



Initial Growth and Physiological Response of Rice with Ammonium Sulfate and *Chromolaena odorata* Under Water Stress

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Abstract: Drought stress is one of the biggest challenges in rice production, requiring effective fertilization strategies to improve plant resilience. This study evaluated the effect of ammonium sulfate ((NH₄)₂SO₄) and *Chromolaena odorata* (Cho) fertilization on early growth and physiological responses of rice (*Oryza sativa* L. cv. Inpago 11) under field capacity (100% WFC) and drought stress (50% WFC) conditions. Experiments were conducted using a divided plot design with three fertilizer treatments control, (NH₄)₂SO₄, and (NH₄)₂SO₄ + Cho and two levels of water availability. Results showed that at 100% WFC, (NH₄)₂SO₄ + Cho fertilization significantly increased maximum growth potential (48.09%), vigor index (29.77%), and growth speed (16.60%.etmal⁻¹) compared to the control. Total chlorophyll and carotenoid contents also increased by 50% and 63.8% in the (NH₄)₂SO₄ + Cho treatment compared to the control. At 50% WFC, differences between treatments became insignificant for most parameters, but proline accumulation increased by 62.5% in the (NH₄)₂SO₄ + Cho treatment compared to the control. This study shows that the combination of (NH₄)₂SO₄ + Cho effectively improves the early growth and physiological resilience of rice under optimal conditions. Still, its effectiveness is reduced under drought stress, emphasizing the importance of adaptive fertilization strategies to water availability.

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1. Introduction

Rice (*Oryza sativa* L.) is one of the world's most important food crops, critical to many countries' food security and economies (Li et al., 2018). However, various abiotic stresses, including drought, often limit rice production (Moonmoon and Islam, 2017). Growth can be disrupted by drought stress, development, and productivity of rice plants, especially at critical phases such as germinating and early growing. Therefore, developing strategies to improve rice drought stress tolerance is important in maintaining the stability of rice production amidst climate change and limited water resources (Fahad et al., 2017).

Fertilization is one of the cultivation practices that play a significant role in the growth and yield of rice plants (Ma et al., 2023). Ammonium sulfate ((NH₄)₂SO₄) is a commonly used inorganic fertilizer in rice cultivation as an important nitrogen source for plant growth. However, the effectiveness of inorganic fertilization can be reduced under abiotic stress conditions such as drought, so an alternative or complementary approach is needed (Al-Yasi et al., 2020). Organic matter use, such as *Chromolaena odorata*, as a source of supplemental nutrients and soil improver has gained attention. In the last few years (Ahmad et al., 2022). *Chromolaena odorata* is an invasive weed with potential high biomass as a source of organic matter and good nutritional content (Fikrinda et al., 2021).

Although the role of ammonium sulfate and *Chromolaena odorata* in improving rice growth and productivity was reported in several previous studies (Ding et al., 2015; Cao et al., 2018; Fikrinda et al., 2021). However, rice's early growth and physiological responses to combining these materials under drought stress conditions have not been extensively investigated. Understanding how rice responds morphologically and physiologically to ammonium sulfate and *Chromolaena odorata* fertilization in the early growth phase under drought stress may provide essential insights into developing more effective nutrient and water management strategies for abiotic stress-resilient rice production.

Previous studies have evaluated rice's growth and physiological responses to ammonium sulfate fertilization under drought-stress conditions. Applying ammonium sulfate at optimal doses can improve growth, chlorophyll content, and water use efficiency in drought-stressed rice (Guo et al., 2008; Gao et al., 2010). Similar results were found by Shiwei Guo et al. (2007), who indicated that ammonium sulfate fertilization could alleviate the negative effect of drought stress on rice growth and yield by increasing nutrient uptake and photosynthetic efficiency. However, both studies also emphasized optimizing the dose and timing of ammonium sulfate application to avoid toxicity effects or increased osmotic stress on rice plants.

Meanwhile, the utilization of Organic matter (Dayan, 2024) source *Chromolaena odorata* in rice cultivation has been explored in several studies. Previous research has shown *Chromolaena odorata*'s potential as an effective raw material for organic fertilizer. Several studies found that *Chromolaena odorata*-based fertilizers, either in compost, bokashi, or liquid fertilizer, can significantly improve soil fertility. This is indicated by an increase in the content of soil nutrients such as nitrogen, phosphorus, and potassium after applying the fertilizer (Ahmad and Lamangantjo, 2018; Agustina et al., 2019; Nurmas et al., 2023). In addition, the use of *Chromolaena odorata* compost was also shown to improve soil chemical properties and increase nutrient absorption in rice and lettuce plants (Munir, 2019; Suri and Yudono, 2020). These studies suggest that *Chromolaena odorata* contains nutrients necessary to support plant growth and can be a potential alternative to organic fertilizer.

Further research has also shown the positive effects of using *Chromolaena odorata*-based fertilizers in growing and producing various crops. Liquid fertilizer from *Chromolaena odorata* significantly increases the growth of purple eggplants and shallots, with an optimal concentration of 60% (Ahmad et al., 2022). Meanwhile, combining *Chromolaena odorata* ash and bagasse increased stem diameter, stem height, and leaf yield in Hibiscus sabdariffa plants. Liquid fertilizer made from a mixture of cattle urine and *Chromolaena odorata* was also effective in increasing growth and biomass production in fodder grasses such as *Panicum maximum*, *Brachiaria decumbens*, and *Pennisetum purpureum* (Hasan et al., 2019). In addition, *Chromolaena odorata* compost was also shown to improve chili plant performance in terms of fruit number and freshness. Dry weight⁻¹, and plant dry weight (Dewi et al., 2018). These findings further strengthen the potential of *Chromolaena odorata* as an effective organic fertilizer raw material to increase the productivity of various crops.

Several researchers have also studied the effects of inorganic and organic fertilizer combinations on rice growth and productivity. Combining inorganic and organic nitrogen fertilizers can increase rice's growth, nutrient uptake, and yield compared to the application of inorganic fertilizers alone (Gusti et al., 2022). Integrating inorganic and organic fertilizers can improve soil fertility, increase fertilizer use efficiency, and support more sustainable rice farming (Iqbal et al., 2021). These results indicate the potential of the combination of ammonium sulfate and *Chromolaena odorata* in improving rice growth and productivity, especially under abiotic stress conditions.

Although previous research has provided insights into the response of rice to ammonium sulfate fertilization, *Chromolaena odorata* utilization, and inorganic-organic fertilizer combinations, there is still a gap in knowledge regarding the early growth and physiological response of rice to specific

combinations of ammonium sulfate and *Chromolaena odorata* under drought stress conditions. The growth stage and final yield of plants have been the focus of most previous studies, while information on the response of rice in the germination and early growth phase is still limited. In addition, the physiological mechanisms underlying the response of rice to these fertilizer combinations under drought stress conditions are also not fully understood. Therefore, this study is expected to contribute to filling these knowledge gaps and provide helpful information for developing nutrient management strategies and improving rice tolerance to drought stress.

This study focuses on rice's early growth and physiological responses to a specific combination of ammonium sulfate and *Chromolaena odorata* under drought stress. In contrast to most previous studies that focused on the final growth and yield phases of the plant, this study paid particular attention to the germination and early growth phases of rice, which are critical periods in determining the success of plant stabilization and its yield potential (Cao et al., 2018). In addition, this study also explored the physiological mechanisms underlying the response of rice to these fertilizer combinations under drought stress conditions by analyzing various physiological parameters such as chlorophyll, carotenoid, and proline contents. Understanding these physiological mechanisms is important to reveal the adaptation strategies of rice to drought stress and identify physiological characteristics that can be used as markers of rice resistance to abiotic stress (Hassan et al., 2023).

The primary objectives of this study were to evaluate the early growth and physiological responses of rice treated with ammonium sulfate and *Chromolaena odorata* under drought-stress conditions. More specifically, the objectives of this study: (1) analyze the effect of ammonium sulfate and *Chromolaena odorata* fertilization on early growth parameters of rice, such as maximum growth potential, germination rate, vigor index, germination power, and growth speed, under field capacity and drought stress conditions; (2) evaluate the physiological response of rice to ammonium sulfate and *Chromolaena odorata* fertilization under drought stress conditions, by measuring chlorophyll, carotenoid, and proline contents; and (3) identify the most effective fertilization combination in improving early growth and physiological resistance of rice to drought stress. The results will be used to develop more effective nutrient management strategies to increase rice productivity and sustainability in a changing climate.

2. Materials and Methods

The research was conducted from July to August 2020 in the Faculty of Agriculture, Universitas Muhammadiyah Gresik greenhouse, 12 meters above sea level. The primary materials used in this research are the rice cv. Inpago 11. Inpago 11 has resistance to drought. Microclimate data obtained during the research include the average daily temperature between 27 °C to 37 °C with air humidity between 45% to 87% observed using a digital thermohygrometer. The type of soil used is latosol soil. Soil N content is 0.08%, and soil moisture content is 11%.

The experiment was designed in Split Plot Randomized Design (SPRD) with two factors. The main plot consisted of three fertilization types: non-fertilization, ammonium sulfate & *Chromolaena odorata*, and only ammonium sulfate. The subplot of the percentage of water supply based on the field capacity of the soil consisted of two levels, including 100% water field capacity (WFC) and 50% WFC. The treatment was repeated five times. Water stress was applied in the initial vegetative phase and maximum vegetative phase.

2.1. Soil requirement per polybag

The need for dry soil per polybag is 25 kg polybag⁻¹ with a soil moisture content of 11%, assuming a soil content weight of 1.1 g cm⁻³. The following is the calculation of the dry weight requirement of soil/polybag, namely:

$$\text{Polybag volume} = \pi r^2 t \quad (1)$$

$$\text{Soil content weight} = \frac{\text{absolute dry soil weight}}{\text{polybag volume}} \quad (2)$$

$$\text{air - dry soil weight} : \frac{(100 + \text{soil moisture content})\%}{100\%} \times \text{absolute dry soil weight} \quad (3)$$

2.2. Ammonium sulfate fertilizer requirement

The recommended dose of ammonium sulfate fertilization for rice plants is 100 kg ha⁻¹. Ammonium sulfate fertilizer required by plants.polybag⁻¹, with a polybag size of 30 cm x 30 cm is 1 g polybag⁻¹, with the following dosage calculation:

$$1 \text{ ha soil volume} = \text{land area} \times \text{soil depth} \quad (4)$$

$$1 \text{ ha soil weight} = \text{soil volume} \times \text{soil specific gravity} \quad (5)$$

$$\text{fertilizer requirements. Polybag}^{-1} = \frac{\text{soil weight in polybag}}{\text{soil weight ha}^{-1}} \times \text{recommended rate of fertilizer} \quad (6)$$

Description: Specific gravity of soil: 1.3 kg dm⁻³; Land area per Ha is 10 000 m²; Soil depth is 20 cm

2.3. Water requirements

The water requirement, at field capacity (FC) of 45% soil water with 11% soil moisture content (MC) is 7.9 liters. So, the 50% water stress treatment is 3.95 liters, and the 80% water stress treatment is 1.58 liters. The following is the calculation of water requirements at the field capacity of soil water per polybag is

$$\begin{aligned} \text{water requirement. Polybag}^{-1} \\ \text{at field capacity} \\ \text{(water weight)} \end{aligned} = \frac{(\text{groundwater FC} - \text{groundwater MC})\%}{100\%} \times \text{absolute dry soil weight} \quad (7)$$

$$\begin{aligned} \text{water requirement Polybag}^{-1} \\ \text{(water volume)} \end{aligned} = \frac{\text{water weight}}{\text{water density}} \quad (8)$$

Description: Density of water = 1 g cm⁻³

2.4. *Chromolaena odorata* needs

Chromolaena odorata is applied in liquid form. *Chromolaena odorata* extract is made by chopping fresh *Chromolaena odorata* leaves into 2-3 cm sizes. Furthermore, it is dried and finely ground in powder form. *Chromolaena odorata* extract is made using 10 kg of *Chromolaena odorata* powder macerated into 50 liters of water, stirring every 6 hours. The application of *Chromolaena odorata* extract per polybag is 200 ml with a ratio of water and *Chromolaena odorata* extract of 15:1.

2.5. Growth observation parameters

Observation parameters were carried out in the early growth phase at the age of 10 days (the days after planting) and the optimum vegetative phase at the age of 40 days.

2.6. Maximum growth potential (MGP) (%)

In the early growth phase, the maximum growth potential (%) was observed at the age of 7 dap, namely by calculating:

$$\text{MGP (\%)} = \frac{\sum \text{growing seeds}}{\sum \text{planting seeds}} \times 100\% \quad (9)$$

2.7. Germination Rate (days)

$$\text{germination rate} = \frac{N1T1 + N2T2 + \dots \dots \dots + N10T10}{\sum \text{total seeds germinated}} \quad (10)$$

Notes: N=number of seeds germinated each day; T=number of time between the beginning of the test and the end of the observation time.

2.8. Vigor index (VI)(%)

Vigor index (%), Observation of the vigor index is carried out on the number of normal sprouts on the *first count*, namely on day 5 (ISTA, 2010), namely

$$VI (\%) = \frac{\sum \text{first count normal sprouts}}{\sum \text{planting seeds}} \times 100\% \quad (11)$$

2.9. Germination potency (GP) (%)

Germination (%) was obtained by counting the number of seeds germinating normally (GN) at 5 dap and 7 dap. The formula calculated seed germination:

$$GP (\%) = \frac{\sum \text{1st count GN} + \sum \text{2nd count GN}}{\sum \text{planting seeds}} \times 100 \quad (12)$$

Description: GN = germinated normally

2.10. Growth rate (%/ethmal)

The growth rate was calculated daily for 7 days. The seeds were usually grown. The growth rate was calculated as follows:

$$\text{Growth rate} = \left(\frac{\text{germinated normally}}{\text{ethmal}} \% \right) = \sum_0^{tn} \frac{N}{t} \quad (13)$$

Description: t = observation time; N = percentage of germinated normally at each observation time; tn = final observation time (day 7), 1 *ethmal* = 1 day.

Observations of shoot length when the plants were 10 days old. Observations of plant adaptive responses were made at the optimum vegetative growth of rice plants at 40 days, namely the content of chlorophyll a, chlorophyll b, total chlorophyll, carotene, and proline.

2.11. Chlorophyll and carotenoid measurement

The materials used to analyze the chlorophyll and carotenoid content consisted of paddy leaves, and the James method (Smith and Benitez, 1955) was used under drought-stress conditions. Absorbance measurements using a UV spectrophotometer were performed to determine drought stress resistance at wavelengths (λ) of 649 nm and 665 nm, with three replicates per sample. Chlorophyll content was calculated as follows:

$$\text{Chlorophyll a} = 12.25 \lambda_{663} - 2.79 \lambda_{649} \quad (14)$$

$$\text{Chlorophyll b} = 21.50 \lambda_{649} - 5.10 \lambda_{663} \quad (15)$$

2.12. Proline measurement

The method of Bates et al. (1973) determined the proline content.

2.13. Statistical analysis

Results are expressed as the mean of three replicates in each individual. Data were analyzed by two-way analysis of variance (ANOVA) using Smartstat software (Setiawan, 2023). Multiple treatments were compared using Fisher's LSD Range Test at a 5% error level.

3. Results and Discussion

Early growth of rice showed variable responses to ammonium sulfate ((NH₄)₂SO₄) fertilization and its combination with *Chromolaena odorata* (Cho) under field capacity (100% WFC) and drought stress (50% WFC) conditions. Table 1 presents the average values of various rice early growth parameters, such as maximum growth potential, germination rate, vigor index, germination potency, and growing speed, as affected by fertilization treatments and water availability conditions. In general, (NH₄)₂SO₄ + Cho fertilization gave the best results in maximum growth potential, vigor index, and growing speed parameters compared to other treatments at 100% WFC, while at 50% WFC, differences between fertilization treatments were significant in most parameters (Table 1). These results are in line with previous studies, which showed that the combination of inorganic and organic fertilizers can have a positive effect on the initial growth of plants under optimal conditions, but the impact can be reduced under abiotic stress conditions (Iqbal et al., 2021; Baquy et al., 2022).

Table 1. Average early rice growth at field capacity and stressed to sulfuric acid and *Chromolaena odorata* fertilization

Fertilizer	Maximum Growth Potential (%)		Germination Rate (Days)		Vigor Index		Germination Potency (%)		Growing Speed (%.etmal) ⁻¹	
	WFC (%)		WFC (%)		WFC (%)		WFC (%)		WFC (%)	
	100	50	100	50	100	50	100	50	100	50
Non	45.71b B	40.48b A	5.63b A	5.73a A	25.71b A	23.81a A	48.10a B	40.72a A	15.26a B	13.06a A
(NH ₄) ₂ SO ₄ + Cho	48.09c B	39.99b A	4.41a A	5.45a B	29.77c B	22.38a A	50.60b B	41.43ab A	16.60c B	13.14a A
(NH ₄) ₂ SO ₄	39.05a B	38.06a A	4.42a A	5.45a B	23.21a A	23.81a A	52.98c B	42.38b A	16.00b B	13.50a A
LSD										
Fertilizer	1.02		0.479		2.493		1.562		0.479	
LSD WFC	0.88		0.415		2.630		1.363		0.441	

Notes: Mean values followed by the same letter are not significantly different according to the LSD Follow-up Test at 0.05 real level. Lowercase letters read vertical direction, comparing 3 fertilizers at the same stress. Capital letters read horizontally, comparing 2 stresses on the same fertilizer.

The fastest germination rate was achieved in treatments with fertilization under 100% WFC conditions, with values of 4.41 days in (NH₄)₂SO₄ + Cho and 4.42 days in (NH₄)₂SO₄ (Table 1). This indicates that fertilization positively affects germination speed, especially under drought-stress conditions. However, the (NH₄)₂SO₄ fertilization treatment alone produced the highest germination potency under 100% WFC conditions, with a value of 52.98% at 100% WFC. Under the stress condition of 50% WFC, the germination potential of seeds with (NH₄)₂SO₄ + Cho fertilization treatment was not significantly different from the same fertilization at 100% WFC. These results indicate that ammonium sulfate fertilization with *Chromolaena odorata* can increase the percentage of germinated seeds and accelerate the germination speed at 50%WFC conditions .

The vigor index and growing speed parameters showed similar response patterns to fertilization treatments and water availability conditions. (NH₄)₂SO₄ + Cho fertilization produced the highest values for both parameters at 100% WFC, with a vigor index value of 29.77 and a growing speed of 16.60%/ethmal (Table 1). However, at 50% WFC, the differences between fertilization treatments were insignificant for both parameters. These results indicate that combining ammonium sulfate and *Chromolaena odorata* can increase seedling vigor and growth speed under optimal conditions, but the effect is reduced under drought-stress conditions. It has been reported in several studies that the application of organic materials, such as By improving the physical, chemical, and biological properties of the soil, *Chromolaena odorata* can increase seedling vigor and early plant growth (Agustina et al., 2019; Munir, 2019; Fikrinda et al., 2021). However, the effectiveness of organic matter can decrease under water stress conditions due to the inhibition of the decomposition process and mineralization of nutrients.

Overall, this study’s findings indicate that the early growth response of rice to ammonium sulfate and *Chromolaena odorata* fertilization is influenced by water availability conditions (Gökkaya and Arslan, 2023). Under field capacity conditions (100% WFC), the fertilization of $(\text{NH}_4)_2\text{SO}_4 + \text{Cho}$ tended to give the best results for most early growth parameters. In comparison, under drought stress conditions (50% WFC), the differences between fertilization treatments were insignificant in most parameters. These results indicate that the effectiveness of fertilization, especially the combination of inorganic and organic fertilizers, may be reduced under abiotic stress conditions such as drought. Several studies have also reported that plant response to fertilization may differ depending on the water stress level, soil type, and other environmental factors (Al-Yasi et al., 2020; Fikrinda et al., 2021; Gusti et al., 2022; Ma et al., 2023). Therefore, when selecting fertilizer types and doses, the plant’s growing environment must be considered to optimize growth and productivity.

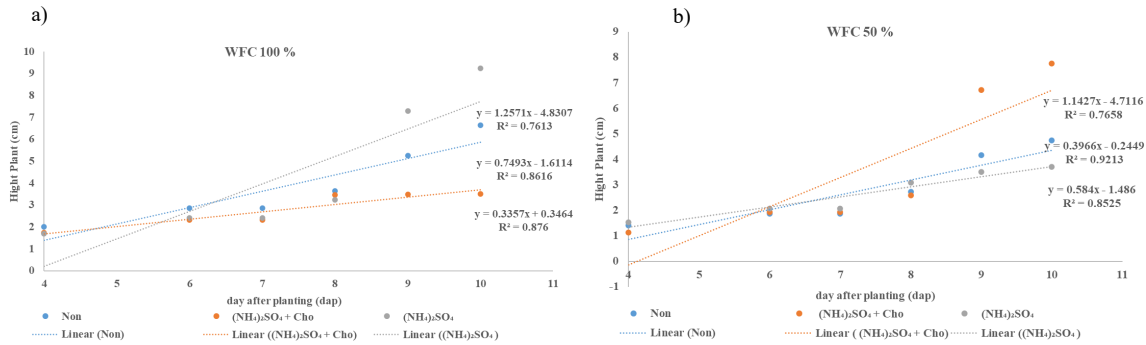


Figure 1. Plant height growth with sulfuric acid and *Chromolaena odorata* fertilization at (a) field capacity and (b) stress.

Based on Figure 1, it can be concluded several things about the effect of fertilizer application $(\text{NH}_4)_2\text{SO}_4$, $(\text{NH}_4)_2\text{SO}_4 + \text{Cho}$, and Non (no fertilizer) on plant height under 100% and 50% water field capacity (WFC) conditions.

At 100% WFC, the $(\text{NH}_4)_2\text{SO}_4 + \text{Cho}$ treatment alone produced the highest plants with line equation $y = 1.2571x - 4.8307$ and $R^2 = 0.7615$, followed by Non (no fertilizer) treatment with equation $y = 0.7465x + 1.6114$ and $R^2 = 0.8616$. The $(\text{NH}_4)_2\text{SO}_4 + \text{Cho}$ treatment produced the shortest plant height with the equation $y = 0.2357x + 0.2464$ and $R^2 = 0.8776$. All treatments’ high R^2 values (>0.75) indicated a strong relationship between plant age and height.

At 50% WFC, similar results were also observed where the $(\text{NH}_4)_2\text{SO}_4 + \text{Cho}$ treatment produced the highest plants with the equation $y = 1.1427x - 4.7116$ and $R^2 = 0.7656$, followed by $(\text{NH}_4)_2\text{SO}_4$ with $y = 0.3966x + 0.2449$ and $R^2 = 0.9213$. The Non-return treatment produced the lowest plants with $y = 0.5643x - 1.486$ and $R^2 = 0.8825$.

Overall, at both WFC levels, the addition of $(\text{NH}_4)_2\text{SO}_4$ fertilizer, both with Cho and without Cho, was able to increase plant height growth compared to the Non-treatment. The combination of $(\text{NH}_4)_2\text{SO}_4 + \text{Cho}$ gave the best results at both WFCs. However, plant height at 100% WFC tended to be better than 50% for all fertilizer treatments.

These results indicate the importance of fertilization using $(\text{NH}_4)_2\text{SO}_4$ and the addition of Cho to optimize plant height growth, as well as the need to maintain soil moisture content at optimal levels around field capacity (WFC 100%) so that plants can grow optimally water (Fikrinda et al., 2021; Ma et al., 2023).

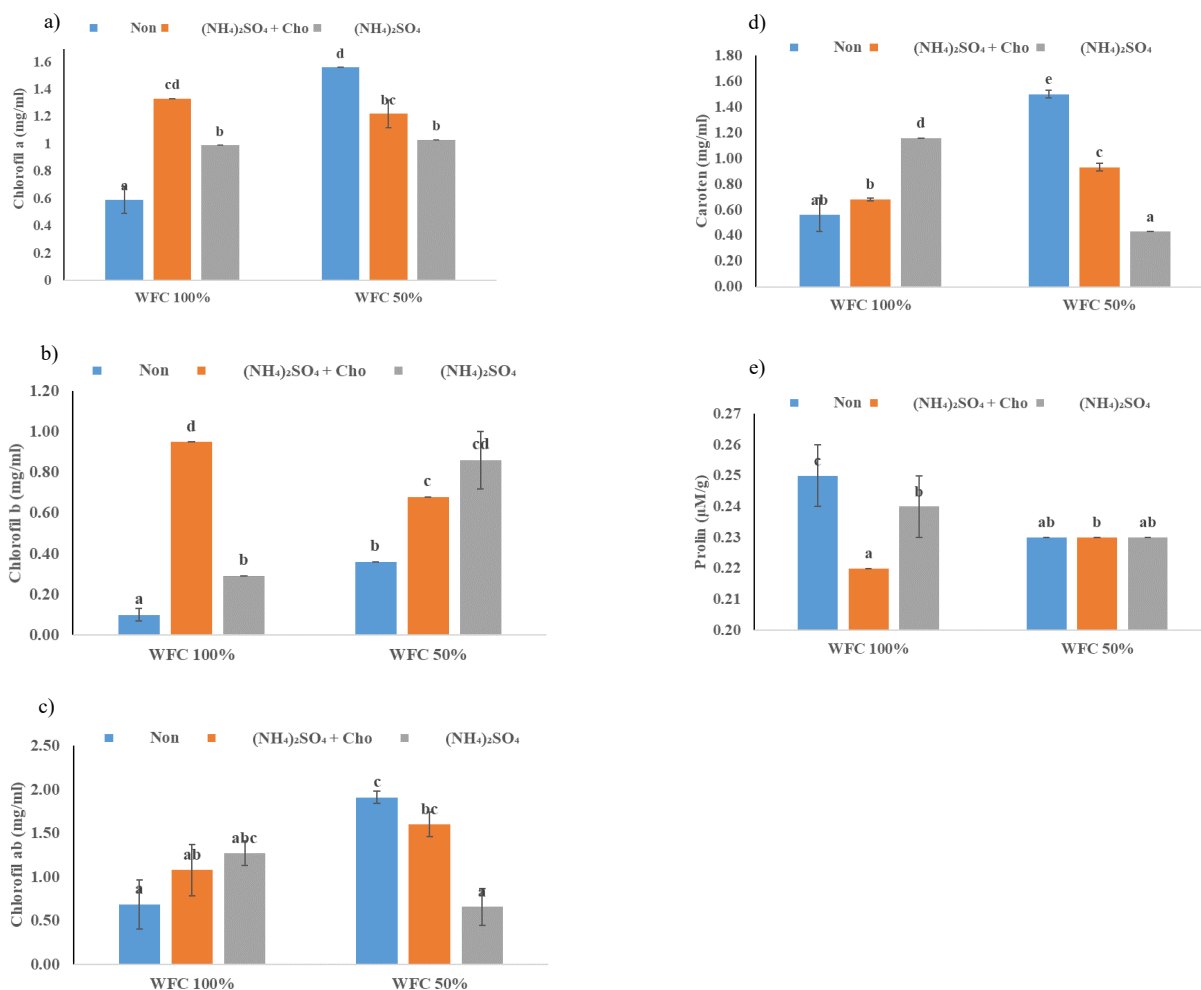


Figure 2. Physiological parameters of rice early growth: (a) Chlorophyll a, (b) Chlorophyll b, (c) Chlorophyll ab, (d) carotene, and (e) proline, under field capacity conditions and stressed to ammonium sulfate fertilization and *Chromolaena odorata*.

Figure 2 shows the physiological response of rice plants in general, indicating that (NH₄)₂SO₄ + Cho fertilization tends to provide the best physiological response under field capacity conditions. Except for the response on proline content. In contrast, differences between fertilization treatments are not always significant under drought stress conditions. This indicates that drought stress can reduce fertilization’s effectiveness in improving rice plants’ physiological status.

The highest chlorophyll a (Figure 2a) and chlorophyll b (Figure 2b) contents were achieved in the (NH₄)₂SO₄ + Cho treatment under field capacity conditions, with values of 1.6 mg g⁻¹ and 0.8 mg g⁻¹ respectively, significantly different from the other treatments. However, under drought-stressed conditions, the differences in chlorophyll a and b contents among fertilizer treatments were insignificant (Rahimi et al., 2023). A similar pattern was also observed in total chlorophyll content (Figure 2c), where (NH₄)₂SO₄ + Cho fertilization produced the highest value (1.2 mg g⁻¹) under field capacity conditions but under drought stress conditions was not significantly different from the other treatments. Chlorophyll is the main photosynthetic pigment, which is important in light absorption and energy conversion. A decrease in chlorophyll’s content under drought stress can impair plants’ photosynthetic capacity, which has impacted growth and Productivity (Gujjar et al., 2020).

Carotenoids (Figure 2d) are secondary pigments involved in photoprotection and abiotic stress response. Under field capacity conditions, (NH₄)₂SO₄ + Cho fertilization produced the highest carotenoid content (2.05 mg g⁻¹), which is significantly different from the other treatment options. However, under drought-stressed conditions, the difference in carotenoid content between fertilization treatments was insignificant. The increase in carotenoid content under drought stress conditions may

reflect the plant's adaptation mechanism to protect the photosynthetic apparatus from oxidative damage (Tiwari et al., 2021).

Proline (Figure 2e) is an amino acid that accumulates in plants in response to abiotic stress, acting as an osmoprotectant and antioxidant. Under field capacity conditions, it was not significantly different. In proline content between fertilizer treatments. However, under drought stress conditions, $(\text{NH}_4)_2\text{SO}_4 + \text{Cho}$ and $(\text{NH}_4)_2\text{SO}_4$ alone produced higher proline content ($0.26 \mu\text{mol g}^{-1}$ and $0.25 \mu\text{mol g}^{-1}$, respectively) compared to the control (Non), although the difference was not significant. Exogenous proline application increases the activity of antioxidant enzymes such as superoxide dismutase (SOD), peroxidase (POD), and catalase (CAT). It increases the content of total soluble protein (TSP), leaf proline, and glycine betaine, which contribute to the increase in chlorophyll content and yield of rice plants under drought stress (Hanif et al., 2021; Urmi et al., 2023). The proline accumulation under drought stress conditions may contribute to plants maintaining cell turgor and protecting cellular structures from damage (Ibrahim et al., 2022).

According to the statistical analysis in Figure 2, $(\text{NH}_4)_2\text{SO}_4 + \text{Cho}$ fertilization consistently gave the best physiological response under field capacity conditions, compared to the other treatments, with significant increases in chlorophyll a, chlorophyll b, total chlorophyll, and carotenoids. However, under drought stress conditions, the effectiveness of fertilization in improving plant physiological status was reduced, with insignificant differences between treatments for most parameters. This emphasizes the importance of fertilizer management that is adaptive to water availability conditions to optimize rice plants' physiological response and productivity.

Conclusion

This study showed that combined fertilization of ammonium sulfate ($(\text{NH}_4)_2\text{SO}_4$) and *Chromolaena odorata* (Cho) gave the best results on early growth and physiological responses of rice under field capacity conditions (100% WFC), with significant increases in growth parameters and photosynthetic pigment content. However, the effectiveness of this fertilization was reduced under drought stress conditions (50% WFC), where differences between treatments became insignificant for most parameters. Proline accumulation increased under drought stress conditions, especially with $(\text{NH}_4)_2\text{SO}_4 + \text{Cho}$ and $(\text{NH}_4)_2\text{SO}_4$ fertilization alone, suggesting a plant adaptation mechanism to stress. These results emphasize the importance of fertilizer management that is adaptive to water availability conditions to optimize growth, physiological responses, and productivity of rice plants, as well as the potential of combinations of inorganic and organic fertilizers in increasing rice resilience to drought stress. The implications of this study indicate that fertilization strategies need to consider the conditions of the plant-growing environment, especially water availability, to face the challenges of climate change and limited water resources in rice production.

Ethical Statement

Ethical approval is not required for this study

Conflict of Interest

The authors report there are no competing interests to declare.

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Author Contributions

Concept – R.A.; Design – R.A.; Supervision – E.S.R.; Materials – R.A., R.F.; Data Collection and Processing – R.A., R.F.; Analysis and Interpretation – R.A., E.S.R.; Writing – R.A.; Critical Review – E.S.R. Translator – H.A.

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