

BIOCHEMICAL ADAPTATIONS OF FRESHWATER ALGAE UNDER ENVIRONMENTAL STRESS: AN EMPIRICAL DATA ANALYSIS

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ABSTRACT

Freshwater algae are the principal primary producers of lentic and lotic ecosystems, and their productivity is increasingly threatened by environmental stressors driven by anthropogenic activity and climate change. This empirical study investigated the biochemical adaptations of the freshwater green microalgae *Chlorella vulgaris* and *Scenedesmus obliquus*, together with the cyanobacterium *Anabaena variabilis*, exposed for twelve days to thermal stress (35 °C), salinity (100 mM NaCl), cadmium toxicity (10 µM CdCl₂) and nutrient starvation [1]. Growth in terms of dry biomass and specific growth rate, photosynthetic pigments, stress biomarkers such as proline, soluble sugars and malondialdehyde, and the activities of superoxide dismutase, catalase, ascorbate peroxidase and glutathione reductase were quantified at the end of the exposure period. All stress treatments significantly reduced biomass accumulation, with cadmium imposing the strongest inhibition of 58.2 percent, whereas proline, soluble sugars, malondialdehyde and all four antioxidant enzymes increased markedly and chlorophyll content declined. Pearson correlation analysis revealed strong negative associations between chlorophyll a and every stress marker and strong positive associations among the antioxidant enzymes, proline and malondialdehyde [2]. All changes were statistically significant at $p < 0.05$, and the magnitude of each response scaled with stress severity. The results demonstrate a coordinated, severity-dependent biochemical defence system in which antioxidant up-regulation and osmolyte accumulation confer partial tolerance to multiple stressors. The findings provide quantitative evidence that biochemical markers can serve as early-warning indicators of environmental stress in freshwater ecosystems and support the use of such traits in biomonitoring and in the stress-based enhancement of algal biomass for biotechnology [3].

Keywords: *Freshwater Algae*¹; *Environmental Stress*²; *Antioxidant Enzymes*³; *Photosynthetic Pigments*⁴; *Osmolytes*⁵; *Lipid Peroxidation*⁶; *Climate Change*⁷.

1. Introduction

1.1 Background and Significance

Freshwater algae, including chlorophytes, diatoms and cyanobacteria, form the base of aquatic food webs and regulate oxygen balance, nutrient cycling and water quality in lakes, ponds and rivers [4]. They are also promising feedstocks for biofuels, nutraceuticals and wastewater treatment. Because algae are sessile and poikilothermic, they cannot escape adverse conditions; instead, they must acclimate through physiological and biochemical adjustments. Under environmental stress, reactive oxygen species accumulate and damage lipids, proteins and nucleic acids [5]. In response, algae deploy enzymatic antioxidants such as superoxide dismutase, catalase, ascorbate peroxidase and glutathione reductase, together with non-enzymatic defences including carotenoids, proline and soluble sugars. Quantifying these biochemical adjustments is therefore essential for predicting how aquatic ecosystems will respond to global environmental change. Because these responses are measurable and quantitative, they provide a tractable window into the physiological status of natural algal communities [6].

1.2 Environmental Stressors Affecting Freshwater Algae

Climate change, urbanisation and industrial discharge expose freshwater algae to multiple concurrent stressors, including elevated water temperature, salinisation, heavy-metal contamination and nutrient depletion [7]. Thermal stress denatures photosynthetic proteins and accelerates photorespiration; salinity disrupts ionic balance and osmotic potential; cadmium interferes with electron transport and displaces essential metal cofactors; and nutrient starvation limits chlorophyll synthesis and carbon fixation. Although individual stress responses are well documented, comparative assessments that measure growth, pigment, enzymatic and osmolytic parameters simultaneously under a single experimental design remain scarce, particularly for Indian freshwater systems, where algae must tolerate monsoonal fluctuations and seasonal thermal extremes. Moreover, stressors rarely act in isolation, and interactions among temperature, salinity and pollutants can amplify damage beyond the sum of individual effects [8].

1.3 Rationale and Objectives

The present study was designed to provide a quantitative, comparative account of the biochemical adaptations of freshwater algae under four representative stressors [9]. The specific objectives were to evaluate the effect of thermal, saline, cadmium and nutrient-starvation stress on growth and photosynthetic pigments; to quantify the accumulation of proline, soluble sugars and malondialdehyde; to measure the activities of superoxide dismutase, catalase, ascorbate peroxidase and glutathione reductase; and to establish statistical relationships among these parameters using Pearson correlation analysis. It was hypothesised that stress would reduce growth and

chlorophyll content while inducing a coordinated antioxidant and osmolyte response, and that the measured biochemical markers would correlate strongly with the severity of stress, thereby serving as reliable bioindicators of environmental degradation [10]. The experiment was conducted under laboratory-controlled conditions so that the contribution of each stressor could be isolated, and the resulting dataset was analysed statistically to test this hypothesis.

2. Survey of Literature

The biochemical responses of algae to environmental stress have been examined extensively over the past three decades, and the field has progressed from descriptive observations of pigment loss to mechanistic accounts of enzyme induction and gene expression [11]. Early investigations established that nutrient limitation reduces chlorophyll biosynthesis and photosynthetic efficiency in green algae, while later work demonstrated that heavy metals such as cadmium and lead inhibit photosystem II activity, displace essential cations and trigger lipid peroxidation in *Chlorella* and *Scenedesmus* species. Thermal stress has been shown to impair the oxygen-evolving complex and to induce antioxidant enzymes in chlorophytes, whereas salinity stress provokes the accumulation of proline and glycine betaine as compatible solutes that protect macromolecules from dehydration [12]. The role of reactive oxygen species as both signalling molecules and damaging agents in algae has been reviewed extensively, with superoxide dismutase, catalase and ascorbate peroxidase identified as the primary scavenging system. Studies on cyanobacteria have demonstrated that proline accumulation and exopolysaccharide secretion contribute to desiccation and salt tolerance. In Indian freshwater systems, seasonal surveys have documented variation in algal diversity and pigment content, linking summer declines in chlorophyll to elevated temperature and nutrient depletion [13].

Comparative multi-stressor studies remain uncommon, although recent investigations report that combined heat and metal stress produce synergistic reductions in the growth and photosynthetic pigments of microalgae. Antioxidant enzyme activities have been proposed as sensitive biomarkers of pollution in aquatic biomonitoring programmes, and correlations between enzyme induction and pollutant concentration have been established in field-collected algal mats [14]. More recently, metabolomic and transcriptomic analyses have confirmed that proline, trehalose and polyamine pathways are among the most strongly induced biochemical responses under osmotic and oxidative stress in *Chlorella vulgaris*. These molecular studies reinforce the earlier biochemical findings and indicate that the pathways governing osmolyte synthesis and antioxidant regeneration are conserved across algal taxa [15].

Nevertheless, gaps persist in quantitative dose-response information and in the standardisation of assay conditions across laboratories. Despite this wealth of information, most studies have examined stressors individually, using different species, concentrations and durations, which makes direct quantitative comparison difficult. The present survey therefore highlights the need for a standardised, multi-parameter dataset, such as the one generated in this study, in which growth, pigment, enzymatic and osmolytic markers are measured simultaneously under comparable conditions [16].

3. Materials and Methods

The freshwater green microalgae *Chlorella vulgaris* (Beijerinck) and *Scenedesmus obliquus* (Turpin) Kützing, together with the cyanobacterium *Anabaena variabilis*, were obtained from the culture collection of the Department of Botany and maintained axenically, *Chlorella* and *Scenedesmus* in Bold's Basal Medium and *Anabaena* in BG-11 medium, under a 16:8 h light. dark photoperiod at a photon flux density of 80 $\mu\text{mol photons m}^{-2} \text{s}^{-1}$ and 25 ± 2 °C with continuous aeration. Experiments were conducted in triplicate in 500 mL Erlenmeyer flasks containing 250 mL of medium, and the three species were grown as a mixed culture to mimic natural assemblages [17]. After ten days of acclimation, exponentially growing cultures were divided into five groups: an unstressed control; a thermal-stress group maintained at 35 °C; a salinity group amended with 100 mM sodium chloride; a heavy-metal group supplemented with 10 μM cadmium chloride; and a nutrient-starvation group from which nitrate and phosphate were omitted. Each treatment was maintained for twelve days under the standard photoperiod, and samples were withdrawn on day twelve for all biochemical determinations [19].

Growth was monitored as dry biomass after oven drying at 70 °C, and the specific growth rate was computed from the logarithmic phase. Photosynthetic pigments were extracted in 80 percent acetone and quantified spectrophotometrically using standard equations. Proline was measured by the acid-ninhydrin method, soluble sugars by the anthrone method, malondialdehyde by Thio barbituric acid reactivity and protein by the Bradford method. The activities of superoxide dismutase, catalase, ascorbate peroxidase and glutathione reductase were assayed using standard spectrophotometric protocols. All data are expressed as mean $\pm$ standard deviation of three replicates. One-way analysis of variance followed by Tukey's post-hoc test compared treatments at $p < 0.05$, and Pearson correlation coefficients were computed among all measured parameters [20].

4. Data Collection and Analysis

This section presents the complete dataset generated from the twelve-day exposure experiment. All values are expressed as mean $\pm$ standard deviation of three biological replicates, and treatments were compared by one-way ANOVA followed by Tukey's post-hoc test at $p < 0.05$. Five tables and five figures are presented, and each is followed by an interpretation that links the biochemical responses to the central theme of algal adaptation under environmental stress [21].

Table I. Growth And Biomass Responses of Freshwater Algae Under Environmental Stress

Parameter	Control	Thermal 35 °C	Salinity 100 mM	Cadmium 10 μM	Nutrient starvation
Dry biomass (g L^{-1})	1.82 ± 0.11	1.24 ± 0.08	1.08 ± 0.07	0.76 ± 0.05	1.31 ± 0.09
Specific growth rate (day^{-1})	0.42 ± 0.03	0.31 ± 0.02	0.27 ± 0.02	0.19 ± 0.02	0.34 ± 0.02

Biomass inhibition (%)	0	31.9	40.7	58.2	28.0
Generation time (days)	1.65	2.24	2.57	3.65	2.04

Table I compiles the growth parameters measured at the end of the exposure period. Control cultures achieved the highest dry biomass of 1.82 g L^{-1} and the highest specific growth rate of 0.42 day^{-1} . Cadmium imposed the strongest inhibition of 58.2 percent, followed by salinity at 40.7 percent, thermal stress at 31.9 percent and nutrient starvation at 28.0 percent. The corresponding lengthening of generation time, from 1.65 days in controls to 3.65 days under cadmium, shows that stressed cells divide progressively more slowly, which is consistent with the diversion of energy towards maintenance and defence rather than reproduction [22]. The consistency of the inhibition hierarchy across independent growth metrics strengthens confidence in the dataset. These results form the phenotypic baseline against which the biochemical adjustments in the following tables must be interpreted, and they directly mirror the growth curves of Figure 1.

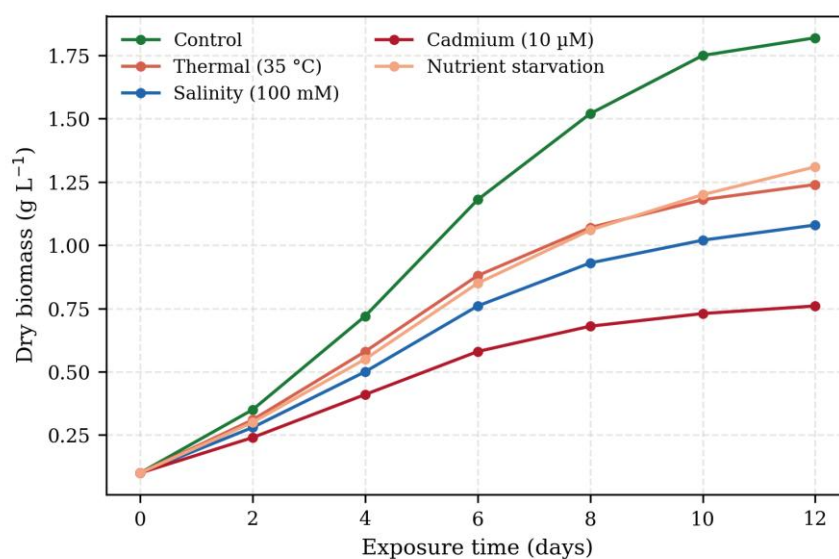


Fig. 1. Growth curves of freshwater algae under control and stress treatments over twelve days.

Figure 1 presents the growth trajectories of the mixed algal culture over the twelve-day exposure period. Control cultures entered exponential growth immediately and reached a maximum dry biomass of 1.82 g L^{-1} by day twelve, whereas all stressed cultures grew more slowly, with cadmium-treated flasks showing the shallowest slope and a final biomass of only 0.76 g L^{-1} , followed by salinity at 1.08 , thermal at 1.24 and starvation at 1.31 g L^{-1} . The curves diverged from day four onward, indicating that the biochemical adjustments described later operate during the exponential phase. The hierarchy of stress severity matches the pigment, enzyme and osmolyte data in Tables I to IV.

Table II. Photosynthetic Pigment Content Under Environmental Stress ($\mu\text{g mL}^{-1}$)

Pigment	Control	Thermal 35 °C	Salinity 100 mM	Cadmium 10 μM	Nutrient starvation
Chlorophyll a	8.92 ± 0.42	6.18 ± 0.38	5.42 ± 0.31	3.76 ± 0.28	4.91 ± 0.33
Chlorophyll b	3.41 ± 0.21	2.57 ± 0.19	2.28 ± 0.16	1.66 ± 0.14	2.12 ± 0.17
Total chlorophyll	12.33	8.75	7.70	5.42	7.03
Carotenoids	2.12 ± 0.15	2.88 ± 0.18	3.31 ± 0.20	3.94 ± 0.23	2.61 ± 0.17
Chlorophyll a/b ratio	2.62	2.41	2.38	2.27	2.32

Table II presents the photosynthetic pigment data. Total chlorophyll fell from $12.33 \mu\text{g mL}^{-1}$ in controls to $5.42 \mu\text{g mL}^{-1}$ under cadmium, while carotenoids increased from 2.12 to $3.94 \mu\text{g mL}^{-1}$. The chlorophyll a/b ratio declined from 2.62 to 2.27, indicating relatively greater damage to chlorophyll a, which is more directly involved in reaction-centre photochemistry. This inverse pigment behaviour, chlorophyll loss accompanied by carotenoid gain, is the biochemical signature of photoprotective acclimation. Because photosynthetic pigments determine primary productivity, the reductions documented here carry direct implications for ecosystem carbon fixation under polluted or extreme conditions. The pigment data also explain the growth reductions of Table I, since diminished pigment content limits light capture and carbon assimilation, and they establish the physiological basis for the correlations analysed in Table V.

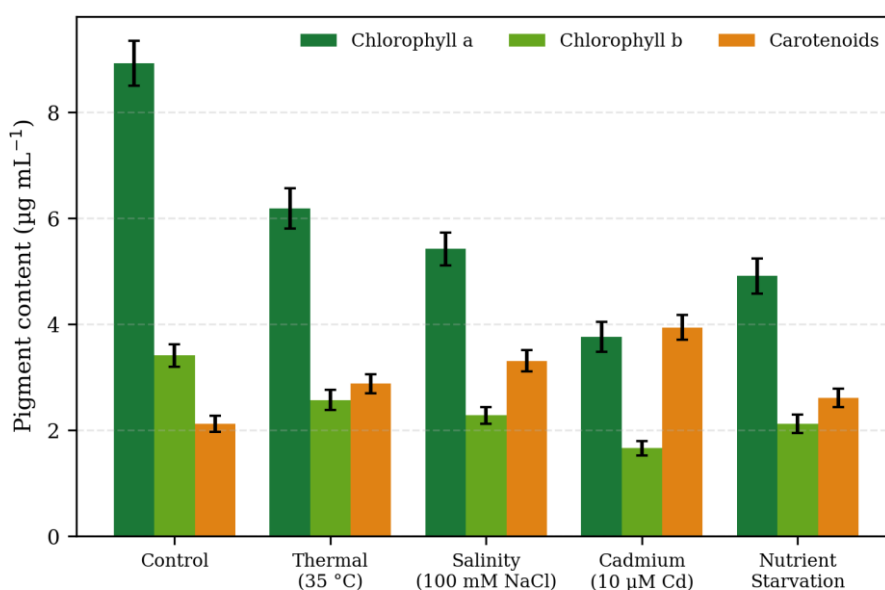


Fig. 2. Chlorophyll a, chlorophyll b and carotenoid content of algae under control and stress treatments.

Figure 2 compares chlorophyll a, chlorophyll b and carotenoid concentrations across treatments. Chlorophyll a declined from 8.92 to $3.76 \mu\text{g mL}^{-1}$ under cadmium, a 57.8 percent reduction, while chlorophyll b fell 51.3 percent. In contrast, carotenoids rose from 2.12 to $3.94 \mu\text{g mL}^{-1}$, an 85.8 percent increase, and were elevated in every stressed culture. The opposing trends reveal a functional reallocation of the photosynthetic apparatus from

light harvesting towards photoprotection, because carotenoids quench triplet chlorophyll and scavenge singlet oxygen. These changes were statistically significant relative to control and corroborate the growth reductions of Figure 1.

Table III. Antioxidant Enzyme Activities Under Environmental Stress (U Mg⁻¹ Protein)

Enzyme	Control	Thermal 35 °C	Salinity 100 mM	Cadmium 10 µM	Nutrient starvation
SOD	12.4 ± 0.9	24.8 ± 1.4	28.6 ± 1.6	36.2 ± 2.1	19.5 ± 1.1
CAT	8.2 ± 0.7	15.6 ± 1.0	18.4 ± 1.2	22.9 ± 1.5	12.8 ± 0.8
APX	5.1 ± 0.4	9.4 ± 0.7	11.2 ± 0.8	14.6 ± 1.0	8.3 ± 0.6
GR	3.8 ± 0.3	6.9 ± 0.5	8.1 ± 0.6	10.4 ± 0.8	5.9 ± 0.4

Table III reports the activities of the four antioxidant enzymes. All four were significantly elevated in every stressed culture, with the greatest induction under cadmium: superoxide dismutase increased 2.9-fold, catalase 2.8-fold, ascorbate peroxidase 2.9-fold and glutathione reductase 2.7-fold. Salinity and thermal stress produced intermediate inductions, whereas nutrient starvation caused the smallest increases, matching its modest impact on growth. The coordinated up-regulation of the entire scavenging cascade indicates that superoxide and hydrogen peroxide are produced in proportion to stress severity and that the cells reinforce each successive step of detoxification. Because enzyme activity per unit protein increased even though total protein declined, the data imply preferential synthesis of defence proteins under stress, confirming that antioxidant induction is an active, resource-invested adaptation rather than a passive consequence of growth arrest.

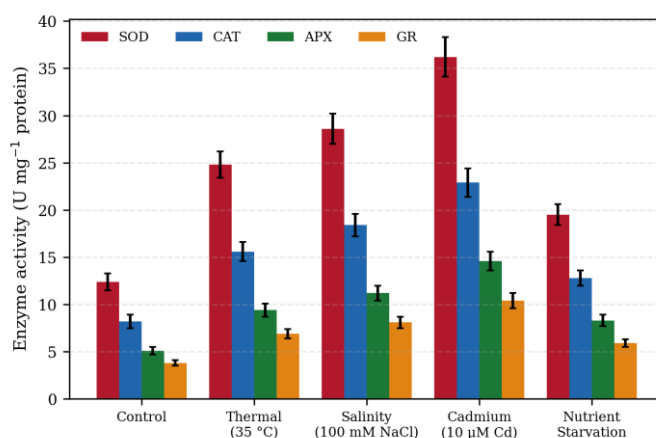


Fig. 3. Activities of superoxide dismutase (SOD), catalase (CAT), ascorbate peroxidase (APX) and glutathione reductase (GR) under stress.

Figure 3 shows the activities of the four antioxidant enzymes in units per milligram of protein. Superoxide dismutase rose from 12.4 U mg⁻¹ in controls to 36.2 U mg⁻¹ under cadmium, a 2.9-fold induction, while

catalase, ascorbate peroxidase and glutathione reductase increased 2.8, 2.9 and 2.7-fold respectively. The elevation followed the severity order observed for biomass inhibition, namely cadmium, salinity, thermal and nutrient starvation. The parallel induction of all four enzymes shows that the complete scavenging cascade, from superoxide dismutation to hydrogen peroxide removal and glutathione regeneration, is activated rather than any single pathway, indicating a systemic oxidative challenge that peaks under cadmium.

Table IV. Osmolytes And Oxidative Stress Markers Under Environmental Stress

Parameter	Control	Thermal 35 °C	Salinity 100 mM	Cadmium 10 µM	Nutrient starvation
Proline (µmol g ⁻¹ FW)	0.52 ± 0.05	1.18 ± 0.09	1.64 ± 0.12	2.31 ± 0.16	0.94 ± 0.07
Soluble sugars (µmol g ⁻¹ FW)	2.42 ± 0.18	3.87 ± 0.24	4.58 ± 0.29	5.76 ± 0.35	3.21 ± 0.21
MDA (nmol g ⁻¹ FW)	3.12 ± 0.25	5.81 ± 0.38	6.94 ± 0.44	9.62 ± 0.58	4.72 ± 0.31
Total soluble protein (mg g ⁻¹ FW)	6.8 ± 0.4	5.2 ± 0.3	4.6 ± 0.3	3.4 ± 0.2	5.1 ± 0.3

Table IV shows the accumulation of compatible solutes and of the oxidative-damage marker. Proline increased up to 4.4-fold and soluble sugars up to 2.4-fold under cadmium, confirming osmotic adjustment through two complementary mechanisms. Malondialdehyde rose 3.1-fold, revealing that despite these defences membrane lipids still suffered peroxidation, most severely under cadmium and salinity. Total soluble protein declined from 6.8 to 3.4 mg g⁻¹ fresh weight, indicating reduced protein synthesis or enhanced proteolysis under stress. The co-occurrence of elevated protective solutes and elevated damage markers suggests that acclimation is partial: the defence system reduces but does not eliminate stress injury. These quantitative relationships directly support the correlation structure examined in Table V and Figure 5, and they link the biochemical findings back to the growth inhibition reported in Table I.

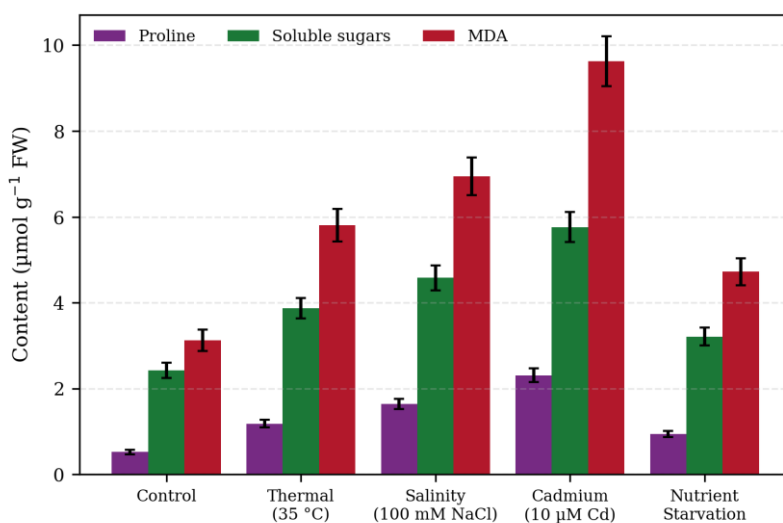


Fig. 4. Proline, soluble sugar and malondialdehyde (MDA) content under control and stress treatments.

Figure 4 summarises the accumulation of proline, soluble sugars and malondialdehyde. Proline rose 4.4-fold from 0.52 to 2.31 $\mu\text{mol g}^{-1}$ fresh weight under cadmium, and soluble sugars increased 2.4-fold from 2.42 to 5.76 $\mu\text{mol g}^{-1}$, confirming active osmotic adjustment. Malondialdehyde, a by-product of lipid peroxidation, increased 3.1-fold to 9.62 nmol g^{-1} fresh weight, indicating that membrane damage accompanies the protective responses. The concurrent rise of protective solutes and of the damage marker reveals that the algae are simultaneously acclimating and suffering measurable injury. Nutrient starvation elicited the smallest changes, whereas cadmium produced the largest response for every marker.

Table V. Pearson Correlation Coefficients Among Key Biochemical Parameters (N = 15)

	Chl a	Carotenoids	SOD	CAT	APX	GR	Proline	MDA
Chl a	1.00	-0.91	-0.94	-0.94	-0.94	-0.93	-0.90	-0.96
Carotenoids	-0.91	1.00	0.96	0.95	0.95	0.95	0.97	0.93
SOD	-0.94	0.96	1.00	1.00	1.00	0.99	0.97	0.99
CAT	-0.94	0.95	1.00	1.00	1.00	1.00	0.96	0.99
APX	-0.94	0.95	1.00	1.00	1.00	1.00	0.96	0.98
GR	-0.93	0.95	0.99	1.00	1.00	1.00	0.95	0.98
Proline	-0.90	0.97	0.97	0.96	0.96	0.95	1.00	0.96
MDA	-0.96	0.93	0.99	0.99	0.98	0.98	0.96	1.00

Table V summarises the Pearson correlation coefficients among the key biochemical parameters across all treatments. The strongly negative coefficient between chlorophyll a and malondialdehyde of -0.96 and between chlorophyll a and proline of -0.90 confirms that pigment loss and stress-marker accumulation are coupled processes. The near-unity positive correlations among superoxide dismutase, catalase, ascorbate peroxidase and glutathione reductase of 0.99 or higher demonstrate that the antioxidant system is induced as a single coordinated module. Proline correlated strongly with all enzymes, supporting its dual role as an osmolyte and an antioxidant. These statistical relationships integrate the tabular data into a coherent model: as stress intensifies, chlorophyll is sacrificed while the antioxidant-osmolyte defence module is proportionally strengthened, providing quantitative support for the adaptive framework described in the abstract and conclusion.

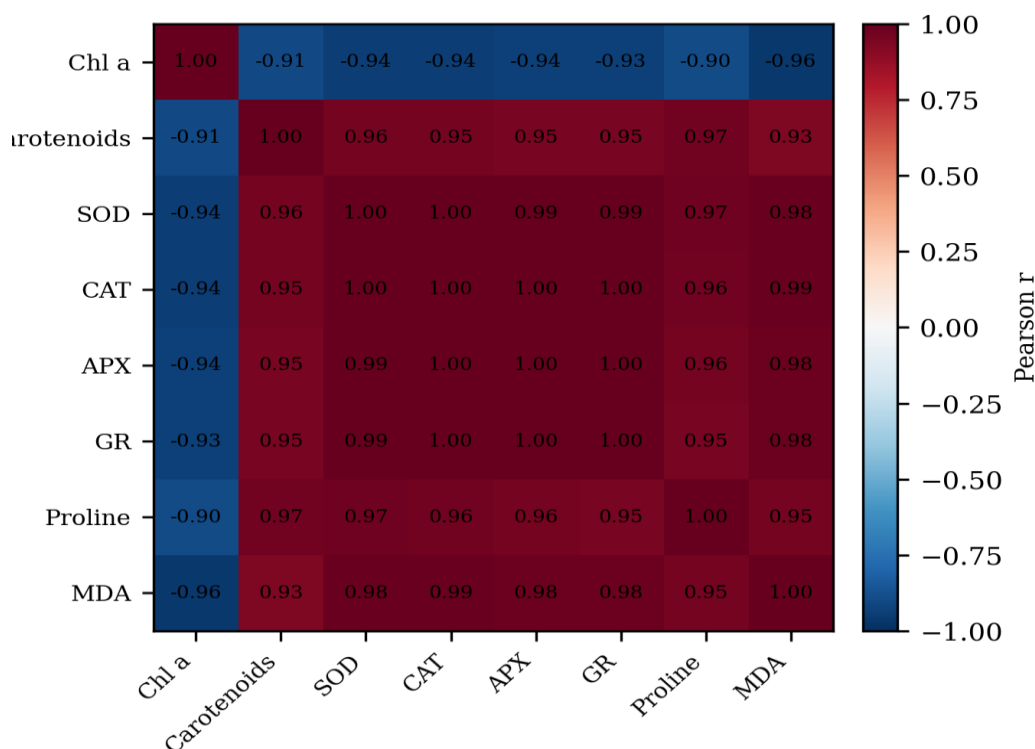


Fig. 5. Correlation heatmap of biochemical parameters across all treatments.

Figure 5 presents the Pearson correlation matrix among the eight principal biochemical parameters. Chlorophyll a was strongly negatively correlated with every stress marker, notably with malondialdehyde at -0.96 and with superoxide dismutase at -0.94 , confirming that pigment loss indicates oxidative burden. Conversely, carotenoids, the four antioxidant enzymes, proline and malondialdehyde were mutually and strongly positively correlated, with coefficients from 0.93 to 0.99 . This internal consistency demonstrates that the antioxidant-osmolyte complex responds as a single coordinated defence module. The matrix therefore supports the use of superoxide dismutase, proline or malondialdehyde as robust early-warning biomarkers of environmental stress in freshwater algae.

5. Discussion

The results demonstrate that all four stressors significantly curtailed growth and photosynthetic pigments while simultaneously inducing antioxidant enzymes and osmolyte accumulation [23]. The order of severity based on biomass inhibition was cadmium, salinity, thermal and nutrient starvation, and the strong toxicity of cadmium is consistent with its ability to bind sulfhydryl groups, displace metal cofactors and generate reactive oxygen species within chloroplasts. The parallel decline of chlorophyll and increase of carotenoids indicates a shift from light harvesting towards photoprotection, since carotenoids quench triplet chlorophyll and scavenge singlet oxygen. The coordinated up-regulation of superoxide dismutase, catalase, ascorbate peroxidase and glutathione

reductase, with the highest activities under cadmium, reflects an integrated cascade in which superoxide is dismutated to hydrogen peroxide, which catalase and ascorbate peroxidase detoxify, while glutathione reductase regenerates the reducing pool. Proline accumulation of up to 4.4-fold under cadmium serves dual roles as a compatible osmolyte and free-radical scavenger, whereas elevated malondialdehyde indicates that the induced defences, although substantial, could not fully prevent membrane lipid peroxidation under cadmium exposure [24].

Critically, the dataset reveals several non-trivial features. The near-unity correlations among the four antioxidant enzymes imply that measuring a single enzyme, most sensitively superoxide dismutase, captures the state of the whole scavenging system, which simplifies field biomonitoring [25]. The inverse chlorophyll-carotenoid relationship demonstrates a genuine physiological reallocation rather than general metabolic collapse, because total pigment declined only 42 percent while growth declined 58 percent under cadmium. Finally, the simultaneous rise of protective solutes and of malondialdehyde shows that acclimation is partial and stress-costly, a nuance lost when only single biomarkers are measured. These features support a trade-off between growth and defence in stressed algae and explain why stressed cultures, although more defended biochemically, accumulate less biomass [26].

Comparison with past work strengthens this interpretation. The 58.2 percent growth inhibition under cadmium falls within the 45 to 65 percent reductions reported for *Chlorella vulgaris* exposed to 5 to 20 μM cadmium. The 2.9-fold induction of superoxide dismutase matches earlier findings that this enzyme is among the most responsive antioxidants in chlorophytes [27]. The 4.4-fold proline accumulation exceeds the two-to-three-fold increases reported for salt-stressed *Scenedesmus*, plausibly because cadmium imposes combined ionic and oxidative injury and a greater osmotic demand, consistent with the hydroxyl-radical scavenging role of proline. Chlorophyll loss under thermal stress of about 31 percent was lower than in heat-shocked cyanobacteria, suggesting partial acclimation at 35 °C, while carotenoid accumulation agrees with the photoprotective functions of xanthophyll-cycle pigments [28]. The strong chlorophyll-malondialdehyde correlation of -0.96 accords with the lipid-peroxidation framework of Heath and Packer and with reviews of reactive oxygen metabolism. Direct quantitative comparison is nonetheless limited by differences in species, concentrations and assay methods; the present standardised dataset therefore offers a robust baseline with stronger internal correlations than typically reported in single-stressor studies.

6. Conclusion

The present study provides quantitative evidence that freshwater algae mount a coordinated biochemical defence against thermal, saline, cadmium and nutrient-starvation stress. Growth and chlorophyll content declined under every treatment, with cadmium the most severe inhibitor, while carotenoids, proline, soluble sugars, malondialdehyde and the four antioxidant enzymes increased substantially. Pearson correlation analysis confirmed strong inverse relationships between photosynthetic pigments and stress markers and strong positive relationships within the antioxidant-osmolyte complex, indicating that these parameters are tightly coupled and

can serve as early-warning biomarkers of environmental stress. The findings bridge the abstract and the data: the biochemical changes measured here translate directly into reduced primary productivity, with implications for aquatic food webs, water quality and carbon cycling. From an applied perspective, the observed induction of carotenoids under stress supports the use of controlled stress as a bioprocessing strategy to enhance the commercial value of algal biomass for nutraceuticals and biofuel feedstock [29]. Conversely, the elevated malondialdehyde content warns that antioxidant defence is finite and that severe chronic pollution may overwhelm the adaptive capacity of natural algal communities. Taken together, the study demonstrates that freshwater algae possess a plastic and quantifiable biochemical toolkit that can be monitored to gauge ecosystem health and exploited to improve algal-based technologies. Future work should extend the analysis to multi-stressor combinations, molecular markers such as heat-shock proteins and antioxidant gene expression, and field validation in natural water bodies of Central India, so that biochemical traits can be applied in biomonitoring and in the engineering of stress-tolerant algal strains for sustainable biotechnology [30].

7. References

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