ARTICLE

# SBFPO Sociedade Brasileira de Floricultura e Plantas Ornamentais

# Morphological, biochemical, and maximum quantum efficiency of photosystem (Fv/Fm) in African marigold (*Tagetes erecta* L.) affected by two soil amendments: rice husk biochar and zeolite

Morfologia, bioquímica e eficiência quântica máxima do fotossistema II (Fv/Fm) em tagetes (*Tagetes erecta* L.) afetadas por dois melhoradores de solo: biochar de casca de arroz e zeólita

Rudabeh Ghaderi<sup>1</sup> 💿, Farzad Nazari<sup>1,</sup> 💿, Mahmoud Koushesh Saba<sup>1,\*</sup> 💿, Himan Nourbakhsh<sup>2</sup> 💿, Negin Nazari<sup>1</sup> 💿

<sup>1</sup> Department of Horticultural Science, Faculty of Agriculture, University of Kurdistan, Sanandaj, Iran
<sup>2</sup> Department of Food Science and Engineering, Faculty of Agriculture, University of Kurdistan, Sanandaj, Iran

**Abstract**: The need for a more efficient agricultural production system has arisen due to factors such as rapid urbanization, climatic anomalies, water scarcity, and declining water quality in modern times. Bedding plants are one of the integral parts of the landscape and the African marigold is one of the most common and particularly popular. This study investigates the effects of biochar and zeolite on the morphological and biochemical properties, as well as the photosynthetic performance, of African marigold (*Tagetes erecta* L.) plants. The experiment followed a completely randomized design with four treatments: soil mixture alone (control), soil mixture amended with 10% (w/w) biochar, or 25 g zeolite kg<sup>-1</sup>, or a combination of both. Results indicated that biochar had a significant positive effect on the maximum quantum efficiency of photosystem II (Fv/Fm). On the other hand, zeolite alone significantly improved various growth parameters, including root and stem length, weight, turgor, flower length, plant height, and Fv/Fm, when compared to the control group. However, the use of zeolite also resulted in reductions in chlorophyll content and dry flower weight. Interestingly, the combined treatment of biochar and zeolite led to a significant increase in total soluble carbohydrates. However, this combined treatment did not have a significant marigold growth and Fv/Fm parameters. These findings suggest that while biochar and zeolite have positive effects on specific aspects of African marigold growth and physiology, their combined application may not be optimal.

Keywords: bedding plants, chlorophyll content, landscape, soil amendments.

**Resumo:** A necessidade de um sistema de produção agrícola mais eficiente surgiu devido a fatores como urbanização rápida, anomalias climáticas, escassez de água e degradação da qualidade da água nos tempos modernos. As plantas anuais são integrantes da paisagem, sendo o tagetes uma das mais comuns e particularmente popular. Este estudo investiga os efeitos do biochar e zeólita nas propriedades morfológicas e bioquímicas, bem como no desempenho fotossintético das plantas de tagetes (*Tagetes erecta* L.). O experimento seguiu um delineamento inteiramente casualizado com quatro tratamentos: mistura de solo (controle), mistura de solo modificada com 10% (m/m) de biochar ou 25 g de zeólita kg<sup>-1</sup>, ou uma combinação de ambos. Os resultados indicaram que o biochar teve um efeito positivo significativo na eficiência quântica máxima do fotossistema II (Fv/Fm). Por outro lado, a zeólita isolada melhorou significativamente vários parâmetros de crescimento, incluindo comprimento das raízes e do caule, peso, turgor, comprimento das flores, altura das plantas e Fv/Fm, quando comparada ao grupo controle. No entanto, o uso da zeólita também resultou em reduções no teor de clorofila a e no peso seco das flores. Curiosamente, o tratamento combinado de biochar e zeólita levou a um aumento significativo nos carboidratos solúveis totais. No entanto, esse tratamento combinado não teve um impacto significativo na maioria dos outros parâmetros de crescimento e Fv/Fm. Esses resultados sugerem que, embora o biochar e a zeólita tenham efeitos positivos em aspectos específicos do crescimento e fisiologia do tagetes, sua aplicação combinada pode não ser ideal.

Palavras-chave: Plantas anuais, teor de clorofila, paisagismo, condicionadores de solo.

# Introduction

Landscapes, such as parks and gardens, are crucial elements of human society (Reid et al., 2022) and play a vital role in improving the quality of life (Addas, 2023). They have long been recognized as important for enhancing the overall quality of human life, as they contribute significantly to biological, environmental, social, and psychological activities (Hunter et al., 2019; Crouse et al., 2021; Reid et al., 2022). Bedding plants (seasonal flowers) hold a key position in landscape design, providing aesthetic appeal and enhancing the ambiance. They serve as temporary space fillers, adding to the beauty of the surroundings (Addas, 2023). Considering the importance of these flowers in the landscape, the nutritional management of these plants significantly affects their production and quality (Khan et al., 2023). Therefore, utilizing the appropriate nutrition regime and cultivation medium becomes crucial in achieving the highest yield and quality (Hassan et al., 2022). Environmental restrictions, high costs, and exclusivity of certain cultivation substrates, such as peat, have led to a growing trend toward cost-effective and easily accessible mineral and organic alternatives like zeolite and biochar (Bashir et al., 2019).

Zeolites, a group of minerals first discovered in 1756, have garnered attention from ornamental plant producers due to their unique properties, including ion exchange capacity, high water absorption, catalytic activity, stable crystal structure, and gas absorption capabilities (Nazari et al., 2007; Rahimi et al., 2021; Amirahmadi et al., 2022). These characteristics make zeolites a promising alternative to traditional peat moss as a growing medium for ornamental plants. Additionally, zeolites offer other beneficial effects in the agricultural sector, such as increasing fertilizer efficiency and gradual release, enhancing water retention capacity in the soil, mitigating drought stress, improving nutrient absorption, promoting plant growth and yield, and reducing pests and diseases (Boros-Lajszner et al., 2018; Amirahmadi et al., 2020).

Biochar, a stable, porous, and fine-grained carbon material, is produced through the pyrolysis of organic matter under oxygen-deprived conditions at temperatures ranging from 300 to 1000 °C (Farooq et al., 2021)cultivated mostly in a semi-arid environment and frequently confronts sporadic dry periods during different stages of plant growth, which drastically reduce its productivity. The current study was enacted

<sup>\*</sup>Corresponding authors: f.nazari@uok.ac.ir, m.saba@uok.ac.ir | https://doi.org/10.1590/2447-536X.v30.e242775 | Editor: Gilmar Schafer, Universidade Federal do Rio Grande do Sul, Brazil Received: July 11, 2024 | Accepted: Oct 02, 2024 | Available online: Oct 25, 2024 | Licensed by CC BY 4.0 (https://creativecommons.org/licenses/by/4.0/)

to evaluate the biochar application and seed priming induced drought tolerance in cowpea. The seeds of cowpea were primed with CaCl2 (1% solution. It plays a crucial role in enhancing the physical, chemical, and biological characteristics of soil (Meng et al., 2021), including increased water holding capacity (Chang et al., 2021), carbon storage, which can raise soil pH, especially on acidic soils (Shetty and Prakash, 2020), also nutrient absorbing and cation exchange capacity (CEC) sorption characteristics (Abbruzzini et al., 2019), buffering capacity, improved soil structure, increased microbial biomass, nutrient provision, and improved soil fertility (Durukan et al., 2020). Furthermore, biochar has been reported to increase the yield of various plants. For instance, cabbage and lettuce treated with biochar exhibited increased final biomass, root biomass, plant height, and leaf count across all crop seasons compared to those without biochar treatment (Ghorbani et al., 2021).

African marigold (*Tagetes erecta* L.) is a warm season flower from the Asteraceae family. It is known for its vibrant colors and extended flowering period, which lasts from spring to early autumn. With its short transplant production time, ease of transplanting, and ability to thrive in various landscapes, African marigold is suitable for cultivation worldwide (Tavakoli Vala et al., 2023). Improving the growth, performance, and marketability of seasonal flowers, including African marigold, is a significant goal in plant production for landscapes. To achieve this, various methods are employed. This research aims to investigate the effects of zeolite and rice husk biochar on the vegetative, reproductive, and physiological characteristics of African marigold under greenhouse conditions.

# Materials and methods

# **Description of the Site and Experiment**

In order to evaluate the effect of zeolite and rice husk biochar on the morphological, biochemical, and photosynthetic characteristics of African marigold (*Tagetes erecta* L.) a greenhouse research was conducted in the Department of Horticultural Science, Faculty of Agriculture, University of Kurdistan. The experiment was performed as a completely randomized design with 5 treatments and 5 repetitions. Each replication had 2 experimental units (one plant in a pot). The treatments are as follows:

1. Soil mixture (field soil, sand, and manure, 1:1:1; v/v/v) as control.

2- Soil mixture amendment with 10% (w w1) rice husk biochar.

3-Soil mixture enriched with 25 g of zeolite per kg.

4- Soil mixture containing 10% (w w<sup>-1</sup>) rice husk biochar and 25 g of zeolite per kg.

#### Preparation of potting media and planting transplants

The biochar used in this study was prepared from rice husks, as described in our previous research (Safari et al., 2023). The natural clinoptilolite type of zeolite was obtained from the Afrazand Company's zeolite mine in Semnan, Iran. The physicochemical characteristics of pure rice husk biochar and pure clinoptilolite zeolite are mentioned in our previous research conducted by Safari et al. (2023) and Rahimi et al. (2021), respectively. Plastic pots with a volume of 3 L, a height of 18 cm, an inner diameter of 17 cm, and a surface area of 227 cm<sup>2</sup> were filled with soil mixtures according to the mentioned proportions. Before planting the transplants, the pots were watered three times with 100 cc of running water, with each watering occurring every three days. To prepare transplants, African marigold F1 seeds prepared from PanAmerican Seed Company were grown in 28-cell cultivation trays that were filled with a mixture of field soil, sand, and peat in a volume ratio of 1:1:1 on August 30, 2022. The culture trays were kept for 20 days in the controlled conditions of the greenhouse (average day and night temperature of 25 and 17 °C, relative humidity of 50%, and a day length of 14 hours). After the transplants reached the 3-leaf stage, one seedling was planted in each pot. Irrigation was carried out immediately, and thereafter, the pots were watered every 4 days. No fertilizer was used to provide nutrients to the plants during the experiment period.

# Measurements

### **Morphological traits**

After the flowers bloomed and fully opened, they were cut, and their length and diameter were measured using a digital caliper. The flowers were

then dried in an oven at a temperature of 70°C for 48 h. The dry weight of the flowers was determined using a digital scale (EK-15KL, A&D Co. Ltd., Tokyo, Japan) with an accuracy of 0.001. To measure the length of the flowering period, we counted the number of days from the appearance of the first flower until the end of flowering. After the flowering stage, we measured additional parameters such as plant height, stem diameter, shoot dry weight, number of lateral branches, number of flowers, and number of leaves.

#### Chlorophyll and carotenoid content

To measure the content of chlorophyll and carotenoid, utilized the method developed by Lichtenthaler and Buschmann (2001). First, 0.2 g of leaf samples were thoroughly ground in a mortar with 10 mL of 80% acetone and 0.01 g of magnesium oxide. Afterward, the mixture was centrifuged (Hettich MIKRO 200, Tuttlingen, Germany) at 3000 g for 10 min. The resulting supernatants were collected, and their absorbance values were evaluated at wavelengths of 663 nm, 645 nm, and 470 nm using a spectrophotometer (AAnalyst 100, PerkinElmer, Inc., Waltham, MA, USA). Finally, calculate the amounts of chlorophyll a, b, total, and carotenoids using the following relationships:

 $\begin{array}{l} {\rm Chl}_{\rm a} = (12.25 \times {\rm A663} - 2.79 \times {\rm A645}) \\ {\rm Chl}_{\rm b} = (21.21 \times {\rm A645} - 5.1 \times {\rm A663}) \\ {\rm Ch}_{\rm Total} = {\rm Chla} + {\rm Chl}_{\rm b} \\ {\rm Car} = (1000_{\rm Total} \times {\rm A470} - 1.8 \times {\rm Chl}_{\rm a} - 85.02 \times {\rm Chl}_{\rm b}) / 198 \end{array}$ 

#### Total soluble carbohydrate content

The method described by Irigoyen et al. (1992) was used to measure the total soluble carbohydrate content. First, 0.2 g of leaf tissue was weighed and ground with 2.5 mL of 95% ethanol in a mortar. The resulting supernatant extract was then transferred to a falcon tube, while the remaining leaf residue was ground again with 5 ml of 70% ethanol. The complete extract was centrifuged at 3500 g for 10 min. Next, 1 mL of the extract was transferred to a test tube and mixed with 3 ml of anthrone reagent. The test tubes were then placed in a hot water bath for 15 min to allow the color to develop. After cooling the tubes, their absorbance was measured at a wavelength of 625 nm using a spectrophotometer. Finally, the concentration of soluble carbohydrates in mg g<sup>-1</sup> of fresh leaf tissue was determined by referring to a standard curve.

#### Maximum quantum efficiency of photosystem II

To measure the maximum quantum efficiency of photosystem II, two healthy and fully developed leaves were chosen from each plant. A plant photosynthesis meter (EARS-miniPPM, Delft, Netherlands) was used to measure this trait and its components in both dark conditions (from 8 o'clock at night) and light conditions (12 o'clock in the afternoon). These measurements were then used to calculate the amount of photosynthesis using the following equations (Klughammer and Schreiber, 2008; Li et al., 2008):

### Statistical analysis

Statistical analysis was performed on all data using SAS version 9.4 software (SAS Institute Inc., Cary, NC, USA). A one-way analysis of variance (ANOVA) was conducted to determine if there were any statistically significant differences among the treatments. The LSD test was used to determine the differences between the treatments, with a significance level of 0.05.

# Results

# Length, fresh weight, turgor, and dry weight of roots

The culture medium type had a significant impact on the root length, root fresh weight, root turgor weight, and root dry weight of African marigold (Fig. 1A-D). The application of biochar, zeolite, and a combination of these amendments increased the root length, root fresh weight, and root turgor weight compared to the control. However, biochar and the combination of biochar + zeolite did not have a significant effect on root dry weight. Overall, the highest values for all four root attributes were obtained when using zeolite alone in the soil mixture (Fig. 1A-D).

#### Rudabeh Ghaderi et al.



Fig. 1. Root length (A), root fresh weight (B), root turgor weight (C) and root dry weight (D) of African marigold grown in a soil mixture (control) or soil mixture amended by biochar, zeolite, or their combination. The same letter(s) above bars are not significantly different according to LSD ( $p \le 0.05$ ). Error bars represent the standard error of means (n = 5).

#### Stem fresh, turgor and dry weight and plant height

Based on the results shown in Fig. 2, it was found that the use of biochar and zeolite, either alone or in combination in the soil mixture, significantly affected growth parameters such as stem fresh weight, stem turgor weight, stem dry weight, and plant height. Individually, the application of biochar and zeolite in the soil mixture had a more positive impact compared to their combined application. It should also be mentioned that zeolite was more effective than biochar, and the soil mixture containing zeolite had the highest amount of these mentioned traits (Fig. 2A-D).





Fig. 2. Stem fresh weight (A), stem turgor weight (B), stem dry weight (C), and plant height of African marigold grown in a soil mixture (control) or soil mixture amended by biochar, zeolite, or their combination. The same letter(s) above bars are not significantly different according to LSD ( $p \le 0.05$ ). Error bars represent the standard error of means (n = 5).

# Length, diameter, fresh and dry weight of flowers

Unlike the vegetative traits, the reproductive traits were less affected by the application of biochar and zeolite (Fig. 3A-D). Zeolite application in the soil significantly increased flower length, while there was no significant difference observed between control plants and those grown in other culture media (Fig. 3A). Flower diameter was not significantly affected by any of the four types of culture medium (Fig. 3B). The fresh and dry weight of the flower was either not significantly affected by the effect of biochar and zeolite, or it was reduced by the use of these two amendments (Fig. 3C-D).





Fig. 3. Flower length (A), flower diameter (B), flower fresh weight (C) and dry weight (D) of African marigold grown in a soil mixture (control) or soil mixture amended by biochar, zeolite, or their combination. The same letter(s) above bars are not significantly different according to LSD ( $p \le 0.05$ ). Error bars represent the standard error of means (n = 5).

В

D

#### **Photosynthetic pigments**

А

С

The use of biochar and zeolite did not significantly increase the amount of photosynthetic pigments in leaves. Although the highest amounts of chlorophyll a, b, total, and carotenoids were obtained with the use of biochar alone, the difference compared to the control was not significant. Even in contrast to this, the lowest value of the four mentioned traits was obtained when zeolite alone was applied. However, this reduction was not significant compared to the control (Fig. 4A-D).





**Fig. 4.** Chlorophyll a (A), chlorophyll b (B), total chlorophyll (C) and carotenoid (D) of African marigold grown in a soil mixture (control) or soil mixture amended by biochar, zeolite, or their combination. The same letter(s) above bars are not significantly different according to LSD ( $p \le 0.05$ ). Error bars represent the standard error of means (n = 5).

#### Total soluble carbohydrates

The use of zeolite alone, as well as its combination with biochar, resulted in a significant increase in the amount of leaf soluble carbohydrates. The highest amount of this attribute was achieved when they were used together. There was no significant difference in the amount of leaf soluble carbohydrates when biochar was used alone in the soil mixture (Fig. 5A).

#### Maximum quantum efficiency of photosystem (Fv/Fm)

The maximum quantum efficiency of photosystem II (Fv/Fm) is significantly affected by the application of biochar and zeolite alone in soil mixture. However, the combination of these two amendments had no significant effect on this trait (Fig. 5B).



Fig. 5. Total soluble carbohydrates (A) and maximum quantum efficiency of photosystem (Fv/Fm) (B) of African marigold grown in a soil mixture (control) or soil mixture amended by biochar, zeolite, or their combination. The same letter(s) above bars are not significantly different according to LSD ( $p \le 0.05$ ). Error bars represent the standard error of means (n = 5).

#### Discussion

Biochar, which is a charcoal-like substance made from organic matter, is becoming increasingly popular in agriculture because of its impressive ability to improve soil health and enhance plant growth (Das and Ghosh, 2020). One of the main advantages of biochar is its effect on soil density. It reduces the apparent specific gravity and creates a lighter, more porous soil structure. This increased porosity results in better aeration, allowing roots to breathe more easily and penetrate deeper into the soil. As a result, root growth thrives, leading to improved nutrient absorption and increased microbial activity (Garg et al., 2021).

Research on soybeans shows the clear benefits of biochar, both directly and indirectly. In this study, it was found that biochar led to a significant increase in the weight of both fresh and dry soybean roots and organs. This positive outcome can be attributed to two main reasons: biochar enhances the availability of nutrients in the soil and also improves its biological, physical, and chemical properties (Gavili et al., 2019). Another emerging star in the world of agriculture is zeolite, which serves a unique purpose in managing nutrients. By acting as a slow-release reservoir, zeolite particles gradually release essential elements into the surrounding environment, ensuring a consistent supply for plant growth (Nazari et al., 2014; Nazari, 2019). Additionally, zeolite's adhesive properties enable it to cling to root surfaces, facilitating the breakdown of nutrients and organic materials. This, in turn, promotes healthy root development (Akbari et al., 2021; Mondal et al., 2021). A study conducted on cucumber cultivation demonstrated the efficacy of zeolite, revealing a significant increase in root growth, available nutrient levels (N, P, and K), and CEC compared to control plants (Vassilina et al., 2023).

The porous structure of biochar enhances water retention in the soil, improves nutrient absorption, and enhances soil properties. This, in turn, promotes plant growth in both stressful and non-stressful conditions (Yildirim et al., 2021). In a recent study, it was found that rice husk biochar increased the fresh weight and turgor of African marigold stems compared to the control. Similar findings were reported in studies on corn (Aguirre et al., 2021), quinoa (Tourajzadeh et al., 2024), and tomato (Kul et al., 2021). On the other hand, zeolite helps reduce the leaching of essential nutrients such as P and K, providing a slow-release source throughout the growth period. This ultimately leads to higher production and transport of osmolytes within the plant (Akbari et al., 2021). In our study, the treatment of zeolite significantly increased the fresh weight, dry weight, and turgor of African marigold stems. This finding is consistent with previously observed results in sunflower (Baghbani-Arani et al., 2020) and petunia (Hamidpour et al., 2013). Additionally, the application of rice husk biochar significantly increased the height of the plants compared to the control group. This can be attributed to biochar's ability to enhance nutrient absorption, improve soil properties, increase waterholding capacity, and boost photosynthesis. These factors ultimately result in more frequent cell division and faster growth (Yildirim et al., 2021). Similar results have been found in research on lettuce (Park et al., 2021), a maize-Chinese cabbage rotation system (Zhao et al., 2022), and wheat (Khan et al., 2021), where the application of biochar led to taller plants and improved growth. In our study, zeolite also significantly increased plant height, likely due to its high water-holding capacity and enhanced CEC, which promote efficient nutrient storage and irrigation management. Furthermore, zeolite replaces ammonium and potassium with calcium on exchange sites, making these vital nutrients more readily available to plants. This, combined with increased P solubility and improved soil characteristics such as aeration, pH, and moisture retention, contributes to significant growth enhancements (Mondal et al., 2021). Studies on radish (AL-fatlawi et al., 2023), sweet pepper (Castronuovo et al., 2023), and African marigold (Nazari et al., 2007) confirm the positive impact of zeolite on plant height and growth.

Contrary to expectations, this study revealed that rice husk biochar significantly reduced the fresh and dry weight of African marigold flowers without affecting their length or diameter. This finding differs from previous research, where biochar enhanced reproductive and functional characteristics in various plants such as wheat (Khan et al., 2021), tomato (Kul et al., 2021), and corn (Aguirre et al., 2021). Similarly, zeolite had no substantial effect on flower length, diameter, and fresh weight but significantly reduced dry weight. This contrasts with results observed in petunia (Hamidpour et al., 2013) and Moldavian dragonhead medicinal plants (Karimzadeh Asl et al., 2018), where zeolite promoted growth. Several factors might explain these discrepancies, including variations in water quality, environmental conditions during the experiment, the specific African marigold variety used, and the inherent characteristics of the soil employed. All of these could influence plant responses to biochar and zeolite amendments. Further research investigating these factors in a controlled setting could provide valuable insights into the observed inconsistencies and improve our understanding of how biochar and zeolite interact with African marigold during the flower development stage.

The Fv/Fm ratio is a suitable index for assessing light inhibition and stress-induced disorders in photochemical centers. This ratio indicates the maximum quantum efficiency of photosystem II in converting absorbed light into chemical energy (Alemu, 2020). By studying chlorophyll fluorescence performance in plant leaves, changes in heat dissipation and photochemical efficiency can be observed (De Cannière et al., 2024). Measuring chlorophyll fluorescence is a valuable method for assessing damage to the photosynthetic apparatus and determining the physiological state of the plant. This method effectively reveals the health of the thylakoid membrane, the plant's tolerance to environmental stress, the degree of damage incurred, and the relative efficiency of electron transfer from photosystem II to photosystem I (Findurová et al., 2023). Any stress factor that affects the plant's photosynthetic system can damage components of the

electron transport chain and photosystem II, disrupting electron transfer and causing the loss of light energy as heat and fluorescence. The application of biochar can enhance photosynthetic efficiency by reducing the optical inhibition of photosystem II, ultimately increasing the rate of photosynthesis (Zhang et al., 2020). Research has shown that biochar improves the photosynthetic activity of Phragmites by enhancing soil characteristics and facilitating nutrient uptake (Abideen et al., 2020). Similarly, another study found that biochar-treated cucumber (Nadeem et al., 2017) and okra plants (Batool et al., 2015) exhibited increased photosynthesis and photosynthetic components. However, in the present study, no significant changes were observed in the rate of photosynthesis or its components following the application of biochar and zeolite. This lack of impact could be attributed to the use of an unsuitable dosage of biochar or zeolite. Additionally, the treatments did not have any effect on the amount of chlorophyll and photosynthetic pigments, which may have contributed to the absence of observed effects on photosynthetic components.

Adding biochar to the soil improves its properties in several ways. It enhances nutrient and water access, increases nutrient storage, boosts CEC, reduces compaction, and fosters beneficial microorganisms (Ding et al., 2010). These factors collectively contribute to better plant growth, as we observed in our study. Furthermore, the total soluble carbohydrate content in these plants also increased, which aligns with previous findings in beans (Farhangi-Abriz and Torabian, 2017) and perennial ryegrass (Rahimi et al., 2021; Safari et al., 2023). On the other hand, zeolite, with its crystalline structure, acts as a reservoir for water and nutrients, gradually releasing them to plants. Its high CEC also makes it a valuable soil amendment, as it modifies the physical and chemical properties to promote optimal plant growth (Akbari et al., 2021). Similar to studies on cucumber (Vassilina et al., 2023) and radish (AL-fatlawi et al., 2023), our research found that zeolite application elevated the total soluble carbohydrate level.

The positive effects of biochar on soil physicochemical properties have been extensively documented. Its gradual release of elements helps reduce carbon, P, and N loss, resulting in improved soil fertility, increased chlorophyll content, and enhanced plant growth and physiological characteristics (Mukherjee and Zimmerman, 2013; Safari et al., 2023). It is worth noting that biochar also enhances magnesium absorption, which is a key component of photosynthetic pigments (Dong et al., 2019). Previous studies have shown that biochar application leads to increased chlorophyll content in crops such as corn (Adejumo et al., 2016), peanut (Agegnehu et al., 2015), and perennial ryegrass (Safari et al., 2023). Similarly, zeolite has been found to enhance chlorophyll content in tuberose plants by improving nutrient uptake, water access, and soil properties (Bahadoran et al., 2012; Nazari, 2019). However, in this study, the combined application of biochar and zeolite did not significantly affect plant photosynthetic pigments. There could be several explanations for this observation. For example, the high application rates of biochar and zeolite may have resulted in salinity and toxicity from the released elements, which could have hindered chlorophyll production. Additionally, the Allelochemical properties of biochar's phenolic compounds may have had a negative impact on pigment production.

# Conclusions

This study aimed to investigate the effects of biochar, zeolite, and soil on the growth characteristics of African marigolds. The results revealed interesting findings regarding the impact of these amendments on various parameters. Soil cultivation alone resulted in the highest dry flower weight, indicating its importance in supporting plant growth. Remarkably, the addition of biochar significantly enhanced the maximum efficiency of Photosystem II, which converts light energy into chemical energy. This suggests that biochar has the potential to improve photosynthetic performance in plants. On the other hand, zeolite application had a significant positive effect on root and stem length, weight, turgidity, flower length, plant height, and photosystem II efficiency. However, it unexpectedly led to a decrease in chlorophyll content. These contrasting effects indicate the complexity of plant responses to zeolite and highlight the need for further investigation into its mechanisms of action. Furthermore, the combined treatment of biochar and zeolite with soil increased total soluble carbohydrate content. Although this combined treatment did not significantly enhance the growth rate or overall plant condition, it suggests a potential synergy between biochar and zeolite in promoting carbohydrate accumulation. Overall, zeolite

alone demonstrated a higher effectiveness compared to other treatments, as it positively influenced multiple growth parameters. However, further research is warranted to explore different application levels of biochar and zeolite, as well as their potential interactions, to optimize their effects on plant growth and development. Understanding the underlying mechanisms and fine-tuning the application strategies will contribute to the advancement of sustainable agricultural practices.

### Author contribution

**FN:** conceptualization, designed the experiment, supervision, methodology, data curation, validation, software, editing, provided scientific advice and guidance. **MKS:** conceptualization, designed the experiment, supervision, methodology, data curation, validation, software, editing, provided scientific advice and guidance **RG:** performed the experiment, writing – original draft **NN:** performed the experiment, writing – original draft. **HN:** methodology, data curation, validation, software. **ALL AUTHORS:** read and approved the final manuscript.

#### Acknowledgments

The authors would like to express their gratitude to the University of Kurdistan for their provision of facilities and financial support for this research.

## **Conflict of Interest**

The authors declare there are no competing interests.

# Data Availability Statement

Data will be made available on request.

#### References

ABBRUZZINI, T.F.; DAVIES, C.A.; TOLEDO, F.H.; CERRI, C.E.P. Dynamic biochar effects on nitrogen use efficiency, crop yield, and soil nitrous oxide emissions during a tropical wheat-growing season. Journal of Environmental Management, v.252, p.109638, 2019. https://doi.org/10.1016/j.jenvman.2019.109638

ABIDEEN, Z.; KOYRO, H.W.; HUCHZERMEYER, B.; ANSARI, R.; ZULFIQAR, F.; GUL, B.J.P.B. Ameliorating effects of biochar on photosynthetic efficiency and antioxidant defense of *Phragmites karka* under drought stress. **Plant Biology**, v.22, n.2, p.259-266, 2020. https://doi.org/10.1111/plb.13054

ADDAS, A. Influence of urban green spaces on quality of life and health with smart city design. Land, v.12, n.5, p.960, 2023. https://doi.org/10.3390/land12050960

ADEJUMO, S.A.; OWOLABI, M.O.; ODESOLA, I.F. Agro-physiologica effects of compost and biochar produced at different temperatures on growth, photosynthetic pigment and micronutrients uptake of maize crop. **African Journal of Agricultural Research**, v.11, n.8, p.661-673, 2016. https://doi.org/10.5897/ajar2015.9895

AGEGNEHU, G.; BIRD, M.I.; NELSON, P.N.; BASS, A.M. The ameliorating effects of biochar and compost on soil quality and plant growth on a Ferralsol. **Soil Research**, v.53, n.1, p.1-12, 2015. https://doi.org/10.1071/sr14118

AGUIRRE, J.L.; MARTIN, M.T.; GONZALEZ, S.; PEINADO, M. Effects and economic sustainability of biochar application on corn production in a mediterranean climate. **Molecules**, v.26, n.11, p.3313, 2021. https://doi. org/10.3390/ molecules 26113313

AKBARI, H.; MODARRES-SASANVY, S.A.M.; HEIDARZADEH, A. Fertilizer systems deployment and zeolite application on nutrients status and nitrogen use efficiency. **Journal of Plant Nutrition**, v.44, n.2, p.196-212, 2021. https://doi.org/10.1080/01904167.2020.1806299

ALEMU, S.T. Photosynthesis limiting stresses under climate change scenarios and role of chlorophyll fluorescence: A review article. **Cogent Food and Agriculture**, v.6, n.1, p.1785136, 2020. https://doi.org/10.108 0/23311932.2020.1785136

AL-FATLAWI, H.H.; FLEIH, S.A.; ABDULLAH, K.M. Qualitative traits of two types of radish (*Raphanus sativus* L.) and their content of medically active substance sulforaphane in response to treatments with zeolite and nano-potassium. Journal of Kerbala for Agricultural Sciences, v.10, n.2, p.81-91, 2023. https://doi.org/10.59658/jkas.v10i2.1190

AMIRAHMADI, E.; GORBANI, M.; MOUDRY, J. Effects of zeolite on aggregation, nutrient availability, and growth characteristics of corn (*Zea mays* L.) in cadmium-contaminated soils. **Water, Air, and Soil Pollution**, v.233, n.11, p.436, 2022. https://doi.org/10.1007/S11270-022-05910-4/metrics

AMIRAHMADI, E.; MOHAMMAD HOJJATI, S.; KAMMANN, C.; GHORBANI, M.; BIPARVA, P. The potential effectiveness of biochar application to reduce soil Cd bioavailability and encourage oak seedling growth. **Applied Sciences**, v.10, n.10, p.3410, 2020. https://doi.org/10.3390/app10103410

BAGHBANI-ARANI, A.; JAMI, M.G.; NAMDARI, A.; KARAMI BORZ-ABAD, R. Influence of irrigation regimes, zeolite, inorganic and organic manures on water use efficiency, soil fertility and yield of sunflower in a sandy soil. **Communications in Soil Science and Plant Analysis**, v.51, n.6, p.711-725, 2020. https://doi.org/10.1080/00103624.2020.1729791

BAHADORAN, M.; SALEHI, H.; ESHGHI, S. Growth and flowering of tuberose as affected by adding natural zeolite to the culture medium. **Journal of Plant Nutrition**, v.35, n.10, p.1491-1496, 2012. https://doi.or g/10.1080/01904167.2012.689909

BASHIR, S.; SALAM, A.; REHMAN, M.; KHAN, S.; GULSHAN, A.B.; IQBAL, J.; HU, H. Effective role of biochar, zeolite and steel slag on leaching behavior of Cd and its fractionations in soil column study. **Bulletin of Environmental Contamination and Toxicology**, v.102, p.567-572, 2019. https://doi.org/10.1007/S00128-019-02573-6/metrics

BATOOL, A.; TAJ, S.; RASHID, A.; KHALID, A.; QADEER, S.; SALEEM, A.R.; GHUFRAN, M.A. Potential of soil amendments (Biochar and Gypsum) in increasing water use efficiency of *Abelmoschus esculentus* L. Moench. Frontiers in Plant Science, v.6, p.138796, 2015. https://doi.org/10.3389/FPLS.2015.00733/bibtex

BOROS-LAJSZNER, E.; WYSZKOWSKA, J.; KUCHARSKI, J. Use of zeolite to neutralise nickel in a soil environment. Environmental Monitoring and Assessment, v.190, n.54, p.1-13, 2018. https://doi.org/10.1007/S10661-017-6427-Z/TABLES/7

CASTRONUOVO, D.; SATRIANI, A.; RIVELLI, A.R.; COMEGNA, A.; BELVISO, C.; COPPOLA, A.; LOVELLI, S. Effects of zeolite and deficit irrigation on sweet pepper growth. **Horticulture**, v.9, n.11, p.1230, 2023. https://doi.org/10.3390/ horticulturae 9111230

CHANG, Y.; ROSSI, L.; ZOTARELLI, L.; GAO, B.; SHAHID, M.A.; SARKHOSH, A. Biochar improves soil physical characteristics and strengthens root architecture in Muscadine grape (*Vitis rotundifolia* L.). Chemical and Biological Technologies in Agriculture, v.8, n.7, p.1-11, 2021. https://doi.org/10.1186/S40538-020-00204-5/ figures /3

CROUSE, D.L.; PIMAULT, L.; CHRISTIDIS, T.; LAVIGME, E.; THOMSON, E.M.; VILLENEUVE, P. J. Residential greenness and indicators of stress and mental well-being in a Canadian national-level survey. **Environmental Research**, v.192, p.110267, 2021. https://doi.org/10.1016/j.envres.2020.110267

DAS, S.K.; GHOSH, G.K. Soil health management through low cost biochar technology. **Biochar Applications in Agriculture and Environment Management,** p.193-206, 2020. https://doi.org/10.1007/978-3-030-40997-5\_9

DE CANNIÈRE, S.; BAUR, M.J.; CHAPARRO, D.; JAGDHUBER, T.; JONARD, F. Water availability and atmospheric dryness controls on spaceborne sun-induced chlorophyll fluorescence yield. **Remote Sensing of Environment**, v.301, n.113922, 2024. https://doi.org/10.1016/j.rse.2023.113922

DING, Y.; LIU, Y.X.; WU, W.X.; SHI, D.Z.; YANG, M.; ZHONG, Z.K. Evaluation of biochar effects on nitrogen retention and leaching in multilayered soil columns. **Water, Air, and Soil Pollution**, v.213, p.47-55, 2010. https://doi.org/10.1007/S11270-010-0366-4/metrics

DONG, L.; RAVELOMBOLA, W.; WENG, Y.; QIN, J.; BHATTARAI, G.; ZIA, B.; SHI, A. Seedling salt tolerance for above ground-related traits in cowpea (*Vigna unguiculata* (L.) Walp). **Euphytica**, v.215, n.53 p.1-22, 2019. https://doi.org/10.1007/S10681-019-2379-4/ metrics

DURUKAN, H.; DEMIRBAS, A.; TURKEKUL, I. Effects of biochar rates on yield and nutrient uptake of sugar beet plants grown under drought stress. **Communications in Soil Science and Plant Analysis**, v.51, n.21, p.2735-2745, 2020. https://doi.org/10.1080/00103624.2020.1849257

FARHANGI-ABRIZ, S.; TORABIAN, S. Antioxidant enzyme and osmotic adjustment changes in bean seedlings as affected by biochar under salt stress. **Ecotoxicology and Environmental Safety**, v.137, p.64-70, 2017. https://doi.org/10.1016/j.ecoeny.2016.11.029

FAROOQ, M.; ROMDHANE, L.; REHMAN, A.; AL-ALAWI, A.K.; AL-BUSAIDI, W.M.; ASAD, S.A.; LEE, D.J. Integration of seed priming and biochar application improves drought tolerance in cowpea. Journal of Plant Growth Regulation, v.40, p.1972-1980, 2021. https://doi.org/10.1007/S00344-020-10245-7/ metrics

FINDUROVÁ, H.; VESELA, B.; PANZAROVA, K.; PYTELA, J.; TRTILEK, M.; KLEM, K. Phenotyping drought tolerance and yield performance of barley using a combination of imaging methods. **Environmental and Experimental Botany**, v.209, n.105314, 2023. https://doi.org/10.1016/j.envexpbot.2023.105314

GARG, A.; HUANG, H.; CAI, W.; REDDY, N.G.; CHEN, P.; HAN, Y.; ZHU, H.H. Influence of soil density on gas permeability and water retention in soils amended with in-house produced biochar. Journal of Rock Mechanics and Geotechnical Engineering, v.13, n.3, p.593-602, 2021. https://doi.org/10.1016/j.jrmge.2021.10.007

GAVILI, E.; MOOSAVI, A.A.; HAGHIGHI, A.A.K. Does biochar mitigate the adverse effects of drought on the agronomic traits and yield components of soybean? **Industrial Crops and Products**, v.128, p.445-454, 2019. https://doi.org/10.1016/j.indcrop.2018.11.047

GHORBANI, M.; AMIRAHMADI, E.; ZAMANIAN, K. In-situ biochar production associated with paddies: Direct involvement of farmers in greenhouse gases reduction policies besides increasing nutrients availability and rice production. Land Degradation and Development, v.32, n.14, p.3893-3904, 2021. https://doi.org/10.1002/ldr.4006

HAMIDPOUR, M.; FATHI, S.; ROOSTA, H.R. Effects of zeolite and vermicompost on growth characteristics and concentration of some nutrients in *Petunia hybrida*. Journal of Science and Technology of Greenhouse Culture, v.4, n.13, P.95-103, 2013. https://www. cabidigitallibrary.org/doi/full/10.5555/20133146080

HASSAN, W.; SABA, T.; JABBI, F.; WANG, B.; CAI, A.; WU, J. Improved and sustainable agroecosystem, food security and environmental resilience through zero tillage with emphasis on soils of temperate and subtropical climate regions: A review. **International Soil and Water Conservation Research**, v.10, n.3, p.530-545, 2022. https://doi.org/10.1016/j.iswcr.2022.01.005

HUNTER, R.F.; CLELAND, C.; CLEARY, A.; DROOMERS, M.; WhEELER, B.W.; SINNETT, D.; NIEUWENHUIJSEN, M.J.; BRAUBACH, M. Environmental, health, wellbeing, social and equity effects of urban green space interventions: A meta-narrative evidence synthesis. **Environment International**, v.130, p.104923, 2019. https:// doi.org/10.1016/j.envint.2019.104923 IRIGOYEN, J.J.; EINERICH, D.W.; SANCHEZ DIAZ, M. Water stress induced changes in concentrations of proline and total soluble sugars in nodulated alfalfa (*Medicago sativa*) plants. **Physiologia Plantarum**, v.84, n.1, p.55-60, 1992. https://doi.org/10.1111/j.1399-3054.1992.tb08764.x

KARIMZADEH, A.S.L., K.; GHORBANPOUR, M.; MAREFATZADEH KHAMENEH, M.; HATAMI, M. Influence of drought stress, biofertilizers and zeolite on morphological traits and essential oil constituents in *Dracocephalum moldavica* L. Journal of Medicinal Plants, v.17, n.67, p.91-112, 2018. http://jmp.ir/article-1-2267-en.html

KHAN, F.; IQBAL, M.; MOHIBULLAH, M.; AAMIR, S.S.; JATOI, S.A. Behavior of annual flower production against varied nutrition management. **SABRAO Journal of Breeding and Genetics**, v.55, n.5, p.1604-1615, 2023. http://doi.org/10.54910/sabrao2023.55.5.14

KHAN, Z.; RAHMAN, M.H.U.; HAIDER, G.; AMIR, R.; IKRAM, R.M.; AHMAD, S.; DANISH, S. Chemical and biological enhancement effects of biochar on wheat growth and yield under arid field conditions. **Sustainability**, v.13, n.11, p.5890, 2021. https://doi.org/10.3390/su13115890/s1

KLUGHAMMER, C.; SCHREIBER, U. Complementary PS II quantum yields calculated from simple fluorescence parameters measured by PAM fluorometry and the Saturation Pulse method. **PAM Application Notes**, v.1, n.2, p. 201-247, 2008.

KUL, R.; ARJUMEND, T.; EKINCI, M.; YILDIRIM, E.; TURAN, M.; ARGIN, S. Biochar as an organic soil conditioner for mitigating salinity stress in tomato. **Soil Science and Plant Nutrition**, v.67, n.6, p.693-706, 2021. https://doi.org/10.1080/00380768.2021.1998924

LI, S.; GU, S.; LIU, W.; HAN, H.; ZHANG, Q. Water quality in relation to land use and land cover in the upper Han River Basin, China. **Catena**, v.75, n.2, p.216-222, 2008. https://doi.org/10.1016/j.catena.2008.06.005

LICHTENTHALER, H.K.; BUSHMANN, C. Extraction of phtosynthetic tissues: chlorophylls and carotenoids. Current Protocols in Food Analytical Chemistry, v.1, n.1, p.F4-2, 2001.

MENG, Q.; ZHAO, S.; GENG, R.; ZHAO, Y.; WANG, Y.; YU, F.; MA, X. Does biochar application enhance soil salinization risk in black soil of northeast China (a laboratory incubation experiment)? **Archives of Agronomy and Soil Science**, v.67, n.11, p.1566-1577, 2021. https://doi.org/10.1080/03650340.2020.1800642

MONDAL, M.; BISWAS, B.; GARAI, S.; SARKAR, S.; BANERJEE, H.; BRAHMACHARI, K.; HOSSAIN, A. Zeolites enhance soil health, crop productivity and environmental safety. **Agronomy**, v.11, n.3, p.448, 2021. https://doi.org/10.3390/agronomy11030448

MUKHERJEE, A.; ZIMMERAMN, A.R. Organic carbon and nutrient release from a range of laboratory-produced biochars and biochar-soil mixtures. **Geoderma**, v.193, p.122-130, 2013. https://doi.org/10.1016/J. geoderma.2012.10.002

NADEEM, S.M.; IMRAN, M.; NAVEED, M.; KHAN, M.Y.; AHMAD, M.; ZAHIR, Z.A.; CROWLEY, D.E. Synergistic use of biochar, compost and plant growth promoting rhizobacteria for enhancing cucumber growth under water deficit conditions. Journal of the Science of Food and Agriculture, v.97, n.15, p.5139-5145, 2017. https://doi.org/10.1002/jsfa.8393

NAZARI, F. Vegetative and reproductive responses of tuberose (*Polianthes tuberosa* L. cv. Dezful) to application of different amounts of natural clinoptilolite zeolite in potting medium. Journal of Soil and Plant Interactions-Isfahan University of Technology, v.9, n.4, p.81-94, 2019.

NAZARI, F.; FARAHMAND, H.; GHASEMI, G.M. The effects of different amounts of natural zeolite on vegetative and reproductive characteristics of *Narcissus tazetta* L. cv. Shahla. Journal of Plant **Production**, v.37, n.2, p.39-48, 2014. https://sid.ir/paper/165166/en

NAZARI, F.; KHOSH-KHUI, M.; SALEHI, H.; ESHGHI, S. Effect of natural zeolite on vegetative and physiological characteristics of African marigold (*Tagetes erecta* L."Queen"). Horticulture, Environment, and Biotechnology, v.48, p.241-245, 2007.

PARK, J.H.; KANG, S.W.; YUN, J.J.; LEE, S.G.; KIM, S.H.; BEAK, J.S.; CHO, J.S. Effects of co-application of biochars and composts on lettuce growth. **Korean Journal of Soil Science and Fertilizer**, v.54, n.2, p.151-160, 2021. https://doi.org/10.7745/kjssf.2021.54.2.151

RAHIMI, E.; NAZARI, F.; JAVADI, T.; SAMADI, S.; DA SILVA, J.A.T. Potassium-enriched clinoptilolite zeolite mitigates the adverse impacts of salinity stress in perennial ryegrass (*Lolium perenne* L.) by increasing silicon absorption and improving the K/Na ratio. Journal of Environmental Management, v.285, p.112142, 2021. https://doi.org/10.1016/j.jenvman.2021.112142

REID, C.E.; RIEVES, E.S.; CARLSON, K. Perceptions of green space usage, abundance, and quality of green space were associated with better mental health during the COVID-19 pandemic among residents of Denver. **PloS One**, v.17, n.3, p.e0263779, 2022. https://doi.org/10.1371/journal.pone.0263779

SAFARI, S.; NAZARI, F.; VAFAEE, Y.; TEIXEIRA DA SILVA, J.A. Impact of rice husk biochar on drought stress tolerance in perennial ryegrass (*Lolium perenne* L.). **Journal of Plant Growth Regulation**, v.42, n.2, p.810-826, 2023. https://doi.org/10.1007/s00344-022-10588-3

SHETTY, R.; PRAKASH, N.B. Effect of different biochars on acid soil and growth parameters of rice plants under aluminium toxicity. **Scientific Reports**, v.10, n.12249, p.1, 2020.

TAVAKOLI VALA, M.; NAZARI, F.; BABAEI, S. The ameliorative effect of rice husk biochar on morpho-physiological and biochemical characteristics of African marigold (*Tagetes erecta* L.) under drought stress. **Flower and Ornamental Plants**, v.8, n.1, p.21-46, 2023. http:// flowerjournal.ir/article-1-269-en.html

TOURAJZADEH, O.; PIRI, H.; NASERIN, A.; MAHDI CAHRI, M. Effect of nano biochar addition and deficit irrigation on growth, physiology and water productivity of quinoa plants under salinity conditions. **Environmental and Experimental Botany**, v.217, p.105564, 2024. https://doi.org/10.1016/j.envexpbot.2023.105564

VASSILINA, T.; NASIYEV, B.; RVAIDAROVA, G.; SHIBIKEYEVA, A.; SEITKALI, N.; SALYKOVA, A.; YERTAYEVA, Z. The effects of clinoptilolite type of zeolite and synthesised zeolite-enriched fertilizer on yield parameters of Cucumber (*Cucumis sativus*) plant and some chemical properties in dark chestnut soil. **Eurasian Journal of Soil Science**, v.12, n.3, p.277-281, 2023. https://doi.org/10.18393/ejss.1284506

YILDIRIM, E.; EKINCI, M.; TURAN, M. Impact of biochar in mitigating the negative effect of drought stress on cabbage seedlings. **Journal of Soil Science and Plant Nutrition**, v.21, n.3, p.2297-2309, 2021. https://doi. org/10.1007/S42729-021-00522-z/metrics

ZHANG, P.; YANG, F.; ZHANG, H.; LIU, L.; LIU, X.; CHEN, J.; LI, C. Beneficial effects of biochar-based organic fertilizer on nitrogen assimilation, antioxidant capacities, and photosynthesis of sugar beet (*Beta vulgaris* L.) under saline-alkaline stress. **Agronomy**, v.10, n.10, p.1562, 2020.

ZHAO, H.; XIE, T.; XIAO, H.; GAO, M. Biochar-based fertilizer improved crop yields and N utilization efficiency in a maize–Chinese cabbage rotation system. **Agriculture**, v.12, n.7, p.1030, 2022. https://doi.org/10.3390/agriculture12071030