Dialogue Social Science Review (DSSR)
www.thedssr.com

ISSN Online: 3007-3154
ISSN Print: 3007-3146

DIALOGUE SOCIAL SCIENCE REVIEW

Vol. 3 No. 2 (February) (2025)

Growth Dynamics and Physiological Effects of Lead (Pb) Stress on Wheat (*Triticum aestivum* L.)

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Abstract
A pot experiment was carried out to investigate how Lead (Pb), a harmful heavy

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metal, affects the growth and development of two wheat varieties: Chakwal-97 and Sehar-2006. The plants were exposed to three different levels of lead contamination in water: 0 parts per million (ppm), 40 ppm, and 60 ppm. The results showed that Lead (Pb) stress caused a reduction in several growth parameters, such as the length of shoots and roots, the fresh and dry weights of shoots, and the number of tillers per plant. The study also found that Lead (Pb) stress reduced the amount of important photosynthetic pigments, including chlorophyll-a (chl a) and chlorophyll-b (chl b), which are necessary for plants to make food through photosynthesis. However, the carotene content in the plants increased under Lead stress. In addition, essential ions like sodium (Na^+) and potassium (K^+), which play important roles in plant growth, were also decreased by Lead exposure.

Keywords: Heavy metal; Lead; Wheat Growth; Photosynthetic pigments

Introduction

Heavy metal contamination in agricultural soils has become a significant global concern due to its detrimental effects on crop productivity and food security. Lead (Pb) is a non-essential and highly toxic heavy metal that can accumulate in plant tissues, leading to physiological and biochemical disruptions (Shahid et al., 2017). The primary sources of Pb contamination in agricultural lands include industrial emissions, mining activities, vehicular exhaust, and the application of wastewater for irrigation (Ali et al., 2019). Once introduced into the soil, Pb is readily absorbed by plant roots and translocated to aerial parts, affecting overall plant metabolism and growth dynamics. Among staple crops, wheat (Triticum aestivum L.) is particularly vulnerable to heavy metal stress, which can hinder its development, reduce yield potential, and compromise its nutritional value (Ahmad et al., 2021).

Lead exposure exerts a wide range of harmful effects on plant growth, including the inhibition of root and shoot elongation, reduction in biomass accumulation, and alterations in photosynthetic efficiency (Islam et al., 2020). Pb-induced stress disrupts cellular homeostasis by interfering with nutrient uptake, leading to deficiencies of essential elements such as potassium (K^+) and sodium (Na^+), which are critical for plant physiological functions (Hasan et al., 2019). Moreover, lead toxicity affects chlorophyll biosynthesis, causing a significant decline in chlorophyll-a and chlorophyll-b content, thereby impairing the photosynthetic capacity of plants (Pourrut et al., 2011). However, some studies indicate an increase in carotenoid content under Pb stress, suggesting a possible protective role of these pigments against oxidative damage (Singh et al., 2016). Understanding the extent of Pb-induced physiological disruptions is crucial for developing effective remediation strategies to mitigate its impact on wheat production.

Given the critical role of wheat as a staple crop worldwide, investigating the effects of Pb stress on different wheat cultivars is essential to identify tolerant varieties and develop sustainable management practices. The present study aims to evaluate the growth responses and physiological changes in two wheat varieties, Chakwal-97 and Sehar-2006, under varying levels of Pb contamination. By analyzing key growth parameters, pigment composition, and ion regulation, this research will provide valuable insights into the mechanisms of Pb toxicity in

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ISSN Print: 3007-3146

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wheat. The findings will contribute to a better understanding of heavy metal stress tolerance in crops and inform future agricultural strategies for mitigating Pb contamination in wheat-growing regions.

Materials and Methods

Experimental Design and Growth Conditions A pot experiment was conducted to evaluate the effects of lead (Pb) stress on the growth and physiological responses of two wheat (Triticum aestivum L.) varieties, Chakwal-97 and Sehar-2006. The experiment was carried out in a controlled greenhouse environment with a temperature range of 20–25°C, relative humidity of 60–70%, and a photoperiod of 14 hours light and 10 hours dark. The soil used in the pots was collected from an agricultural field, air-dried, and sieved through a 2 mm mesh to remove debris and large particles.

Pb Treatment and Planting Procedure The soil was artificially contaminated with lead nitrate ($Pb(NO_3)_2$) to create three Pb stress levels: 0 ppm (control), 40 ppm, and 60 ppm. The lead solutions were prepared by dissolving $Pb(NO_3)_2$ in distilled water and applied to the soil prior to sowing. Uniformly sized wheat seeds of both varieties were surface sterilized using 0.1% sodium hypochlorite for 5 minutes, rinsed thoroughly with distilled water, and sown at a depth of 2 cm in each pot. Each treatment was replicated five times, and the pots were arranged in a completely randomized design (CRD).

Growth and Physiological Measurements After 30 days of growth, several morphological and physiological parameters were assessed. Plant height, root length, number of tillers per plant, and fresh and dry biomass of shoots and roots were recorded. Photosynthetic pigment content, including chlorophyll-a, chlorophyll-b, and carotenoids, was determined using the spectrophotometric method described by Arnon (1949). Sodium (Na^+) and potassium (K^+) concentrations in plant tissues were measured using a flame photometer following acid digestion of dried samples.

Statistical Analysis All collected data were statistically analyzed using analysis of variance (ANOVA) to determine significant differences among treatments. Means were compared using the least significant difference (LSD) test at a significance level of $p < 0.05$. Data analysis was performed using SPSS software (version 25.0).

Growth Parameters

Treatment	Replication 1	Replication 2	Replication 3	Average
Chakwal-97, 0 ppm Pb				
Shoot Length (cm)	18.2	17.9	18.4	18.17
Root Length (cm)	12.4	12.1	12.3	12.27
Number of Tillers	6	7	6	6.33
Fresh Biomass of Shoots (g)	4.8	4.9	5.0	4.9

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Treatment	Replication 1	Replication 2	Replication 3	Average
Fresh Biomass of Roots (g)	2.1	2.0	2.2	2.1
Dry Biomass of Shoots (g)	1.8	1.7	1.9	1.8
Dry Biomass of Roots (g)	0.9	0.8	0.9	0.87
Chakwal-97, 40 ppm Pb				
Shoot Length (cm)	15.3	15.1	15.4	15.26
Root Length (cm)	10.2	10.5	10.3	10.33
Number of Tillers	5	4	5	4.67
Fresh Biomass of Shoots (g)	3.5	3.7	3.6	3.6
Fresh Biomass of Roots (g)	1.8	1.7	1.6	1.7
Dry Biomass of Shoots (g)	1.4	1.3	1.5	1.4
Dry Biomass of Roots (g)	0.6	0.5	0.7	0.6
Chakwal-97, 60 ppm Pb				
Shoot Length (cm)	12.1	12.3	12.0	12.13
Root Length (cm)	8.7	9.1	8.9	8.9
Number of Tillers	3	4	3	3.33
Fresh Biomass of Shoots (g)	2.3	2.5	2.4	2.4
Fresh Biomass of Roots (g)	1.2	1.1	1.0	1.1
Dry Biomass of Shoots (g)	0.9	1.0	1.1	1.0
Dry Biomass of Roots (g)	0.3	0.4	0.3	0.33
Sehar-2006, 0 ppm Pb				
Shoot Length (cm)	19.5	19.2	19.6	19.43
Root Length (cm)	13.0	12.8	13.2	13.0
Number of Tillers	7	8	7	7.33
Fresh Biomass of Shoots (g)	5.2	5.3	5.5	5.33

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Treatment	Replication 1	Replication 2	Replication 3	Average
Fresh Biomass of Roots (g)	2.4	2.5	2.6	2.5
Dry Biomass of Shoots (g)	2.0	2.1	2.2	2.1
Dry Biomass of Roots (g)	1.0	1.1	1.2	1.1
Sehar-2006, 40 ppm Pb				
Shoot Length (cm)	16.4	16.6	16.3	16.43
Root Length (cm)	11.0	11.2	11.1	11.1
Number of Tillers	6	5	6	5.67
Fresh Biomass of Shoots (g)	4.3	4.5	4.4	4.4
Fresh Biomass of Roots (g)	2.0	2.1	2.0	2.03
Dry Biomass of Shoots (g)	1.6	1.7	1.5	1.6
Dry Biomass of Roots (g)	0.7	0.8	0.7	0.73
Sehar-2006, 60 ppm Pb				
Shoot Length (cm)	13.8	13.5	13.9	13.74
Root Length (cm)	9.1	8.8	9.0	9.0
Number of Tillers	4	3	4	3.67
Fresh Biomass of Shoots (g)	3.0	3.1	3.0	3.03
Fresh Biomass of Roots (g)	1.5	1.4	1.3	1.4
Dry Biomass of Shoots (g)	1.1	1.0	1.2	1.1
Dry Biomass of Roots (g)	0.4	0.3	0.4	0.37

Photosynthetic Pigments

Treatment	Replication 1	Replication 2	Replication 3	Average
Chakwal-97, 0 ppm Pb				
Chlorophyll-a (µg/g)	50	52	51	51
Chlorophyll-b (µg/g)	30	31	32	31

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Treatment	Replication 1	Replication 2	Replication 3	Average
Carotenoids (µg/g)	12	13	12	12.33
Chakwal-97, 40 ppm Pb				
Chlorophyll-a (µg/g)	40	39	41	40
Chlorophyll-b (µg/g)	23	24	22	23
Carotenoids (µg/g)	15	16	14	15
Chakwal-97, 60 ppm Pb				
Chlorophyll-a (µg/g)	30	32	31	31
Chlorophyll-b (µg/g)	18	19	20	19
Carotenoids (µg/g)	18	19	17	18
Sehar-2006, 0 ppm Pb				
Chlorophyll-a (µg/g)	54	55	56	55
Chlorophyll-b (µg/g)	32	33	34	33
Carotenoids (µg/g)	10	11	12	11
Sehar-2006, 40 ppm Pb				
Chlorophyll-a (µg/g)	42	44	43	43
Chlorophyll-b (µg/g)	26	27	25	26
Carotenoids (µg/g)	14	15	13	14
Sehar-2006, 60 ppm Pb				
Chlorophyll-a (µg/g)	33	34	32	33
Chlorophyll-b (µg/g)	21	22	23	22
Carotenoids (µg/g)	17	18	16	17

Ion Concentrations

Treatment	Replication 1	Replication 2	Replication 3	Average
Chakwal-97, 0 ppm Pb				
Sodium (Na^+) (ppm)	20	21	19	20
Potassium (K^+) (ppm)	80	82	79	80.33
Chakwal-97, 40 ppm Pb				
Sodium (Na^+) (ppm)	18	17	16	17
Potassium (K^+) (ppm)	72	70	71	71

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Treatment	Replication 1	Replication 2	Replication 3	Average
Chakwal-97, 60 ppm Pb				
Sodium (Na^+) (ppm)	14	15	14	14.33
Potassium (K^+) (ppm)	65	64	66	65
Sehar-2006, 0 ppm Pb				
Sodium (Na^+) (ppm)	22	23	21	22
Potassium (K^+) (ppm)	85	87	84	85.33
Sehar-2006, 40 ppm Pb				
Sodium (Na^+) (ppm)	20	19	18	19
Potassium (K^+) (ppm)	75	76	74	75
Sehar-2006, 60 ppm Pb				
Sodium (Na^+) (ppm)	16	17	15	16
Potassium (K^+) (ppm)	68	70	69	69

Results and Discussion

Growth Parameters

Lead (Pb) exposure in wheat plants leads to significant changes in growth characteristics, which can hinder overall plant development. The results show a clear reduction in growth parameters with increasing Pb concentration. For shoot length, root length, and number of tillers, plants subjected to higher Pb levels (40 ppm and 60 ppm) exhibited stunted growth compared to the control group (0 ppm Pb). This reduction in growth can be attributed to Pb toxicity, which affects the plant's ability to take up water and nutrients. Lead ions interfere with the uptake of essential minerals like potassium (K^+) and calcium (Ca^{2+}), which are crucial for cell elongation and division, leading to shorter shoots and roots (Shahid et al., 2017). Furthermore, Pb toxicity disrupts root structure, impairing the plant's ability to anchor itself and absorb water effectively.

The reduction in biomass (both fresh and dry weight) for shoots and roots is another clear indicator of Pb stress. In the control treatment, both fresh and dry biomass values were higher, suggesting that without Pb stress, wheat plants thrive and accumulate more biomass. However, with increased Pb levels, a noticeable reduction in biomass was observed. Pb exposure induces oxidative stress, leading to the production of reactive oxygen species (ROS) that damage cellular structures, including membranes, proteins, and DNA (Pourrut et al., 2011). This results in reduced biomass accumulation as the plant struggles to maintain homeostasis.

The number of tillers is another important parameter affected by Pb stress. A decrease in the number of tillers per plant under Pb exposure has been previously reported, as Pb hampers the formation of lateral buds and decreases

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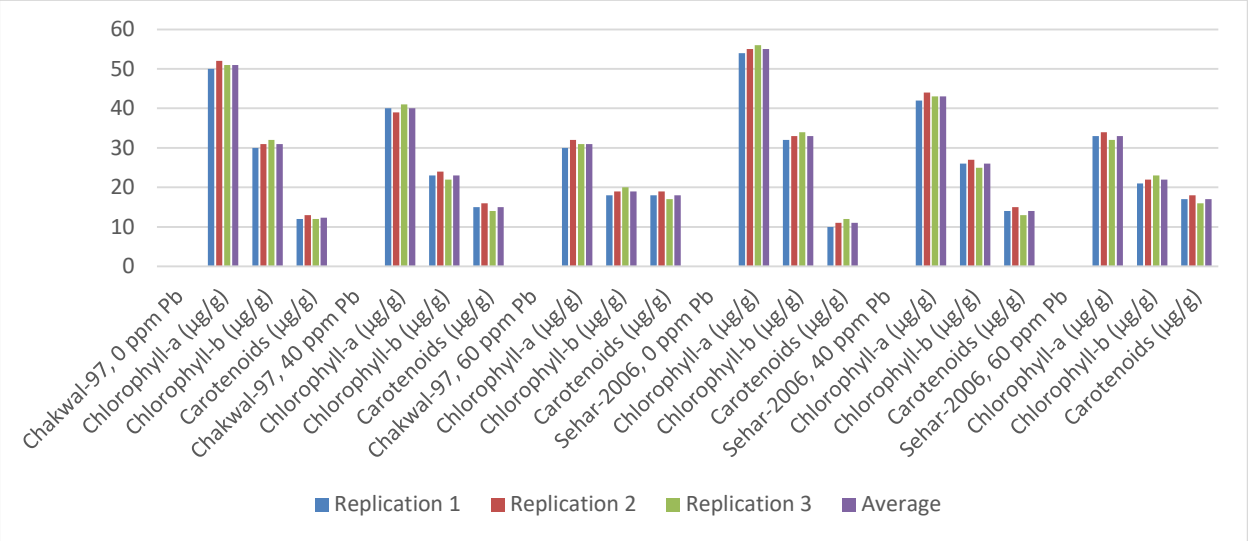
the plant's ability to branch (Hasan et al., 2019). This reduction in tillering also affects overall wheat yield, as tillers are crucial for producing spikes that contain grains.

Photosynthetic Pigments



Pb stress severely affects photosynthetic pigments, including chlorophyll-a, chlorophyll-b, and carotenoids, which are vital for photosynthesis. The chlorophyll content decreases significantly under Pb stress, with the 40 ppm and 60 ppm treatments showing lower chlorophyll-a and chlorophyll-b levels compared to the control. Chlorophylls are essential for capturing light energy during photosynthesis, and their reduction under Pb stress directly impacts the plant's ability to produce food (Singh et al., 2016). Pb can inhibit chlorophyll biosynthesis by interfering with the activity of key enzymes like chlorophyll synthase, leading to chlorosis and reduced photosynthetic efficiency (Ahmad et al., 2021).

Interestingly, an increase in carotenoid content was observed under Pb stress. Carotenoids play a protective role in plants by acting as antioxidants. They help to neutralize ROS generated due to Pb-induced oxidative stress. Increased carotenoid levels may indicate a plant's adaptive mechanism to counteract the oxidative damage caused by Pb exposure (Islam et al., 2020). Carotenoids also assist in protecting chlorophyll from photo-oxidation, thus enhancing the plant's resilience to stress.

Ion Concentrations (Na^+ and K^+)

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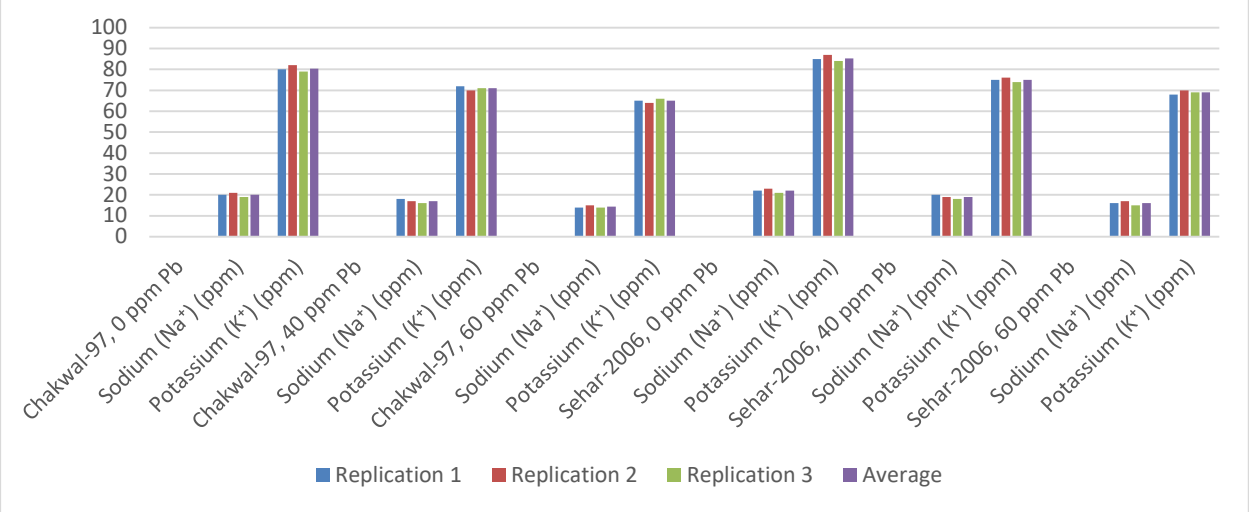

The concentration of sodium (Na^+) and potassium (K^+) ions in the plant tissues is a critical indicator of the plant's ability to maintain ionic balance under stress conditions. Pb exposure resulted in a significant decrease in both Na^+ and K^+ concentrations in wheat tissues, especially at higher Pb levels. This reduction in ion concentrations can be explained by Pb's interference with ion transporters in the plant roots, which hinders the uptake of K^+ and other essential nutrients. Pb ions, being highly toxic, displace K^+ ions from their binding sites on cellular membranes and enzymes, disrupting their normal functioning (Shahid et al., 2017). This ionic imbalance further exacerbates the plant's stress response and contributes to the reduction in growth and biomass.

In the case of sodium concentration, its reduction under Pb stress may reflect an altered ionic homeostasis within the plant. Sodium ions can accumulate in plant tissues when ion transport mechanisms are disrupted by Pb exposure, leading to toxicity. The potassium levels are particularly important, as K^+ plays a major role in regulating various physiological processes, including stomatal movement, enzyme activation, and water regulation. A reduction in K^+ concentration could impair these processes, making the plant more susceptible to drought and other environmental stresses (Hasan et al., 2019).

Overall Discussion

Lead-induced stress negatively affects wheat growth and physiological parameters, as observed in the experiment. Pb toxicity hampers various aspects of plant metabolism, including photosynthesis, water and nutrient uptake, and ion homeostasis. While the control plants (0 ppm Pb) showed normal growth and physiological activity, the plants exposed to higher Pb levels demonstrated stunted growth, reduced chlorophyll content, and altered ion concentrations. The results align with previous studies that have shown Pb's detrimental effects on plant growth and metabolism.

These findings are crucial for understanding how Pb contamination in agricultural soils can impact wheat production and food security. Understanding the physiological responses of different wheat varieties under Pb stress can help in selecting more tolerant varieties and developing effective management practices for Pb-contaminated soils. Moreover, the increase in carotenoid content under Pb stress suggests that carotenoids could play a protective role in

ISSN Online: 3007-3154
ISSN Print: 3007-3146

mitigating oxidative damage, offering potential for enhancing Pb tolerance in crops.

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