in plants under environmental stress: A review of recent experimental evidence. *International Journal of Molecular Sciences,* 21(22), 8695. https://doi.org/10.3390/ijms21228695

Hernandez , T. D. B, Slater, B. K., Shaffer J. M and Basta N. (2023). Comparison of methods for determining organic carbon content of urban soil in central Ohio. *Geodermal Regional*, 34: e00680. https://doi.org/10.1016/j.geodrs.2023.e00680

Hodges, D. M., DeLong, J. M., Forney, C. F. and Prange, R. K. 1999. Improving the thiobarbituric acid-reactive-substances assay for estimating lipid peroxidation in plant tissues containing anthocyanin and other interfering compounds. *Planta*, 207:604-611. doi:10.1007/s004250050524

International Atomic Energy Agency (IAEA) (2008). *Field Estimation of Soil Water Content: A Practical Guide to Methods, Instrumentation and Sensor Technology*. Retrieved from www.pub.iaea.org on January 2020

Kathimba, F. K., Kimani, P. M. Narla, R. D. and Kiirika, L. M. (2021). Characterization of tomato germplasm accessions for breeding research, *Journal of Agricultural Biotechnology and Sustainable Development*: 13(2):20-27. https://doi.org/10.589JABSD2021.0386

Khalid, M., Rehman, H. M., Ahmed, N., Nawaz, S., Saleem, F., Ahmad, S., Uzair, M., Rana, I. A., Atif, R. M., Zaman, Q. U. and Lam, H.-M. (2022). Using exogenous melatonin, glutathione, proline, and glycine betaine treatments to combat abiotic stresses in crops. *International Journal of Molecular Sciences*, 23(21), 12913. https://doi.org/10.3390/ijms232112913

Koralage, I S. A., Weersinghe, P., Silva, N. R. N and De Silva, C. S. (2015). The Determination of Available Phosphorus in Soil: a Quick and Simple Method. QUSL Journal, 8: 1-17. Retrieved from ouslj.sljol.info

Massey, V. and Williams, C.H. (1965). On the Reaction Mechanism of Yeast. Glutathione Reductase, *Journal of Biological Chemistry*, 240(11): 4470-4480.https://doi.org/10.1016/S0021-92258(18)9085-7

Nagata, M. and Yamashita, I. (1992). *Simple method for simultaneous determination of chlorophyll and carotenoids in tomato fruit.* Nippon Shokuhin Kogyo Gakkaishi 39:925-928.doi:10.3136/nskkk1962.39.925

Nakano, Y. and Asada, K. 1981. Hydrogen peroxide is scavenged by ascorbate-specific peroxidase in spinach chloroplasts. *Plant and cell physiology* 22:867-880. https://doi.org/10.1093/oxfordjournals.pcp.a076232

Patanè, C., Cosentino, S. L., Romano, D. and Toscano, S. (2022). Relative water content, proline, and antioxidant enzymes in leaves of long shelf-life tomatoes under drought stress and rewatering. *Plants*, 11(22), 3045. https://doi.org/10.3390/plants11223045

Rutherford, P. M., McGill, W. D., Arocena, J. M. and Figueiredo, C. T. (2007). Total Nitrogen. *Soil Sampling and Methods* 13: 239-241. https://doi/10.1201/9781420005271.ch22

Sachdev, S., Ansari, S. A. and Ansari, M. I. (2023). *ROS generation in plant cells orchestrated by stress. In Reactive oxygen species in plants: The right balance* (pp. 23–43). Springer. https://doi.org/10.1007/978-3-031-24894-7_2

Tufail, T., Bader Ul Ain, H., Noreen, S., Ikram, A., Arshad, M. T. and Abdullahi, M. A. (2024). Nutritional benefits of lycopene and beta-carotene: A comprehensive overview. Food Science & Nutrition, 12(11), 8715–8741. https://doi.org/10.1002/fsn3.4102

Ungureanu, N., Vlăduţ, V. and Voicu, G. (2020). Water scarcity and wastewater reuse in crop irrigation. *Sustainability*, 12(21), 9055. https://doi.org/10.3390/su12219055

Waheed, K., Nawaz, H., Hanif, M. A. and Rehman, R. (2020). Tomato. In Medicinal plants of South Asia: Novel sources for drug discovery (Chapter 46). Springer.

Wang, C., Li, M., Duan, X., Abu-Izneid, T., Rauf, A. and Khan, Z. (2023). Phytochemical and nutritional profiling of tomatoes; Impact of processing on bioavailability – A comprehensive review. *Food Reviews International*, 39(8), 5986–6010.https://doi.org/10.1080/87559129.2022.2097692

Xi, Y., Kong, F. and Chi, Z. (2021). ROS induce β-carotene biosynthesis caused by ~ changes of photosynthesis efficiency and energy metabolism in Dunaliella salina under stress conditions. *Frontiers in Bioengineering and Biotechnology,* 8, 613768. https://doi.org/10.3389/fbioe.2020.613768

Zandi, P. and Schnug, E. (2022). Reactive oxygen species, antioxidant responses and implications from a microbial modulation perspective. Biology, 11(2), 155. https://doi.org/10.3390/biology11020155

Zulfiqar, F. and Ashraf, M. (2023). Proline alleviates abiotic stress-induced oxidative stress in plants. Journal of Plant Growth Regulation, 42, 4629–4651. https://doi.org/10.1007/s00344-022-10839-3