Life Science and Biotechnology
Volume 2, Number 1, May 2024
Pages: 29 – 32

Growth and physiological adaptation of Ageratum conyzoides L. under salinity stress

Yusi Ananda Putri[1], Ulfatul Inayah[1], Abdillah Maulana Farhan[1], Edia Fitri Dwinianti1*[2]
[1]Biology Department, Faculty of Mathematics and Natural Sciences, University of Jember, Indonesia
[2]Meru Betiri National Park Office, Jember, East Java, Indonesia
* Correspondence Author: ediafitridwinianti@gmail.com

Abstract

Physiological stress affects all plants, including salinity stress, which poses significant challenges. *Ageratum conyzoides* L. was selected as the subject of this study due to its reputed ability to thrive under various stress conditions, owing to its excellent adaptive capacity. The study employed salt solutions at concentrations of 1000 ppm, 3000 ppm, and 5000 ppm, administered as a single treatment over four weeks. The aim was to assess the impact of salinity stress on the growth, stomatal density, and chlorophyll content of *A. conyzoides*. Results indicate that salt treatment adversely affected the height of *A. conyzoides*. Stomatal density was highest at the 5000 ppm concentration, attributed to intensified transpiration in response to stress compared to lower concentrations. Chlorophyll content also showed a reduction at higher salt concentrations. These findings highlight the physiological responses of *A. conyzoides* to salinity stress, providing insights into its adaptation mechanisms under adverse environmental conditions.

Keywords: *Ageratum conyzoides*, chlorophyll, salinity, stress

Introduction

Ageratum conyzoides, commonly known as bandotan, is a plant that thrives in subtropical and tropical regions such as Indonesia. This plant is characterized by its stems and leaves covered entirely with fine white hairs. Known for its environmental resilience, *A. conyzoides* exhibits exceptional adaptive capabilities, enabling its survival even in dense shrub habitats, where it can behave invasively (Atisha & Mita, 2018; Ulum et al, 2023a).

Salt (NaCl) is a compound containing sodium, an essential micronutrient for plants. Sodium plays a crucial role in plant physiology by partially replacing potassium necessary for optimal growth. Chlorine is absorbed by plants in the form of Cl- ions. Its function is directly related to regulating osmotic pressure within plant cells (Syakir et al., 2008). Salt stress suppresses plant growth by inhibiting cell enlargement, cell division, protein production, and overall biomass. Symptoms of plant growth under high soil salinity include abnormal growth patterns, tip drying, and chlorosis (Triyani et al., 2013). Stressed plants undergo metabolic changes to adapt to environmental fluctuations.

Salinity stress significantly impacts the morphology, physiology, and biochemistry of *A. conyzoides* (Ulum et al, 2023). It adversely affects plant growth and development, inducing osmotic and ionic stress due to salt accumulation around the roots, reducing the plant's ability to absorb water. Furthermore, excessive uptake of salt constituents can lead to toxicity in plants (Ginting et al., 2019; Barus et al., 2021). Salinity stress alters the growth, chlorophyll content, and stomatal density of *A. conyzoides*, necessitating further investigation.

Therefore, the objective of this study is to identify the effects of salinity stress on the growth, stomatal density, and chlorophyll content of *A. conyzoides*. By exploring these impacts, we aim to contribute to the understanding of how this plant responds to saline conditions, providing insights that could inform strategies for its management and conservation in diverse ecosystems.

Materials and Methods

Plant Material

Ageratum conyzoides used in this study were seedlings with a height of 5 cm that grew wild around the botanical garden of the Department of Biology, FMIPA, University of Jember. The research time was March 10^{th} - April 10^{th}, 2023. Greenhouse abiotic conditions were humidity 75-78.3%, temperature 30 ^{0}C, and light intensity 181-437 Lux.

Plant nursery and stress treatment

This study used a completely randomized design (CRD) with 3 treatments and 1 control. Repetition was done three times. Each pot contained 3 plants.

Planting media was put into the pot until it reached 12 cm and the distance between one plant and another was approximately 3 cm. The salinity concentrations used were 0 ppm, 1000 ppm, 3000 ppm, and 5000 ppm. Each pot is watered with 400 ml of salt water. Stress treatment was conducted once a week.

Stress effect measurement

The observation parameters were plant height, stomatal density, and chlorophyll content. Growth measurements were taken once a week. The analysis of stomatal density and chlorophyll content was conducted in the fourth week after treatment.

Stomatal density observations were made by applying clear nail polish to the adaxial surface of the leaf. After drying, the nail polish was removed by attaching clear tape and lifting it. The stomata mold from the nail polish was attached to the glass object and observed using a microscope with a magnification of 400x. The number of *A. conyzoides* stomata was counted using a microscope with the help of the Optilab tool (Ulum et al, 2023).

The chlorophyll content was measured by weighing 0.1 g of leaf and cutting it into small pieces. Subsequently, the leaf samples were immersed in 10 ml of 96% alcohol in the absence of light for three days, allowing the chlorophyll to dissolve in the alcohol solution. The chlorophyll content was then quantified using a spectrophotometer at wavelengths of 649 nm and 665 nm. The absorbance values were converted into mg/L units using the formula (Ulum et al, 2023b).

Chlorophyll a (mg/L) = (13.7 x A665) – (5.67 x A649)

Chlorophyll b (mg/L) = (25.8 x A649) − (7.7 x A665)

Total Chlorophyll (mg/L) = (20 x OD649 + 6,1 x OD645)

Statistical analysis

Data were analyzed with R version 4.1.2 for Windows (R Foundation for Statistical Computing). Statistical data visualization was using ggplot2 (Wickham, et. al. 2016).

Results

Salinity effect on the plant height

The growth of plants is influenced by a number of external factors, one of which is the impact of salinity levels or salt concentration. This also applies to *A. conyzoides*, although they possess a remarkable ability to adapt to stress (Ulum et al, 2023a). Regression analysis has shown significant differences in the physical characteristics of *A. conyzoide* under salinity stress treatments, as illustrated in Figure 1.

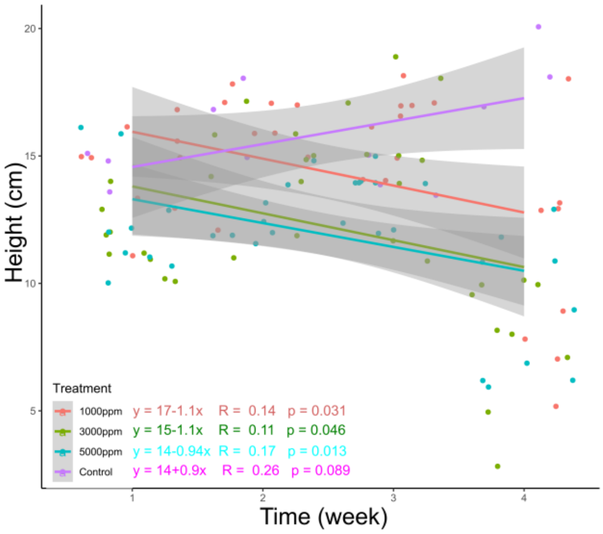

Figure 1. Effect of salinity stress on the height of *A. conyzoides* based on Regression analysis with correlation value.

The analysis showed a significant difference in the plant height of *A. conyzoides*. In the control section, *A. conyzoides* showed more effective growth compared to those subjected to salinity stress. This is due to the potential of salinity to affect the metabolic system of *A. conyzoide*, resulting in slower growth (Busaifi et al., 2016). In the salinity stress treatment, the 1000 ppm concentration has the greatest inhibition with an average *A. conyzoides* height of only 9.45 cm, while the 3000 ppm and 5000 ppm concentrations have an average height of 9.96 cm and 10.53 cm, respectively. This difference is also influenced by the absorption conditions in each individual, with the *A. conyzoides* at 1000 ppm presumably having a greater absorption capacity so that the height is shorter than the other concentrations. Plants experiencing salinity stress generally do not show direct damage, but their growth is depressed and gradually changes. The decrease in height growth is caused by osmotic stress, which inhibits water absorption, and the influence of excess Na and Cl ions due to NaCl application, which also inhibits cell division and enlargement (Romadloni and Wicaksono, 2018).

Salinity effect on the stomatal density

Stomata are a crucial part of plants involved in metabolic processes (Ulum et al, 2023b). Salinity stress induced by salt application causes stomata to become more compact, as observed in Figure 2. The salinity stress has been shown to increase stomatal density (Figure 3). This increased stomatal density occurs because *A. conyzoides* receives a salt supply, prompting the formation of a greater number of stomata compared to normal/control conditions to facilitate the process of transpiration.

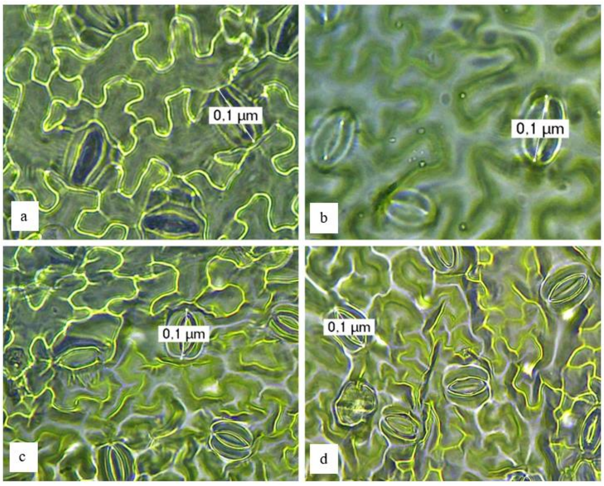

Figure 2. The stomatal structure of *A. conyzoides*. A = control; b = 1000 ppm; c = 3000 ppm; d = 5000 ppm)

The highest stomatal density was observed in the 5000 ppm salt water salinity stress treatment. This phenomenon was caused by a significant increase in the evaporation process in *A. conyzoide*, resulting in the highest number of stomatal densities compared to the other treatments. This evaporation is an adaptive response of *A. conyzoides* to the direct entry of saline substances, allowing the plant to regulate the entry and exit of substances into the plant body. According to Junandi et al. (2019), increased salinity can reduce photosynthetic capacity, which affects metabolic processes. Excessive uptake of the element sodium (Na) results in reduced uptake of water and potassium (K). Impaired water uptake interferes with the photosynthetic process by closing the stomata, thus reducing the supply of CO_2 to the chloroplast. This shows that salinity stress not only affects stomatal physiology but also affects the ionic balance and photosynthetic efficiency of *A. conyzoide*.

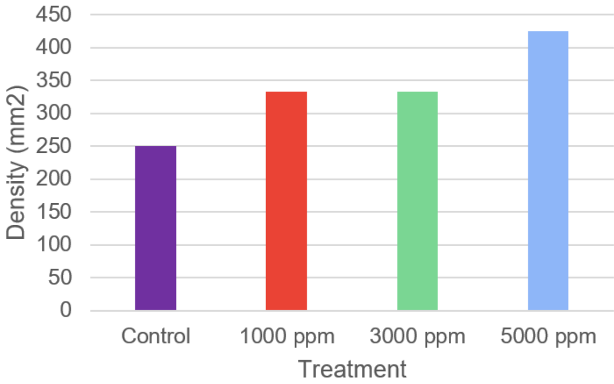

Figure 3. Effect of salinity stress on the stomatal density of *A. conyzoides*.

Salinity effect on the chlorophyll

Chlorophyll content under salinity stress generally shows leaf chlorosis with varying degrees of damage (Purwaningrahayu and Taufik, 2017). These measurements showed the highest chlorophyll content in the control treatment with a value of 11.04 mg/L. Factors affecting chlorophyll formation include genes, light (Ulum et al, 2021), and elements such as N, Mg, and Fe, which act as components and catalysts in chlorophyll synthesis (Pratama, 2015).

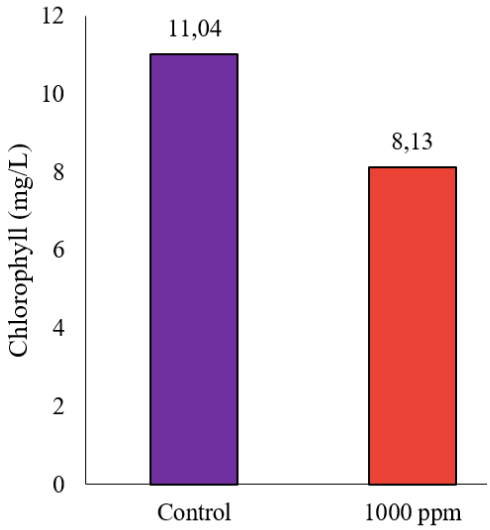

Figure 3. Effect of salinity stress on the Chlorophyll of *A. conyzoides*

Chlorophyll content decreases with increasing salinity. The decrease in chlorophyll content in plants under salinity stress conditions has been considered a typical symptom of oxidative stress and was related to inhibition of chlorophyll synthesis (Santos, 2004). In addition, salinity stress causes an increase in the formation of reactive oxygen species which can damage cellular components (Taibi et al., 2016).

Acknowledgments

This research was conducted in the project course of Plant Ecophysiology.

References

Atisha, S. A. Dan S. R. Mita. 2018. Herban bandotan (*Ageratum conyzoides* L.) sebagai pengobatan luka terbuka. *Farmaka*, 16(3): 1-10.

Barus, W. A., A. Munar, I. Sofia, dan E. Lubis. 2021. Kontribusi asam salisilat untuk ketahanan cekaman salinitas pada tanaman. Jurnal Penelitian Bidang Ilmu Pertanian. 19(2): 9-19.

Ginting, F. P., Y. Asbur, Y. Purwaningrum, M. S. Rahayu, dan N. Nurhayati. 2019. Pengaruh stres salinitas terhadap pertumbuhan, komposisi mineral, kadar prolin, zat antioksidan kedelai. Agriland: Jurnal Ilmu Pertanian. 7(1), 48-54.

Junandi, J., M. Mukarlina, dan R. Linda. 2019. Pengaruh cekaman salinitas garam NaCl terhadap pertumbuhan kacang tunggak (*Vigna unguiculata* L. Walp) pada tanah gambut. Jurnal Protobiont. 8(3): 101-105

Pratama, A. J. 2015. Analisis Kandungan Klorofil Gandasuli (*Hedychium gardnerianum* Shephard ex Ker-Gawl) pada Tiga Daerah Perkembangan Daun yang Berbeda. In Prosiding KPSDA. 1(1): 216-219.

Romadloni, A dan K. P. Wicaksono. 2018. Pengaruh beberapa level salinitas terhadap perkecambahan kacang hijau (*Vigna radiata* L.) Varietas Vima 1. Jurnal Produksi Tanaman. 6(8): 1663 – 1670.

Santos, C.V., 2004. Regulation of chlorophyll biosynthesis and degradation by salt stress in sunflower leaves. Scientia Horticulturae. 103: 93–99.

Syakir, M., N. Maslahah, dan M. Januwati. 2008. Pengaruh salinitas terhadap pertumbuhan, produksi dan mutu sambiloto (*Andrographis paniculata* Ness). Jurnal Buletin Littro. 19(2): 129-137.

Taibi, K., Taibi, F., Abderrahim, L. A., Ennajah, A., Belkhodja, M., & Mulet, J. M. 2016. Effect of salt stress on growth, chlorophyll content, lipid peroxidation and antioxidant defence system in *Phaselus vulgaris* L. South African Journal of Botany. 105: 306 - 312. DOI: http://dx.doi.org/10.1016/j.sajb.2016.03.011.

Triyani, A., Suwarto, dan S. Nurchasanah. 2013. Toleransi genotip kedelai (*Glycin max* L. Merril.) terhadap konsentrasi garam NaCl pada fase vegetatif. Agronomika. 13(1): 201- 213.

Ulum, F.B., Akbar, S. M., Andiana, J., Rosyadi, A., & Setyati, D. Pengaruh Cekaman Salinitas terhadap Pertumbuhan dan Perkembangan Bandotan (*Ageratum conyzoides* L.). *Journal of Natural Science*, 4(3): 152-162. *DOI:* 10.34007/jonas.v4i3.435

Ulum, F.B, Setyati, D. and Su'udi, M., 2023. Epiphytic ferns and orchids adaptation mechanism based on stomatal structure and chlorophyll content in Urban Forest of Jember University, Indonesia. Life Science and Biotechnology, 1(1), pp.10-15. DOI: https://doi.org/10.19184/lsb.v1i1.39678

Ulum, F.B., Hadacek, F. and Hörandl, E., 2021. Polyploidy improves photosynthesis regulation within the Ranunculus auricomus complex (Ranunculaceae). Biology, 10(8), p.811.. https://doi.org/10.3390/biology10080811

Wickham, H. and Wickham, H., (2016). Getting Started with ggplot2. ggplot2: Elegant graphics for data analysis, pp.11-31.