Special Issue: Novel Advances in Agricultural Science and Technology for Sustainable Farming in Tropical Region

Drought Stress Effect on Red Onion: Morphological, Physiological, and Yield Attributes

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Abstract | Shallot (*Allium ascalonicum* L.), the most significant plant in the Alliaceae family, is used extensively worldwide. The three main objectives of this study are to: (i) ascertain how drought stress affect shallot growth and yield, as well as water usage efficiency, (ii) N levels affect shallot growth and yield, as well as water usage efficiency; and (iii) ascertain which combination of N levels and water deficit maximizes shallot crop yields. The shallot experiment employed a completely randomized design (CRD) and a 3 × 3 factorial design with three replications. First factor was 25 % of field capacity (FC1); 50 % of field capacity (FC2) and 75 % field capacity (FC3) is attributed to this factor. The second factor is nitrogen fertilizer, namely N1 = 100 kg N ha^{-1}, N2 = 150 kg N ha^{-1}, and N3 = 200 kg N ha^{-1}. The study's findings demonstrated that the effects of nitrogen fertilizer and drought stress interacted to affect plant height, leaves number, tiller number, tuber weight, and crop production. water use efficiency (WUE), relative leaf water content (RLWC), and growth rate (CGR). Onion diameter, harvest index, and proline did not exhibit any relationship between nitrogen fertilizer and drought.

Received | May 29, 2024; **Accepted** | February 04, 2025; **Published** | April 02, 2025
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Citation | Purbajanti, E.D., D. Safitri, M.A. Wedyan, D. Roeswitawati, E.S. Savitri, S. Yuniati and T.M. Yuhana. 2025. Drought stress effect on red onion: Morphological, physiological, and yield attributes. *Sarhad Journal of Agriculture,* 39 (Special issue 1): 91-100.
DOI | https://dx.doi.org/10.17582/journal.sja/2023/39/s1.91.100
Keywords | Abiotic stress, *Allium ascalonicum* L., Bulb onion, Nitrogen fertilizer, Shallot, Water deficit

Introduction

Shallots (*Allium ascalonicum* L) is a plant that is usually planted in the lowlands and has shallow roots so it is sensitive to the environment (Ginting *et al*., 2024). Several shallot cultivars are superior cultivars that are widely cultivated by farmers in various regions in Indonesia. The Bima cultivar is a shallot cultivar originating from Brebes (Central Java) and is most often cultivated by farmers in various provinces in Indonesia because of its good adaptability in various regions in Indonesia (Manurung *et al*., 2022).

The factor that influences optimal plant growth is the environment. One of the influencing environmental factors is water (Henik *et al*., 2023; Yetagesu *et al*., 2020). Water has an important role for plants, namely as a basic material for plant metabolism, plays a role in respiration and photosynthesis, a solvent in the soil for plant nutrition, regulates temperature through transpiration, regulates cell turgidity, and is involved in transporting metabolites from roots to leaves (Hendro *et al*., 2022; Sansan *et al*., 2024). Shallots are plants that require sufficient water and are susceptible to drought or inundation (Ginting *et al*., 2024). The most common effect of drought stress is a reduction in water potential, turgor pressure in growing cells, and thus a lack of turgor pressure necessary for their growth. Lack of water accelerates cell differentiation. Under drought stress, root, stem, leaf and fruit growth decreases (Bekele *et al*., 2023). Under these conditions, not all plant organs are affected equally. Typically, due to drought stress, the leaf to stem ratio decreases; Older leaves and leaves exposed to shade usually die more quickly, slowing tillering and increasing tiller mortality (Gedam *et al*., 2021). A plant's reaction to water stress might involve a variety of defensive mechanisms, changes to physiology, morphology, anatomy, or biochemistry, as well as processes for plant growth and development (Gokçe *et al*., 2023). Lack of water can cause abiotic stress in basil plants, which can affect their physiological and anatomical structure, productive balance, and capacity to create secondary metabolites (Wakchaure *et al*., 2023). Applying nitrogen fertilizer is one tactic to lessen the detrimental impacts of water scarcity on shallots. An essential macronutrient for onion growth, nitrogen (N) affects the production of biomass and bulb output. Onion productivity, however, might be adversely affected by an excessive or insufficient N application (Chaudhry *et al*., 2020). It takes effective N management techniques to maximize onion growth and reduce environmental effects. Studies have demonstrated the significance of nitrogen fertilization in augmenting onion growth and yield, specifically in both typical and arid environments (Geisseler *et al*., 2021). For example, using nitrogen fertilizers at the right amounts helps preserve turgor potential and increase leaf water content, which strengthens cell membrane integrity and increases plant resistance to drought stress (Alzoheiry, 2024) In their study of the impact of drought on onion yield, Ropelewska *et al*. (2022) discovered that drought stress during the last stages of growth caused the onions to mature earlier than intended, lowering the yield. The objectives of this research were to: (i) ascertain how drought stress affect shallot growth and yield, as well as water usage efficiency, (ii) N levels affect shallot growth and yield, as well as water usage efficiency; and (iii) ascertain which combination of N levels and water deficit maximizes shallot crop yields. The research result from various studies on shallot cultivation have significant implication for the development of shallot farming practices among farmers. Here the implication (i) Risk management and behavior (Sriyadi ang Yekti, 2021), (ii) cultivation technology (Tome *et al*., 2022), (iii) land management (Irawan *et al*., 2024), (iv) sustainability, production stability, and regional specific (Parmawati *et al*., 2021).

Materials and Methods

Plant materials and growth conditions

The research was carried out from June to December 2022 at the Greenhouse Laboratory and Plant Ecology and Production, Diponegoro University, Faculty of Animal Husbandry and Agriculture, Semarang. The research location is located at latitudes 6° 5534" to 7° 0704" in the south and 110° 1620" to 110° 3029" in the east, at an altitude of 125 m a.s.l., with an average daily temperature of 22 °C to 34 °C, air humidity of 75 %, and annual rainfall of 3 359 mm.

Implementation of research

The shallot experiment used a completely randomized design (CRD) and a 3 × 3 factorial design with three replications. Specifically, FC1 maintains a 25 % field capacity, FC2 a 50 % field capacity, and FC3 a 75 % field capacity. Dryness is the primary component of Oxisol soil. The second element is the nitrogen fertilizer, more precisely N1 = 100 kg N ha^{-1}, N2 = 150 kg N ha^{-1}, and N3 = 200 kg N ha^{-1}. After excavating to a depth of 2 cm, shallot seeds are inserted into the hole and covered with dirt. Following that, basic fertilization is done using manure at the dosage of 2 t ha^{-1} (8 g plant^{-1}), 100 kg ha^{-1} of SP 36 fertilizer (0.4 g plant^{-1}), and 100 kg ha^{-1} of KCl (0.4 g plant^{-1}) administered concurrently at planting time. Depending on the treatment, urea is administered between the ages of 2 wk and 4 wk. By not watering the plants until they keep water at 75 % FC, 50 % FC, and 25 % FC until harvest time, drought stress starts at the age of 2 wk after planting (WAP), or when the onion plants start to create bulbs.

Measurements and calculated parameters

- The height of shallot plants is measured in meter from the base of the plant touching the ground to the tip of the tallest leaf. Measurements were carried out every 2 WAP, until the end of the generative phase.
- Leaves number was calculated number of leaf plant^{-1}.
- Number of tiller (tiller plant^{-1}) was calculated the number of shallot seedlings is calculated from each shoot that has a leaf growing point that produces bulbs. Measurements were carried out at the end of the research.
- Fresh weight, was calculated all crops are in wet form.
- Tuber weight (g plant^{-1}), the weight of fresh tubers is calculated using a digital scale to produce tubers after harvest. Measurements were carried out at the end of the research, tuber diameter, was calculated average diameter of shallots per plant.
- Number of splited bulbs and number of splits in bulb were also counted. Economic yield (t ha^{-1}) was determined by taking the fresh weight of the bulbs. Fresh leaf, bulb and root weights were used to estimate biological yield (t ha^{-1}). Harvest index (%) was determined by Equation 1 (Ali and Anjum, 2016).

$$\text{Harvest index (\%)} = (\text{Economic yield} / \text{Biological yield}) \times 100 \quad \ldots(1)$$

Crop growth rate (CGR) - Watson (1956)

The CGR explains the dry matter accumulated per unit land area per unit time (g m^{-2} d^{-1}) (W2 –W1) CGR = ρ (t2 − t1) Where, W1 and W2 are whole plant dry weight at time t1 – t2 respectively ρ is the ground area on which W1 and W2 are recorded. CGR of a species are usually closely related to interception of solar radiation and calculated at harvest stages, by Equation 2.

$$CGR = \frac{W2-W1}{t2-t1}\ gm^{-2}day^{-1} \quad \ldots.(2)$$

Where, W1 and W2 were the dry weight of the aerial plant per unit area gained at time t1 and t2 respectively.

Proline analysis

Analysis of proline content was carried out using the Purbajanti *et al.* (2017) method. An amount of 0.5 g of plant roots were ground in 10 mL of 3 % sulfosalicylic acid solution, then filtered using Whatman paper No. 1. As much as 2 mL of filtrate is reacted in 2 mL of ninhydrin acid. (Ninhydrin acid is made by heating 1.25 g of ninhydrin in 30 mL of glacial acetic acid and 20 mL of phosphoric acid until it dissolves). The filtrate and ninhydrin acid are added to glacial acetic acid then heated at 100 °C for 1 h. The reaction is ended by inserting the test tube containing the filtrate into a beaker filled with ice. Toluene was added to the mixture of filtrate, ninhydrin and glacial acetic acid and stirred with a stirrer for 15 sec to 20 sec to form two layers of different colored liquid. Red colored toluene containing proline is taken with a pipette then put into a cuvette and the OD is read with a wavelength of 520 nm. Calculation of proline levels is carried out by making a proline standard, making a stock solution of 2.5 μM and diluting it with 3 % sulfosalicylic acid. Dilution is intended to obtain variations in proline concentration. The solution is reacted with ninhydrin acid and glacial acetic acid. The solution was put into a cuvette and the OD was read with a wavelength of 520 nm.

Calculation of relative leaf water content and water use efficiency

Relative leaf water content (RLWC) was observed in the morning, leaf samples were taken randomly, and determined gravimetrically before dawn. Ten expanded flag leaves were collected to determine fresh weight (FW). Wet leaf weight (TW) was determined after the leaves were floated in distilled water in a closed container at 4 °C in a dark room for 24 h. Dry weight (DW) was determined for the same leaves after drying in an oven for 48 h at 70 °C. RLWC is calculated as in Equation 3. while WUE as in Equation 4 (Purbajanti *et al.*, 2020).

$$RLWC\ (\%) = \frac{(FW-DW)}{(TW-DW)} \times 100\ \% \quad \ldots(3)$$

$$WUE = \frac{\text{Plant dry weight (g)}}{\text{Water requirements of each plant (mm)}} \quad \ldots(4)$$

Statistical analysis

One-way analysis of variance was used to assess the data, then Tukey's test with a significance level of 5 % (Adinurani, 2016; Setyobudi *et al.*, 2024).

Results and Discussion

The study's findings indicate that the effects of nitrogen fertilizer and field capacity on plant height interact. The fertilization of 200 kg N ha^{-1} and 75 %

field capacity (plant height 50.66 cm) was much higher (Figure 1a). The shallot plants grown were taller than those found in Tambunan *et al.* (2014) study, which measured the height of shallot plants grown on a medium consisting of a mixture of sand and vermicompost at 36.45 cm.

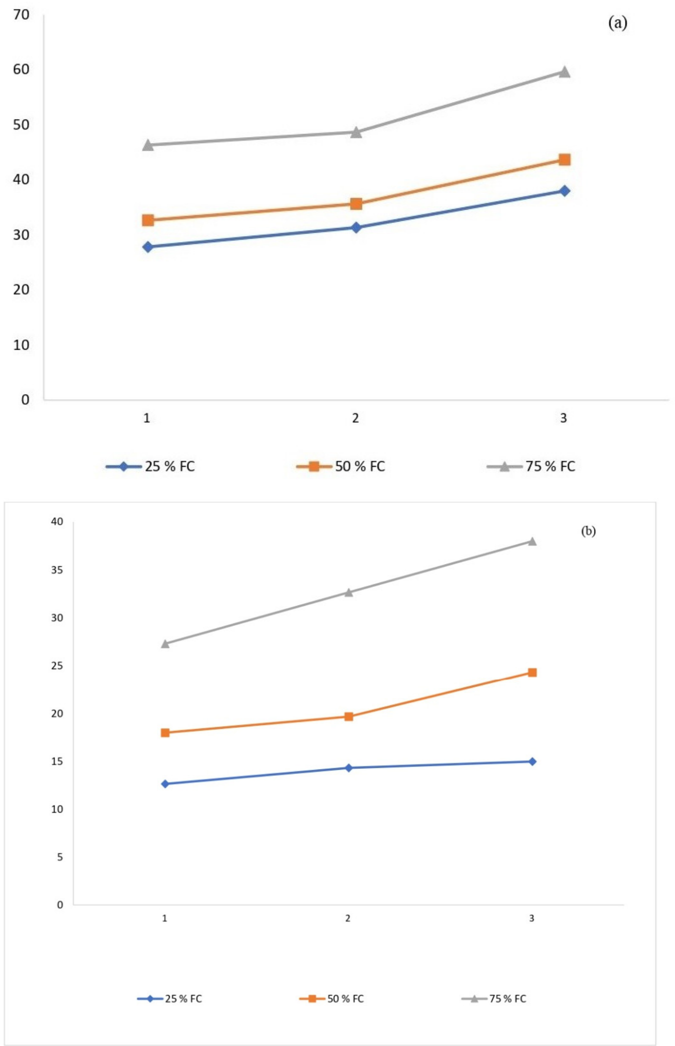


Figure 1: *Onion plan height (a) and leaves number (b) effect of drought stress and dosage nitrogen fertilizer.*

The number of leaves on a shallot plant exhibits a relationship between nitrogen fertilizer and field capacity in proportion to plant height. In the meantime, 200 kg N ha^{-1} of fertilization (38 leaves) was much more than 50 % of the field's capacity (24.3 cm) and 25 % of its capacity (15.0 cm) (Figure 1b). Things that influence plant height are variety, light, wind, water, temperature, soil nutrient content, soil type and soil pH. If all things are considered the same, then plant height is greatly influenced by the conditions of soil field capacity, variety and fertilization given. Plants that lack water often experience reduced growth or even wilt. Plants that lack water tend to grow shorter. Nitrogen is very important for plant growth, especially for the development of leaves and stems. Nitrogen deficiency can cause plants to grow slower and shorter. Plants experiencing drought will show reduced growth even though N fertilizer is given to stimulate growth (Zegeye *et al.*, 2024). Additionally, nutrient absorption decreases in drought-stressed environments. This is partly because of lower soil moisture, which inhibits the pace at which nutrients diffuse from the soil matrix to the absorbing root surface and limits the growth and development of leaves (Nourbakhsh and Cramer, 2022).

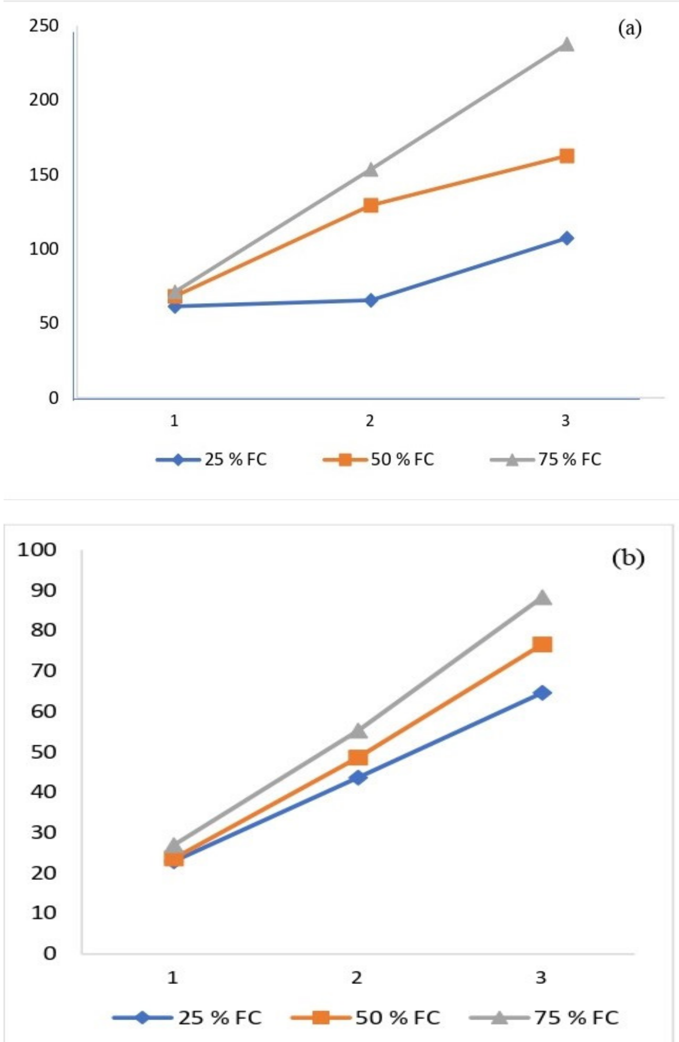


Figure 2: *Onion biomass (a) and weights of shallot bulbs (b) effect of drought stress and dosage nitrogen fertilizer.*

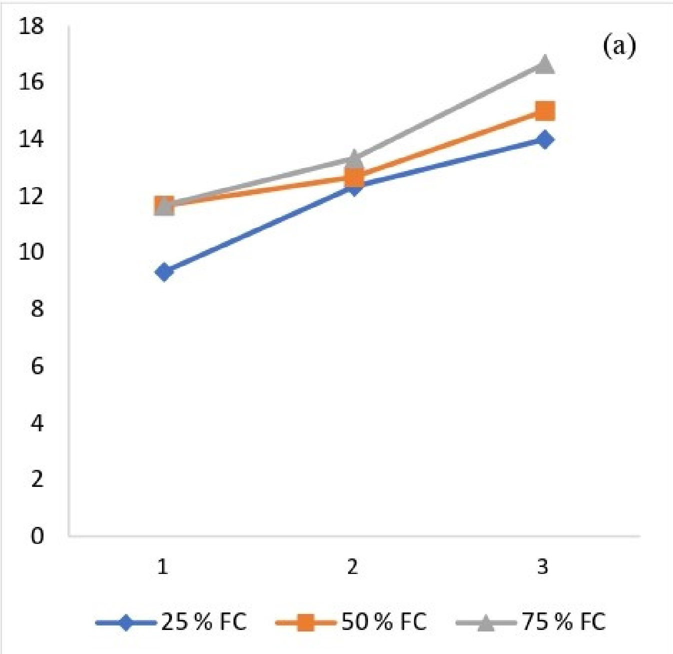
The findings demonstrated that the effects of nitrogen fertilizer and field capacity on biomass weight interacted. In the meantime, the maximum biomass weight was reached by 75 % of field capacity (237.77 g) with fertilization of 200 kg N ha^{-1} (Figure 2a). Relatively high levels of nutrients are required for optimum growth and development at early stage and adequate moisture supply is most suitable during the early growth Additionally, in drought-stressed conditions, nutrient absorption diminishes (Alemu *et al*., 2022). In contrast to studies conducted by Fadila *et al*. (2023), the biomass yield for three onions' worth of seeds per polybag was 47.49 g. Field capacity and nitrogen fertilizer have an effect with bulbs weight. The bulbs weight of 88.33 g was conducted at 200 kg N ha^{-1} and 75 % of field capacity (Figure 2b). Water deficiency is mostly associated with modifications in plant metabolism. The reduction or loss of photosynthetic pigment production is one of them. The canopy growth dynamics, which were greatly influenced by limited water supply during development and bulbification stages leading to restricted cell multiplication and expansion, and stomatal control in response to water stress, are consistent with this yield declination result (Wakchaure *et al*., 2023).

Field capacity and nitrogen fertilizer have an interaction with the quantity of tillers. In the meantime, the number of tillers is 16.7 with fertilization of 200 kg N ha^{-1} and 75 % of field capacity (Figure 3a). The type and amount of fertilizer applied can enhance or inhibit tiller formation. While adequate fertilization promotes growth, excessive use can negatively impact plant health and reduce tillering (Zegeye *et al*., 2024). This is more than the 4.3 shallot seedlings per cluster found in the study of Kurnianingsih *et al*. (2018).

Fertilizer containing nitrogen and field capacity interact. Crop growth rate (3.2 g d^{-1}) at 75 % of field capacity (200 kg N ha^{-1}) (Figure 3b). One of the most significant environmental issues is a severe lack of water, which impedes crop maturation and expansion as well as the output and quality of agricultural goods (Henik *et al*., 2023; Suprapto *et al*., 2018).

The interactions between nitrogen fertilizer and field capacity are also included in the RWLS of shallot plants. 200 kg N ha^{-1} of fertilizer at 75 % field capacity (RWLS 73.2 %) is much more than 50 % of field capacity (66.8 %) and 25 % of field capacity (63.3 %) (Figure 4a). Plant dry weight, leaf water potential (ΨI), and relative leaf water content (RLWC) all sharply decline when water supply is reduced (Solouki *et al*., 2023).

The study's findings indicate that the efficiency of water consumption is influenced by both field capacity and nitrogen fertilizer. The maximum water usage efficiency (5 706.4 mL g^{-1} plant) was attained at 75 % of field capacity fertilization of 200 kg N ha^{-1} (Figure 4b). This was noticeably higher than 50 % of field capacity (2 342.4 mL g^{-1} plant) and 25 % of field capacity (515.2 mL g^{-1} plant). According to Caceres *et al*. (2024), stomata closure is a crucial resistance mechanism that is triggered by Amino Butyric Acid (ABA). This process reduces transpiration and thus stops water loss, increasing water use efficiency (WUE).



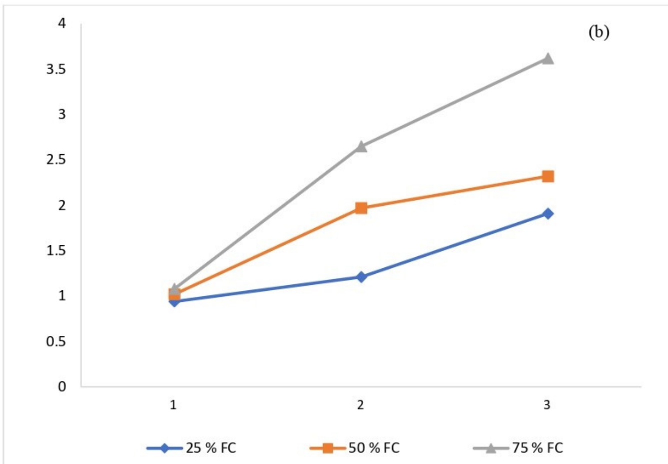


Figure 3: *Onion tiller number (a) and crop growth rate (b) effect of drought stress and dosage nitrogen fertilizer.*

Regarding bulb diameter, there was no relationship between nitrogen fertilizer and field capacity. The maximum tuber diameter in the 75 % of field capacity treatment (2.17 cm) was significantly different (Figure 5a). Regarding nitrogen fertilization, the maximum diameter (2.51 cm) was observed with 200 kg N ha^{-1} (Figure 5b). When compared to control, straining at first bolting drastically reduced the onion umbel's diameter (Ashagrie *et al*., 2021).

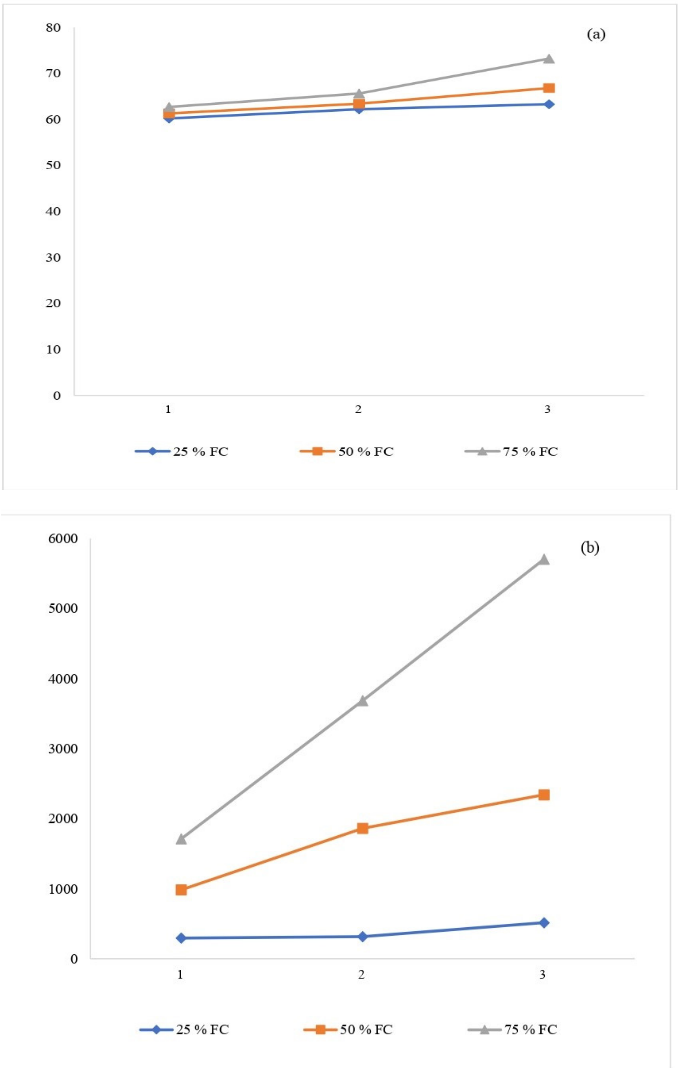


Figure 4: *Onion RWLS (a) and WUE (b) effect of of drought stress and dosage nitrogen fertilizer.*

There is no relationship between nitrogen fertilization and field capacity in the harvest index parameter. Every factor exhibits a discernible impact. 25 % of field capacity (57.61 %) achieves the maximum harvest index, which differs significantly from 50 % or 75 % of field capacity (Figure 6a). Regarding fertilization factor, the harvest index in the 150 kg N ha^{-1} treatment was 46.81%; this was not substantially different from the fertilization factor of 200 kg Nha^{-1}, which was 49.19 % (Figure 6b); nevertheless, it was considerably different from the fertilization factor of 100 kg N ha^{-1}. A harvest index value of more than 75 % indicated that most of the assimilated results from photosynthesis were stored in the form of bulbs (Shiferaw and Wyngard, 2019).

The proline interaction has a maximum value at 25 % of field capacity, which is notably different from 50 % or 75 % of field capacity, but otherwise has no real effect (Figure 7a). N fertilizer of 100 kg N per ha produces the highest proline compared to fertilizer of 150 kg N per ha or 200 kg N ha^{-1} (Figure 7b). This is consistent with the findings of Shanthi *et al.* (2023), which reported that plants exposed to drought create huge amounts of proline. In plants receiving nutritional supplementation, proline synthesis peaked under drought (Wang *et al.*, 2022).

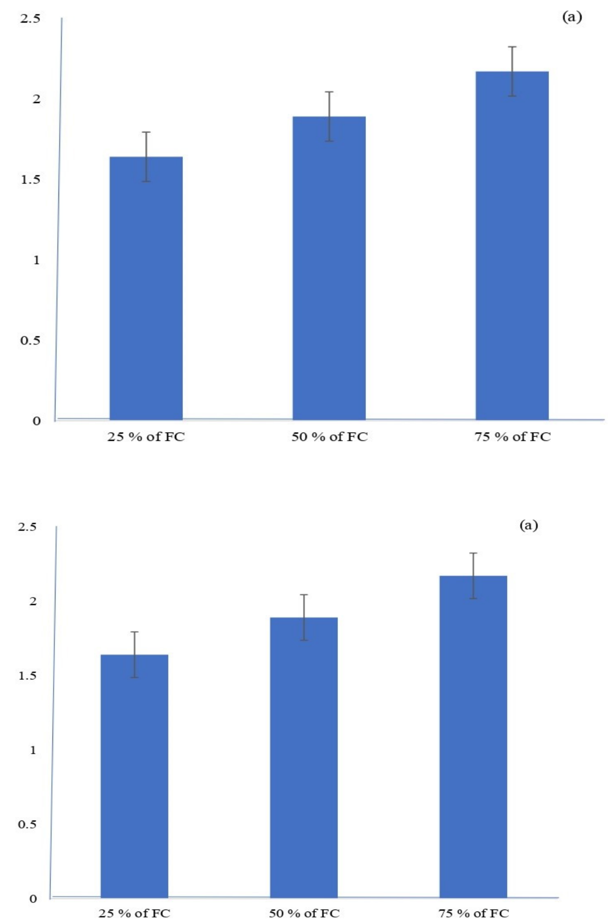


Figure 5: *Onion bulb diameter effect of field capacity (a) and dosage N fertilizer (b).*

The sedentary character of plants has forced them to develop complex mechanisms that allow them to survive and adapt to water shortages, as drought stress is particularly damaging to plant growth and development (Wang *et al.*, 2022). Plants are simultaneously subjected to a variety of abiotic stressors. concurrently, which may negatively affect development and productivity. The combination of these forces may worsen the harm done (Caceres *et al.*, 2024). According to Sansan *et al.* (2024) the root system is the first organ of plants to detect soil dryness. It then uses these signals to communicate with the shoots in different drought circumstances, eliciting diverse stress responses. My research on the growth characteristics of shallots under dry stress conditions yielded the following results: A decrease

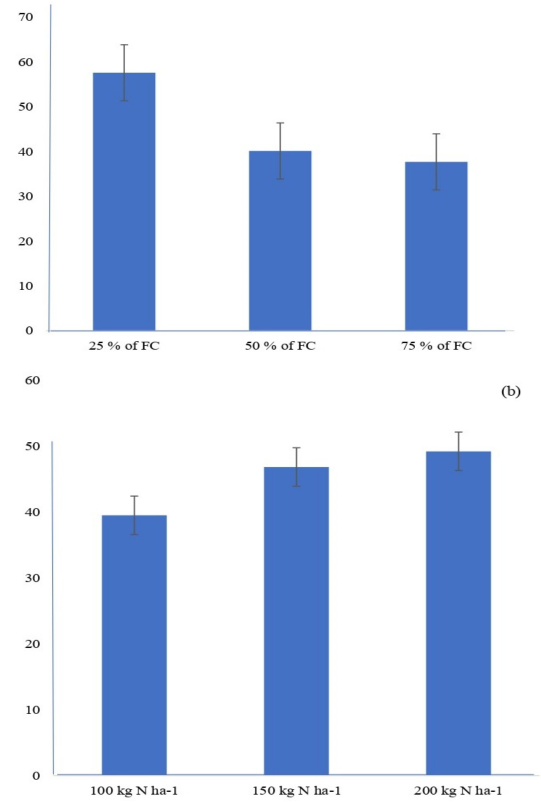


Figure 6: *Onion Harvest Index effect of field capacity (a) and dosage N fertilizer (b).*

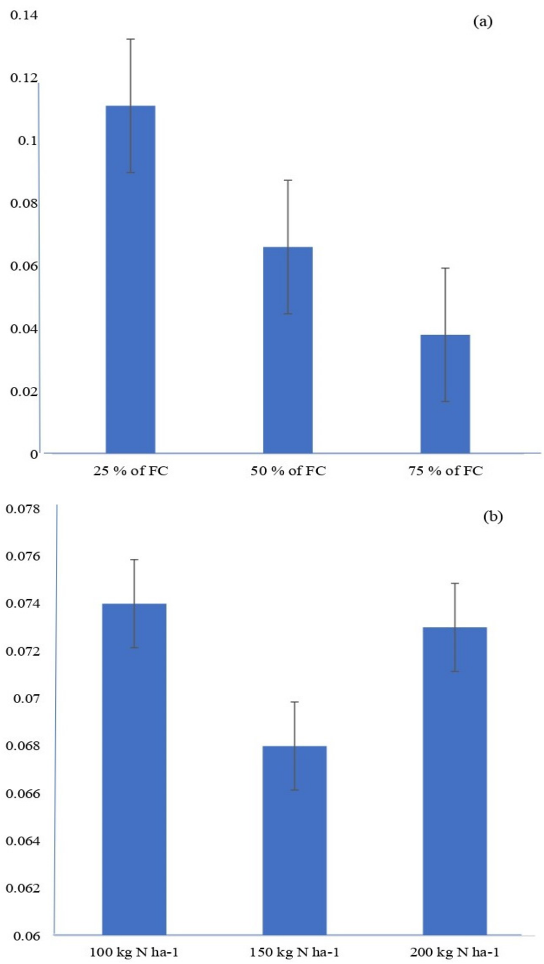


Figure 7: *Onion proline content effect of field capacity (a) and dosage N fertilizer (b)*

in height of 24.99 % (Figure 1a), a decrease in the number of leaves (60.52 %) (Figure 1b), a fall in the number of offspring (16.01 %) (Figure 3a), a decrease in biomass weight (54.85 %) (Figure 2a), and a decrease in tuber diameter (26.78 %) (Figure 5a). In situations where the field capacity was 75 %, 50 %, and 25 %, respectively, drought stress dramatically decreased the relative growth ratio by 67.2 %, 63.9 %, and 61.9 % (Figure 3b). The number of leaves decreases when the plant experiences water stress. This is thought to be because to reduce water loss through transpiration, plants respond by closing the stomata. However, stomata closure also inhibits the entry of CO_2 which is necessary for photosynthesis. The low rate of photosynthesis causes reduced assimilate for the formation and growth of new leaves (Ginting *et al*., 2024). Drought stress causes a decrease in water potential in the soil, making it difficult for roots to absorb the water and nutrients needed for the formation of new shoots. The plant's response to reducing the rate of transpiration and optimizing the distribution of assimilate is to reduce the number of tillers (Gedam *et al*., 2021). In particular, drought stress caused a considerable fall in the water leaf ratio, which may be influenced more by RLWC than by fertilizer. In terms of biomass output and transpiration rate, it is also a major determinant of crop productivity. Water will change the turgidity of the cell, which will change how the stomata open and close. If the stomata are blocked, less sunlight will be converted, which will impact the outcomes of photosynthesis. Plant transpiration is likewise impacted by this; in plants, more water will result in a higher rate of transpiration and vice versa (Pamungkas *et al*., 2022). Furthermore, there is a decrease in nutrient absorption when there is drought stress. Reduced soil moisture contributes to this by slowing the pace at which nutrients diffuse from the soil matrix to the absorbing root surface, which in turn restricts the growth and development of leaves (Nourbakhsh and Cramer, 2022). The best strategy is to use tolerant species, however other factors that affect a plant's resistance to environmental stress include type, conditions of the habitat, and availability of nutrients. Furthermore, this fertilizer can enhance the quantity and quality of plant growth characteristics while reducing the adverse effects of environmental stress (Geisseler *et al*., 2021).

Conclusion and Recommendations

The effect of drought stress (75 % FC) and dosage of nitrogen fertilizer (200 kg N ha^{-1}) significantly increased plant height, leaf number, onion biomass and weight of shallots bulbs, tiller number, CGR, RLWC and WUE. For onion bulb diameter, harvest index and proline were only single treatment factors that had an influence. The highest yield of Onion bulb diameter, harvest index was in the dry stress treatment of 75 % FC, and for the N fertilizer treatment at 200 kg N ha^{-1}, but different from the proline content, the highest value was at 25 % FC, also the highest proline content at 100 kg N ha $^{-1}$.

Acknowledgement

The authors would like to thank Diponegoro University for its financial support (No. 41/ UN 7.5.5.2/HK/2022). All so, the authors are grateful for the research facility provided by the Department of Agroecotechnology at the Faculty of Agriculture and Animal Science at Diponegoro University in Semarang, Indonesia.

Novelty Statement

The authors have seen on Google Scholar that there has been no research on dry stress combined with nitrogen fertilization on *Allium ascalonicum* L, which is present on *Allium cepa* L. The study's findings indicate that the efficiency of water consumption is influenced by both field capacity and nitrogen fertilizer. In the meantime, the maximum water use efficiency was reached at 75 % of field capacity (5 706.4 mL g^{-1} plant) with fertilization of 200 kg N ha^{-1}, which was noticeably higher than 50 % of field capacity (2 342.4 mL g^{-1} plant) and 25 % of field capacity (515.2 mL g^{-1} plant)

Author's Contribution

Endang Dwi Purbajanti: Conceptualized and designed the study, elaborated the intellectual content, performed literature search, manuscript preparation, and manuscript revision.
Dian Safitri: Performed literature search, manuscript preparation, and statistical analysis.
Mohammed Ali Wedyan, Dyah Roeswitawati, Evika Sandi Savitri, and Sri Yuniati: Manuscripts review and literature search.
Titan Memory Yuhana: Graphical data analysis.
All authors have read and approved the final manuscript.

Conflict of interest
The authors have declared no conflict of interest.

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