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Impact Assessment of Salinity Stress on Chlorophyll Contents in Leafy Vegetables

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A B S T R A C T :

Salinity is a critical abiotic stress factor limiting agricultural productivity, particularly in semi-arid regions with marginal soils. This study investigates the effect of varying salinity levels on chlorophyll content in three widely consumed leafy vegetables. Results showed a consistent and statistically significant reduction in chlorophyll content with increasing salinity across all species, with the severity of decline being species-specific. Spinach displayed the highest tolerance by retaining more chlorophyll under stress, followed by Cabbage, while Cilantro exhibited the greatest sensitivity. One-way ANOVA confirmed significant differences among all treatment groups ($p < 0.001$). These findings highlight chlorophyll degradation as a reliable indicator of salt stress and provide critical insights into crop selection for salinity-prone areas. The results offer a physiological basis for developing salt-tolerant varieties and guiding sustainable vegetable cultivation practices in affected regions.

Keywords: Salinity stress, chlorophyll content, salt tolerance, leafy vegetables.

1. INTRODUCTION

Salinity stress is a critical abiotic factor adversely affecting plant growth, particularly in arid and semi-arid regions where irrigation practices lead to salt accumulation in the soil. One of the most pronounced impacts of salt stress in plants is the reduction in chlorophyll content, which directly impairs photosynthesis, growth, and crop productivity. Chlorophyll, the primary pigment responsible for light absorption, is sensitive to ionic imbalance, osmotic stress, and reactive oxygen species (ROS) triggered by elevated salt concentrations in the root zone (Parida and Das 2005).

Several studies have explored the mechanisms through which salinity affects chlorophyll biosynthesis and stability. In their experiment on Lactuca sativa (lettuce), Alzahrani et al. (2018) found a significant decline in total chlorophyll content when plants were exposed to increasing concentrations of NaCl (50–150 mM). Similarly, Khan et al. (2021) observed up to 40% reduction in chlorophyll a and b levels at 100 mM NaCl, indicating damage to thylakoid membranes and pigment instability.

In spinach (Spinacia oleracea), a widely consumed leafy vegetable, chlorophyll reduction has been associated with both ionic and osmotic components of salinity. Dissanayake et al. (2020) reported that Na^+ toxicity and K^+ deficiency in salt-stressed spinach leaves resulted in a 30–50% drop in chlorophyll concentration, along with decreased photosynthetic rate and stomatal conductance. Furthermore, the extent of chlorophyll loss varies with genotype, salinity level, and duration of exposure, emphasizing the need for crop-specific strategies. For instance, Kumar et al. (2019) evaluated different leafy greens and found that kale maintained relatively higher chlorophyll content under salinity compared to amaranth, due to better ion compartmentalization and antioxidant defense mechanisms.

Further, Singh et al. (2020) emphasized that chlorophyll degradation is one of the earliest responses to salt stress and serves as a sensitive indicator for selecting salt-tolerant cultivars. In a case study involving the Vidarbha region, Kale et al. (2019) noted that salinity levels, though moderate, significantly affect vegetable yields and plant physiology, highlighting the need for crop-specific tolerance evaluation. As leafy vegetables are increasingly grown for both subsistence and commercial purposes in this region, understanding their salinity responses will help ensure better land use planning, crop diversification, and resource-efficient farming in marginal lands.

Soil salinity in semi-arid regions like some parts of the Vidarbha region of Maharashtra, has been documented in regional soil surveys and research studies. According to the National Bureau of Soil Survey and Land Use Planning (NBSS&LUP), many soils in Vidarbha region show moderate salinity levels, especially in poorly drained areas or where irrigation water has high salt content (NBSS&LUP 2016). These conditions can lead to the accumulation of sodium salts in the root zone, reducing nutrient uptake and interfering with metabolic functions, particularly in salt-sensitive crops like leafy vegetables (Shrivastava and Kumar 2015). In this context, the study was undertaken to assess the impact of saline stress on chlorophyll contents in leafy vegetables.

2. MATERIALS AND METHODS

2.1. Plant Material

The study was conducted to evaluate the impact of salinity stress on chlorophyll content in selected leafy vegetables under controlled pot conditions using a completely randomized design (CRD). Three commonly consumed leafy vegetables—Spinacia oleracea (spinach), Brassica oleracea var. capitata (Cabbage), and Coriandrum sativum (Cilantro)—were selected based on their regional relevance and known sensitivity to salinity. Certified seeds were procured from an agricultural supplier and sown in plastic pots (5 kg capacity) filled with a sterilized mixture of loam soil and well-decomposed farmyard manure in a 2:1 ratio. After germination, seedlings were thinned to maintain uniform plant density.

2.2. The Experiment

Salinity stress was imposed after 12 days of sowing by irrigating plants with saline solutions prepared using analytical-grade sodium chloride (NaCl). Four salinity levels were maintained: 0 mM (control), 50 mM, 100 mM, and 150 mM NaCl, designated as T0, T1, T2, and T3, respectively. These concentrations were selected based on previous studies to simulate mild, moderate, and high salinity stress conditions in leafy vegetables (Flowers and Yeo 1995; Munns and Tester 2008). The saline solutions were applied twice a week, and care was taken to maintain uniform moisture across all treatments. Soil electrical conductivity (EC) was regularly monitored using a portable EC meter to confirm salinity levels.

2.3. Growth Conditions

Plants were grown in a polyhouse facility under natural light conditions, with an average daytime temperature of 28 ± 2°C and relative humidity of 65–70%.

2.4. Sampling

After 30 days of salinity treatment, fully expanded leaves from the middle portion of each plant were harvested for chlorophyll estimation. Leaf samples were processed following Arnon's method (1949). Absorbance was measured at 645 nm and 663 nm using a UV–visible spectrophotometer.

2.5. Data Analysis

In addition to pigment data, visual observations such as leaf chlorosis and growth reduction were recorded. Data were analyzed using one-way analysis of variance (ANOVA) to evaluate the effect of salinity treatments on chlorophyll content. All statistical analyses were performed using GraphPad Prism software version 7.0.

3. RESULTS AND DISCUSSION

Salinity stress induced a progressive and concentration-dependent decline in total chlorophyll content across all three leafy vegetable species, with the extent of reduction varying among crops. Under control conditions (0 mM NaCl), maximum levels of chlorophyll a, chlorophyll b, and total chlorophyll were observed, indicating optimal physiological functioning and photosynthetic efficiency. Spinacia oleracea exhibited the highest chlorophyll content (3.19 ± 0.07 mg/g; Table 1), which may be attributed to its relatively larger and thicker leaf structure that facilitates higher pigment accumulation. These findings are consistent with previous observations in salt-free conditions (Dissanayake et al. 2020).

At 50 mM NaCl, reductions in chlorophyll content were evident in all three species (Table 1). S. oleracea retained the highest level (2.98 ± 0.02 mg/g), indicating only mild pigment degradation and early osmotic stress symptoms. In contrast, Coriandrum sativum exhibited a significant decline to 1.99 ± 0.04 mg/g, suggesting greater sensitivity to low salinity levels. This finding supports the general trend that aromatic herbs such as C. sativum are less tolerant due to limited ion exclusion and osmoprotective capacity (Kumar et al. 2019).

Table 1: Effect of varying salinity levels on chlorophyll contents of leafy vegetables.

Salinity Level (mM NaCl)	Chlorophyll contents (mg/g)		
	Spinacia oleracea	*Brassica oleracea* var. capitata	*Coriandrum sativum*
T0 - (Control)	3.19 ± 0.07	1.97 ± 0.11	2.31 ± 0.08
T1 - 50	2.98 ± 0.02	1.69 ± 0.07	1.99 ± 0.04
T2 - 100	2.33 ± 0.06	1.21 ± 0.05	1.37 ± 0.07
T3 -150	1.87 ± 0.03	0.88 ± 0.05	0.99 ± 0.02

At 100 mM NaCl, the negative effects of salinity became more pronounced. S. oleracea maintained a moderate level of chlorophyll (2.33 ± 0.06 mg/g), while Brassica oleracea var. capitata dropped to 1.21 ± 0.05 mg/g. C. sativum showed substantial damage, with total chlorophyll content decreasing to

1.37 ± 0.07 mg/g. The marked decline in pigment concentration at this level likely reflects disrupted chlorophyll biosynthesis and enhanced degradation, attributed to ionic toxicity and oxidative stress targeting chloroplast structures (Khan et al. 2021).

Under severe salinity stress (150 mM NaCl), all species exhibited visible physiological symptoms including chlorosis, leaf curling, and growth retardation. S. oleracea, although affected, retained a relatively higher chlorophyll content (1.87 ± 0.03 mg/g), indicating a moderate degree of salt tolerance. B. oleracea showed a 44% reduction from control (1.97 ± 0.11 mg/g), while C. sativum experienced the most drastic drop (0.99 ± 0.02 mg/g), confirming its high susceptibility to salinity (Table 1). These reductions reflect both the direct oxidative degradation of photosynthetic pigments and suppression of chlorophyll synthesis under saline stress (Alzahrani et al. 2018).

Overall, these findings validate the sensitivity of chlorophyll content as a physiological marker for salinity tolerance. Among the three species tested, S. oleracea demonstrated the greatest resilience, followed by B. oleracea, whereas C. sativum exhibited the least tolerance, especially under moderate to high salinity levels. These results may inform crop selection for saline-prone regions, where soil salinity poses a growing constraint on vegetable production.

A one-way analysis of variance (ANOVA) conducted to evaluate the effect of salinity stress on total chlorophyll content revealed that salinity had a statistically significant effect on chlorophyll content in all three species ($p < 0.001$). The highly significant p-values for all three species suggest that the observed differences in chlorophyll concentration across treatments are not due to random variation but are attributable to the imposed salinity levels (Table 2).

Table 2: One way ANOVA of varying salinity level effects on chlorophyll contents of leafy vegetables.

Species	Source	Sum of Squares	df	Mean Square	F-value	p-value
Spinacia oleracea	Treatment	3.355	3	1.118	828.48	2.60×10^{-10}
	Residual	0.011	8	0.001		
Brassica oleracea	Treatment	3.714	3	1.238	229.41	4.27×10^{-8}
	Residual	0.043	8	0.005		
Coriandrum sativum	Treatment	3.614	3	1.205	463.72	2.62×10^{-9}
	Residual	0.021	8	0.003		

4. CONCLUSIONS

The study concludes that increasing salinity levels have a significant and adverse impact on the chlorophyll content of commonly consumed leafy vegetables. The results reflect impaired photosynthetic efficiency and physiological stress. These findings have practical implications for crop selection and management in semi-saline agricultural zones, where identifying salt-tolerant cultivars is essential to sustaining leafy vegetable production under increasing soil salinization pressures.

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