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Influence of biochar and chitosan on productivity of tomato under water-stress conditions

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Key words: Drought, Field capacity, Biochemical parameters, Chlorophyll content, Yield

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Abstract

Adding biochar to the soil as an amendment might help it retain more water, which could increase crop yields. The experiment was conducted at Sher-e-Bangla Agricultural University, Dhaka, Bangladesh during January to April 2022 to determine the effects of chitosan and biochar on tomato plants under water stress in terms of productivity. In this experiment, three water regimes (i) at 80% of field capacity (FC), ii) at 60% of FC, and iii) at 40% of FC were used as treatments. Additionally, three drought mitigating agents were used: i) chitosan (200 μL L^{-1} ha^{-1}), ii) rice husk biochar (20 t/ha), and iii) biochar + chitosan. Biochar was used to transplant 25-day-old, uniform, healthy seedlings into plastic pots. Drought was imposed ten days after transplantation, lasting till flowering. Chitosan was applied using a hand sprayer ten days after implantation. The findings revealed that water stress significantly reduced tomato morphological, physiological, and biochemical properties while increasing pH. However, the effects of water stress on tomato plants were greatly mitigated by the use of biochar and chitosan. The number of leaves, chlorophyll content, days needed from transplanting to the first flowering, fruits per plant, weight of each individual fruit, yield per plant, pH, and titratable acidity content all significantly improved with the application of biochar and chitosan. Consequently, in terms of growth, yield, and biochemical parameters, the combination effect of chitosan and biochar appeared more effective.

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Introduction

Globally, tomatoes *(Lycopersicum esculentum* L.) are mostly grown in home and commercial gardens and are one of the most important and widely used agricultural vegetable crops. Plant growth and productivity are influenced by a wide range of biotic and abiotic stress factors. Various plants under drought stress showed significant reductions in growth parameters, including leaf count, leaf area, and stem length. Fresh-market tomatoes are a popular and adaptable fruit vegetable that contribute significantly to human nutrition worldwide due to their high sugar and acid content. It consists of a significant amount of carbohydrates; acids are key factors to tomato flavor (Gharezi *et al.,* 2012). In addition to affecting fruit quality, moisture stress reduces crop productivity. Under water stress, there have been documented changes in the physicochemical properties of tomatoes, including total soluble solids (TSS), titratable acidity (TA), and firmness. TSS increased in tomatoes under water stress (Shahein *et al.,* 2012).

Drought is a significant environmental threat that affects plant metabolism at numerous levels. Approximately one-third of agricultural land in worldwide lacks adequate water supplies (Zhang *et al.,* 2013). Water scarcity inhibits plant efficiency, negatively impacting photosynthesis, growth, and fruit output. Water-stressed plants affect cellular pathways and overall efficiency. Water-deficit may improve tomato fruit quality by increasing levels of total soluble solids (sugars, amino acids, and organic acids), which accumulate in the fruit (Yin *et al.,* 2010).

The benefits of biochar on soil have been associated with improved nutrient availability, cation exchange capacity (CEC), and soil water-holding capacity (Liu *et al.,* 2013; Olmo *et al.,* 2014). Biochar's porous structure improves water retention in sandy soils, leading to more efficient water usage in agriculture. Adding biochar to soils is currently found to increase crop growth and yield (Petruccelli *et al.,* 2015).

One of the primary characteristics of chitosan, which is a naturally occurring biodegradable substance derived from chitin and found in the shells of crustaceans like shrimp and crabs, is that it is poly-cationic (Bautista-Baños *et al.,* 2006). Chitosan oligomers permeate the majority of cell areas and subsequently affect the following: calcium, MAP kinase, chromatin, deoxyribonucleic acid (DNA), oxidative burst, reactive oxygen species (ROS), pathogenesis-related (PR) genes/proteins, and phytoalexins (Hadwiger, 2013). Chitosan improves stress tolerance by enhancing hypersensitive reactions, lignification, lipid peroxidation, and pathogen defence when used directly to plant tissue. Chitosan might reduce the impact of water stress on production by increasing stomatal conductance and reducing transpiration rate through foliar spray (Ahmed *et al.,* 2020). As a result, the present investigation was conducted to study the effect of biochar and chitosan on morphological, physiological and yield attributes of tomato under water stress.

Materials and methods

Plant materials and growing conditions

Cultivar BARI tomato 8 was used in the experiment, which was conducted in a shed house at Sher-e-Bangla Agricultural University, Dhaka 1207, Bangladesh, under natural lighting. The tomato seeds were received from the Bangladesh Agricultural Research Institute (BARI) in Gazipur, Bangladesh. Seeds were sown in PVC tanks (1.2×0.6×0.6 m) using a soil combination and slow-release fertilizers. At 25 days old, seedlings were transplanted to maintained pots (3 seedlings per pot), which were filled with soil and recommended fertilizer doses.

Treatments and sample collection

Three different water regimes were applied to the plants as part of the treatments. From 30-35 day-old seedlings to maturity, three levels of field capacity were maintained: i) control (80% of field capacity), ii) 60% of field capacity (FC), and iii) 40% of field capacity (FC). Rice husk biochar (20 t/ha), chitosan (200 µL L^{-1}), and chitosan + biochar were applied to the plants. 40% moisture, 40% moisture + chitosan, 40% moisture + biochar, 40% moisture + chitosan + biochar, 60% moisture, 60% moisture + chitosan, 60% moisture + biochar, 60% moisture + chitosan + biochar, 40% moisture, 40% moisture + chitosan, 40% moisture + biochar, 80% moisture + chitosan + biochar are the treatments that will be used in the experiment. There were three replications for each treatment. Many morphological, physiological, and biochemical parameters will be assessed throughout the blossoming period.

Number of leaves per plant

Each plant in the treatment had all of its leaves counted three times, and a mean value was determined. The number of trimmed leaves was also counted.

SPAD chlorophyll meter reading

A SPAD-502 chlorophyll meter (Minolta, Tokyo, Japan) was used to determine the amount of chlorophyll present in the first fully opened leaves. The midpoint of the leaf lamina was measured on both the treated and control plants. During the flowering stages, five randomly selected plants from each treatment and replication were assessed.

Measurements of flower, yield and yield traits

For each treatment and replication, five randomly selected plants were used to record observations on the number of days until tomato flowering. The average of the harvests from all five plants in each treatment and replication was used to calculate the yields per plant (g). On each harvest day, an electronic top pan balance was used to weigh the fruits (g) from each chosen plant. Counting the gathered fruit allowed us to determine the number of fruits/plants.

pH determination and titratable acidity (TA %)

A digital pH meter (HI 2211; Romania) was used to measure the pH of tomato fruit juices that had been subjected to different treatments. Using a mortar and pestle, a 5 g sample was ground to determine the titratable acidity. 100ml was the final volume after filtering and adding distilled water. The stock solution was then poured into a conical flask along with 2 drops of phenolphthalein in an amount of 10 ml. To titrate the solution, 0.1N NaOH was utilized.

Data analysis

The analysis of the data was done with SPSS 20.0. When $P < 0.05$, the value was deemed statistically significant. The mean ± SE of the replicates was used to present all the results. Microsoft Excel was used to make the graphs.

Results and discussion

Number of leaves plant^{-1}

Plants with a moisture level of 80 % had the highest number leaves (22.67), while plants with a moisture level of 40% have the minimum number of leaves (13.67) (Table 1). Plants use biochar and chitosan to reduce drought stress and maintain leaf number under various water stress circumstances. The stressed tomato plants showed a considerable increase in the number of leaves per plant after applying chitosan (Moolphuerk *et al.*, 2022). Addition of biochar to soil improved soil quality and retention of nutrients resulting in increased plant growth (Bonanomi *et al.*, 2017). As a result, plant morphological and physiological attributes were significantly improved, which have a positive effect on the increase of the number of leaves per plant. Comparing biochar to the control, there was a considerable increase in phenol oxidase, acid activity, and fluorescein diacetate hydrolases in the soil.

SPAD reading

Regarding varying moisture levels, plants maintained at 80% moisture level + chitosan exhibited the highest chlorophyll content (56.61), whereas plants maintained at 40% moisture level treated without any chemical showed the lowest chlorophyll content (46.86). In contrast to drought-stressed plants alone, plants that get biochar and chitosan treatment reduce drought stress and have the maximum chlorophyll content under various water-stressed circumstances (Table 1). Plants treated with chitosan + biochar had the maximum chlorophyll content at both 60% and 40% moisture level, indicating a reduction in stress circumstances compared to those of plants that did not use chitosan and biochar. Chitosan applied topically lessened the effects of water stress on photosynthetic rate, which can boost endogenous cytokinins, which are essential for increasing the amount of chlorophyll in leaves (Hassnain *et al.,* 2020). Plants under drought stress can lose a large amount of their chlorophyll content; the more stress there is, the more chlorophyll is lost. Drought stress lowers crop water content, increases stomatal resistance, and lowers photosynthetic rate (Xu *et al.,* 2020).

Days required from transplanting to 1st flowering

Plants with 80% moisture required more days to flower (59.33) however, plants with 40% moisture required fewer days to flower (51.00). Plants use biochar and chitosan to mitigate drought stress and lengthen flowering time in various kinds of water stress conditions. Plants treated with chitosan + biochar took more days to flower (58.00) and (55.00) when compared to drought-stressed plants alone (Table 1). Foliar spraying with oligo chitosan possessed an advantageous impact on the number of flower clusters and days required from transplanting first flowering of tomato plants at different days after planting (Sultana *et al.,* (2017). The physiological processes of plants were altered by drought stress, and biochar supports physiological activity, which leads to plant flowering, improved plant development, and an influence on the number of days until first flowering (Mannan *et al.,* 2020).

Table 1. Effect of biochar and chitosan under various water regimes on morphological, physiological, yield and quality attributes of tomato

Treatments	Number of leaves/plant	SPAD value	Days to flowering	Number of fruits/plant
W_1B_0	19.00 bc	53.66 b	58.33 ab	23.67 b
W_1B_1	21.33 ab	56.61 a	57.00 bc	24.67 ab
W_1B_2	20.02 bc	51.03 bc	57.33 bc	23.67 b
W_1B_3	22.67 a	51.93 bc	59.33 a	25.33 a
W_2B_0	19.04 bc	50.63 c	56.67 bcd	18.67 e
W_2B_1	20.00 bc	50.96 bc	55.67 cd	21.33 cd
W_2B_2	17.67 c	50.96 bc	57.33 bc	20.33 d
W_2B_3	21.33 ab	51.16 bc	58.00 ab	21.66 c
W_3B_0	13.67 d	46.86 e	51.00 f	6.33 h
W_3B_1	18.06 c	49.26 c	53.00 e	10.66 f
W_3B_2	17.67 c	47.7 e	55.00 d	9.33 g
W_3B_3	19.33 bc	50.46 c	55.00 d	11.67 f
CV%	2.07	11.25	16.79	3.32
$LSD_{0.05}$	2.43	2.71	1.95	1.017

Mean with similar letter do not differ significantly. B_0= Control, B_1= chitosan, B_2= biochar, B_3= chitosan + biochar, W_1= 80% field capacity, W_2= 60% field capacity, W_3= 40% field capacity.

Number of fruits plant^{-1}

The application of biochar and chitosan helps plants reduce drought stress and produce the most fruits per plant in a variety of water-stressed environments (Table 1). Plants treated with chitosan + biochar yielded the most fruits per plant at 60% and 40% moisture levels, i.e., 21.66 and 11.67 respectively, which reduced stress circumstances compared to plants that did not use chitosan and biochar. Foliar spraying of chitosan during the vegetative and blooming stages increases the number of fruits per plant and tomato yield (Mahmood *et al.,* 2017). The high concentration of starch and plant hormones, which are essential for promoting cell division and fruit formation, may be the cause of the rise in fruit production. Plant development, nitrogen uptake, auxin and gibberellic acid concentrations, and yield characteristics, such as fruit weight and number of fruits per plant, are all significantly impacted by biochar (Langeroodi *et al.,* 2019).

Individual fruit weight (g)

The plants treated with chitosan + biochar had the highest individual fruit weight (52.55) and (47.33) in both 60% and 40% moisture levels (Fig. 1). These plants were able to reduce stress circumstances in comparison to their non-treated counterparts. Plants that received a chitosan foliar spray produced more fruit (Mukta *et al.,* 2017). The plant produced more photosynthetic material when chitosan was applied, which increased fruit yield and weight. Biochar is an efficient soil amendment that enhances soil properties and sustains long-term soil productivity and yield (Naeem *et al.,* 2017). Water stress mostly affects plants during their growth and development by upsetting the electrolyte balance, which unquestionably results in a reduction of some vital nutrients that reduce fruit weight and ultimately affect production (Abdalla, 2011).

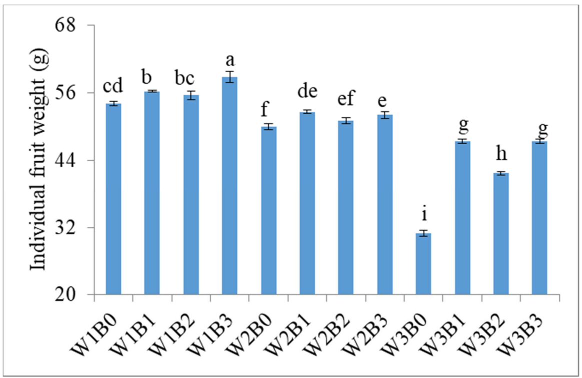

Fig. 1. Effect of biochar and chitosan under various water regimes on individual fruit weight of tomato Mean with similar letter do not differ significantly. B_0= Control, B_1= chitosan, B_2= biochar, B_3= chitosan + biochar, W_1= 80% field capacity, W_2= 60% field capacity, W_3= 40% field capacity

Yield (t/ha)

The production of fruits per hectare in tomatoes was highly impacted by varying water regimes. When plants were kept at varying moisture levels, the highest output (22.30 tons/ha) was noted in those kept at 80% moisture, while the lowest yield (2.92 tons/ha) was found in those kept at 40% moisture. The combination of biochar, chitosan, and different water regimes had a considerable impact on the fruit production per hectare. The highest fruit output (16.85 tons/ha) and (8.20 tons/ha) at 60% and 40% moisture levels, respectively, were found in plants treated with chitosan + biochar (Fig. 2).

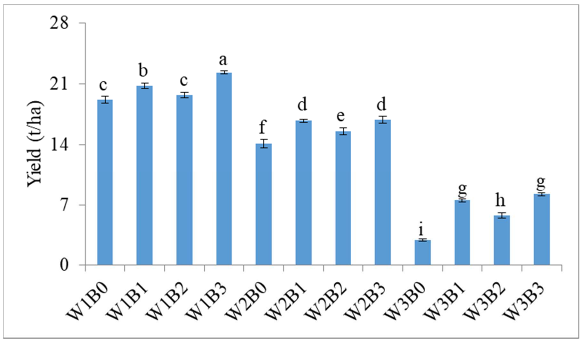

Fig. 2. Effect of biochar and chitosan under various water regimes on yield of tomato

Mean with similar letter do not differ significantly. B_0= Control, B_1= chitosan, B_2= biochar, B_3= chitosan + biochar, W_1= 80% field capacity, W_2= 60% field capacity, W_3= 40% field capacity.

A significant impact on morpho-physiological, biochemical, and molecular functions is caused by drought, and this results in low yield (Bangar *et al.,* 2019). Applications of chitosan have been shown to activate gibberellin in the carpel's ovary, which is essential for cell elongation and, as a result, increases size and volume, ultimately enhancing yield (Mondal *et al.,* 2012). The addition of biochar to the soil increased its water-holding capacity and permeability, both of which increase yield (Langeroodi *et al.,* 2019).

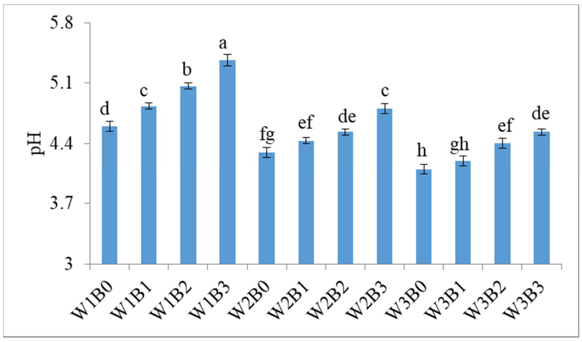

Fig. 3. Effect of biochar and chitosan under various water regimes on pH of tomato

Mean with similar letter do not differ significantly. B_0= Control, B_1= chitosan, B_2= biochar, B_3= chitosan + biochar, W_1= 80% field capacity, W_2= 60% field capacity, W_3= 40% field capacity.

pH

In comparison to drought-stressed plants alone, plants treated with biochar + chitosan in both 60%

and 40% moisture availability displayed the maximum pH (4.80) and (4.53), respectively (Fig. 3). Chitosan influenced the p^H level in fruit juice as chitosan helps to increase the ascorbic acid content in fruits (Gol and Rao, 2011). When tomato plants received less water, they regulate some metabolic activities, such as osmotic adjustment in sink organs, which increases amount of organic acid transformation, thus enhancing acidity content (Rouphael *et al.,* 2008).

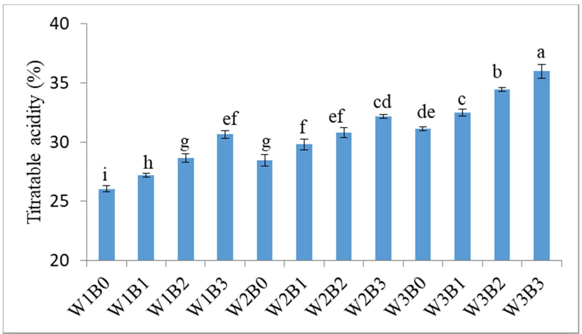

Fig. 4. Effect of biochar and chitosan and various water regimes on titratable acidity of tomato
Mean with similar letter do not differ significantly. B_0= Control, B_1= chitosan, B_2= biochar, B_3= chitosan + biochar, W_1= 80% field capacity, W_2= 60% field capacity, W_3= 40% field capacity.

Titratable Acidity (TA %)

Plants treated with biochar + chitosan had 60% and 40% moisture availability, the largest titrable acidity (32.26) and (36.00) were recorded, respectively, in comparison to plants that were only challenged by drought (Fig. 4). Basit *et al.* (2020) found that titratable acidity in fruit increased significantly in response to chitosan application as compared to the control treatment. Titratable acidity increased due to enhance of organic acid catabolism. Using biochar as a soil ammedment enhanced titratable acidity levels in fruits (Abiven *et al.,* 2015). This occurred due to use of biochar improves fruit acidity and antioxidant component content through increasing nutrient availability in the root zone.

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