CUKUROVA UNIVERSITY
INSTITUTE OF NATURAL AND APPLIED SCIENCES

MSc THESIS

Effects of hydrogels on beans (*Phaseolus vulgaris* L.) growing under salt stress

Kamran ZIKARIA

Horticulture Department

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CUKUROVA UNIVERSITY
INSTITUTE OF NATURAL AND APPLIED SCIENCES

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This Master's Thesis was evaluated by the following Jury Members on 26/09/2024 and was approved by unanimity / majority of votes.

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CONTENTS

ABSTRACT I
ÖZ II
LIST OF TABLES IV
LIST OF FIGURES V
SYMBOLS AND ABBREVIATIONS VI
1. INTRODUCTION 7
2. REVIEW OF LITERATURE 13
3. MATERIALS AND METHODS 23
3.1. Growing conditions 23
3.2. Carried out experimental activities 25
3.3. Plant nutrition 26
3.4. Experimental setup 27
3.5. Treatments 28
3.6. Measurements 28
3.6.1. Morphological parameters 28
3.6.2. Physiological parameters 30
3.6.3. Micro-macro nutrient element analysis in plant leaves and roots 34
3.6.4. Biochemical parameters 35
3.7. Statistical analysis of data 39
4. RESULTS AND DISCUSSIONS 41
4.1. Morphological traits 41
4.2. Physiological traits 44
4.3. Nutrient element analysis in plant leaves and roots 47
4.4. Biochemical traits 52
4.5. Fruit quality traits 57
5. CONCLUSIONS 61
REFERENCES 63
CURRICULUM VITAE 81

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ABSTRACT

Hydrogel polymers have garnered significant interest for their potential to improve water and nutrient retention under abiotic stress conditions. Similarly, the incorporation of biochar can enhance plant growth and development under such stresses. This study investigated the effects of a superabsorbent hydrogel (SAH), composed of alginate-based hydrogel and biochar derived from fruit peels, on common bean performance under salt stress. The experiment involved six treatments: T1 (control), T2 (salt), T3 (salt + 0.5 g SAH), T4 (salt + 1.5 g SAH), T5 (salt + 2.5 g SAH), and T6 (salt + 3.5 g SAH). Salt stress adversely affected plant morphological traits (e.g., plant height, leaf area, root length), physiological traits (e.g., photosynthesis, stomatal conductance, leaf temperature), nutrient uptake, and water absorption. Additionally, salt stress increased reactive oxygen species (ROS) concentrations, leading to enhanced lipid peroxidation and cellular damage. Treatment T3, which included 0.5 g of SAH, mitigated the effects of salt stress by improving root length, root fresh weight, shoot fresh weight, leaf area, number of leaves, leaf water potential, and stomatal conductance. T3 also enhanced water uptake, reduced membrane damage, and decreased sodium and chloride uptake. This treatment resulted in the lowest lipid peroxidation and the highest proline content, along with increased antioxidant enzymatic activity. Based on these findings, it can be concluded that the application of 0.5 g SAH could improve common bean growth and development under salt stress conditions. Since increasing doses of SAH cause Na accumulation in the root, the salt damage-reducing effect may not have been seen with increasing SAH doses.

Keywords: Common Bean, Hydrogel, Biochar, Hydroponics, Plant Growth, NaCl, Stress Mitigation

ÇUKUROVA ÜNİVERSİTESİ
FEN BİLİMLERİ ENSTİTÜSÜ

YÜKSEK LİSANS TEZİ

Hidrojellerin tuz stresi altında büyüyen fasulye (*Phaseolus vulgaris* L.) üzerine etkileri

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ÖZ

Hidrojel polimerler, abiyotik stres koşullarında su ve besin maddesi tutulumunu iyileştirme potansiyeli nedeniyle önemli bir ilgi görmektedir. Benzer şekilde, biyokömür eklenmesi de bu tür stresler altında bitki büyüme ve gelişimini artırabilir. Bu çalışmada, alginat bazlı hidrojel ve meyve kabuklarından türetilmiş biyokömürden oluşan süper emici hidrojeller (SEH) karışımının tuz stresi altındaki fasulye performansı üzerindeki etkileri araştırılmıştır. Deneme, altı uygulamadan oluşmaktadır: T1 (kontrol), T2 (tuz), T3 (tuz + 0.5 g SEH), T4 (tuz + 1.5 g SEH), T5 (tuz + 2.5 g SEH) ve T6 (tuz + 3.5 g SEH). Tuz stresi, bitki morfolojik özelliklerini (örneğin, bitki boyu, yaprak alanı, kök uzunluğu), fizyolojik özellikleri (örneğin, fotosentez, stomal iletkenlik, yaprak sıcaklığı), besin alımını ve su emilimini olumsuz etkilemiştir. Ayrıca, tuz stresi reaktif oksijen türleri konsantrasyonlarını artırarak lipid peroksidasyonunu ve hücresel hasarı artırmıştır. 0.5 g SEH içeren T3 tedavisi, kök uzunluğunu, kök taze ağırlığını, sürgün taze ağırlığını, yaprak alanını, yaprak sayısını, yaprak su potansiyelini ve stomal iletkenliği iyileştirerek tuz stresinin etkilerini hafifletmiştir. T3 ayrıca su alımını artırmış, membran hasarını azaltmış ve sodyum ve klor alımını düşürmüştür. Bu uygulama, en düşük lipid peroksidasyonu ve en yüksek prolin içeriğini sağlamış ve artan antioksidan enzimatik aktivite ile sonuçlanmıştır. Bu bulgulara dayanarak, 0.5g SEH uygulamasının tuz stresi koşullarında fasulyenin büyüme ve gelişimini iyileştirebileceği sonucuna varılabilir. Artan SAH dozları kökte Na birikmesi yaptığından, tuz zararını azaltıcı etki artan SAH dozlarında görülmemmiş olabilir.

Anahtar Kelimeler: Fasulye, Hidrojel, Biochar, Topraksız, Bitki Büyümesi, NaCl, Stres Azaltıcı

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LIST OF TABLES

Table 3. 1. Analysis results of the growing substrate before planting. 24
Table 3. 2. Various activities were conducted during the experiment. 25
Table 3. 3. Names of the mineral fertilizers used to prepare nutrient solution in this experiment... 26
Table 3. 4. The concentration ranges of nutrients used in 100% mineral nutrition control application in the experiment. 26
Table 3. 5. Treatment combinations of the experiment.. 28
Table 4. 1.Effect of superabsorbent hydrogels on some morphological traits of common bean under salt stress. 42
Table 4. 2. Effect of superabsorbent hydrogels on some root traits of common bean under salt stress. 43
Table 4. 3. Effect of superabsorbent hydrogels on some morphological traits of common bean under salt stress. 44
Table 4. 4. Effect of superabsorbent hydrogel on physiological traits of common bean under salt stress. 45
Table 4. 5. Effect of superabsorbent hydrogel on leaf nutrients concentration (%) of common bean under salt stress. 47
Table 4. 6. Effect of superabsorbent hydrogel on leaf nutrients concentration (ppm) of common bean under salt stress. 48
Table 4. 7. Effect of superabsorbent hydrogel on root nutrients (%) uptake of common bean under salt stress. 49
Table 4. 8. Effect of superabsorbent hydrogel on root nutrients (ppm) uptake of common bean under salt stress. 50
Table 4. 9. Effect of superabsorbent hydrogel on stress antioxidant production of common bean under salt stress. 53
Table 4. 10. Effect of superabsorbent hydrogel on total sugar content of common bean under salt stress. 55
Table 4. 11. Effect of superabsorbent hydrogel on leaf temperature of common bean under salt stress. 56
Table 4. 12. Effect of superabsorbent hydrogels on some fruit traits of common bean.................. 56

LIST OF FIGURES

Figure 3. 1. Sowing operations; super absorbent hydrogels application and seedling transferring. 27

Figure 3. 2. Experimental setup of common beans under salt stress in pots. 27

Figure 3. 3. Plants under different treatments (a) control (b) salt (c) salt + 0.5g SAH plant^{-1}, (d) salt + 1.5g SAH plant^{-1} (e) salt + 2.5g SAH plant^{-1} (f) salt + 3.5g SAH plant^{-1}. 29

Figure 3. 4. Common bean pods under salt stress and superabsorbent hydrogel. 29

Figure 3. 5. Common bean roots under salt stress and superabsorbent hydrogel. 30

Figure 3. 6. Determination of plant height and number of leaves. 30

Figure 3. 7. Leaf temperature measurement. 32

Figure 3. 8. Physiological traits measurement (photosynthesis, leaf temperature, etc.). 32

Figure 3. 9. Relative water content determination. 33

Figure 3. 10. Stomatal conductance and chlorophyll content (SPAD) determination. 33

Figure 3. 11. Leaf membrane damage index determination. 33

Figure 3. 12. Leaf water potential determination. 34

Figure 3. 13. Nitrogen determination in leaves and roots. 34

Figure 3. 14. Micro (Fe, MN, Zn, Cu) and macro (K, Ca, Mg) nutrient and Na analysis. 35

Figure 3. 15. Leaf osmotic potential determination. 37

Figure 3. 16. Total sugar content determination. 37

Figure 3. 17. Electrical conductivity and pH determination of drained water. 39

Figure 4. 1. Effect of superabsorbent hydrogel on photosynthesis rate of common bean under salt stress. 46

Figure 4. 2. Effect of superabsorbent hydrogel on physiological responses of common beans under salt stress. 51

Figure 4. 3. Effect of superabsorbent hydrogel on enzymatic activities of common bean under salt stress. 54

Figure 4. 4. Effect of superabsorbent hydrogel on enzymatic activities of common bean under salt stress. 54

Figure 4. 5. Effect of superabsorbent hydrogel on fruit quality traits of common bean under salt stress. 58

Figure 4. 6. Effect of superabsorbent hydrogel on fruit quality traits of common bean under salt stress. 58

Figure 4. 7. Drained water pH. 59

Figure 4. 8. Drained water EC. 59

SYMBOLS AND ABBREVIATIONS

APX : Ascorbate Peroxidase
BC : Biochar
C : Carbon
Ca : Calcium
CAT : Catalase
Cu : Copper
DAS : Days after sowing
DAT : Days after transplant
DMC : Dry Matter Content
Fe : Iron
GR : Glutathione Reductase
K : Potassium
K_2O : Potassium Oxide
MDA : Malondialdehyde
Mg : Magnesium
$MgSO_4$: Magnesium Sulphate
Mn : Manganese
P : Phosphorous
ROS : Reactive Oxygen Species
S : Sulphur
SAH : Superabsorbent hydrogel
SOD : Superoxide Dismutase
Zn : Zinc

1. INTRODUCTION

Common bean (*Phaseolus vulgaris* L.) is an essential food crop known for its high nutrient density and important function as a major vegetable crop. Commonly known as the "grain of hope" because of its abundant nutritional content (Nadeem et al., 2021), it occupies a unique position in the diets of several cultural groups. Turkish common beans are a significant dietary source of vital minerals, calories, and proteins, ranking second only to cereals in terms of nutritional value. On average, Turkish residents consume common beans on a weekly basis, either in their unripe pods, dried seeds, or as part of salads (Nadeem et al., 2018). About 30 million hectares are dedicated to the worldwide cultivation of common beans, resulting in a production output of 27.5 million tons in 2020 (FAO, 2022). Studies suggest that common beans evolved and were initially cultivated in Mesoamerica, then disseminated to Europe through the Columbian exchange, and ultimately arrived in Turkey via the trade routes created by the Ottoman Empire (Nadeem et al., 2021). Therefore, it is not native to Turkey. Nevertheless, the common bean has achieved considerable importance in Turkish agriculture and commerce. Differential farming techniques, local taste preferences, and varied topographies have led to the development of multiple landraces of common beans in various regions of Turkey over time (Nadeem et al., 2018). Common bean production in Turkey has shown a consistent upward trend in recent years. Turkey's yearly output, quantified in dry or fresh weight, has reached 279,518 tons, cementing its position as the third-largest producer worldwide and the leading producer in the Mediterranean area (Yeken et al., 2019).

Abiotic stresses, including drought, salinity, and heat, are important environmental constraints that have a detrimental impact on plant growth and productivity (Lasheen et al. 2024). These stresses pose a substantial risk to cultivable land and result in annual yield losses ranging from 10% to 60% (Alsamadany et al. 2022; EL-Bauome et al. 2022). Saline soils consist of soils with an electrical conductivity (EC) above 4 dS m^{-1} (corresponding to 40 mM Na^+) at a temperature of 25°C, an exchangeable sodium percentage (ESP) below 15%, and a pH level below 8.5 (Sparks, 2003). The buildup of salt in soils can be attributed to various causes, such as excessive application of fertilizers and soil amendments (Shrivastava and Kumar, 2015), irrigation with brackish water, and the infiltration of seawater into groundwater systems, especially in coastal regions. Significant extraction of groundwater for agricultural and other uses might worsen the infiltration of saltwater into freshwater aquifers (Tasnim and Tahsin, 2014). Furthermore, the use of such water for irrigation ultimately results in the gradual buildup of salt in the soil.

Over 20% of the global cultivated land is impacted by salinity, and around 33% of irrigated land is similarly categorized as salt-affected and degraded (Manuel et al., 2017). Salinization of soil deteriorates soil quality by modifying its physical, chemical, and biological characteristics (Bayabil et al., 2021; Warrence et al., 2002; Hemami et al., 2021), resulting in reduced agricultural output (Grundmann et al., 2016). Sustained salt buildup in soil impedes the development and productivity of some crops, especially vegetables, which often have limited tolerance to high salinity (1–2.5 dS m^{-1}) (Manuel et al., 2017). The heightened osmotic potential in saline soils limits the absorption of water by plants, therefore intensifying the difficulty considerably. Sheldon et al. (2017) documented a notable decrease in plants' capacity to metabolize water from saline soils, accompanied by reduced biomass generation caused by unbearable salinity levels.

The efficient control of saline soils is crucial in order to avoid the buildup of salt and improve agricultural outcomes. An optimal approach to reduce salinity is to remove salts from below the root zone by flushing them with freshwater (Bayabil et al., 2022; Perla et al., 2004). In their study, Yan and Marschner (2013) showed that leaching produced a substantial reduction in the electrical conductivity (EC) of three naturally saline soils during an incubation experiment. Nevertheless, leaching is only feasible in areas with a plentiful influx of freshwater, which restricts its use in countries with limited water resources (El-Haddad et al., 2000; Mostafazadeh-Fard et al., 2009). Moreover, the process of leaching presents the potential danger of polluting freshwater systems (Feng et al., 2005; van der Schans et al., 2009).

In plants, abiotic stressors have a profound negative impact on plant biochemical and physiological processes, ultimately leading to diminished growth and reproductive ability (Mahpara et al., 2022). Various environmental stresses, including drought, salinity, and heat, stimulate the generation of reactive oxygen species (ROS) in the chloroplasts, mitochondria, and peroxisomes of plant cells (Rychter, 2006). To cope with these stresses, plants produce a range of suitable organic solutes (Boscaiu and Fita, 2020). At low concentrations, ROS play a crucial role as signalling molecules in stress responses. However, when ROS levels are high, they can lead to substantial oxidative damage by promoting lipid peroxidation, protein breakdown, nucleic acid damage, and DNA denaturation. Oxidative stress hampers the function of enzymes, disturbs the fundamental structures of cells, and can finally result in the demise of cells (Mittler et al., 2011; Cruz et al., 2008; Mattos and Moretti, 2015; Huseynova et al., 2016). Additionally, the photosynthetic system might experience progressive degradation as a result of the buildup of ROS in chloroplasts (Tambussi et al., 2000).

Moller et al. (2007) have suggested that the synthesis of malondialdehyde (MDA) can serve as a predictive measure for the development of ROS, thereby contributing to oxidative damage. Increased concentrations of hydrogen peroxide (H_2O_2) and malondialdehyde (MDA) in plant cells might interfere with adaptive processes responding to abiotic stimuli such salinity and drought, resulting in oxidative stress (Chakraborty et al., 2012). Under conditions of salt stress, the accumulation of MDA is increased, leading to oxidative damage in parenchymal cells (Al-Quraan et al., 2013). Ashraf et al. (2010) showed that elevated levels of MDA have a negative impact on plant growth when subjected to intense salt stress.

Lipid peroxidation, induced by ROS disrupts membrane structure, leading to a loss of membrane selectivity and integrity, as well as imbalances in water and nutrient uptake. These water imbalances in plants can have negative effects on photosynthesis, biomass accumulation, and subsequently root and shoot elongation (Mickky and Aldesuquy, 2019). To counteract ROS, plants have evolved an antioxidant defence system that includes both non-enzymatic and enzymatic antioxidants, such as SOD, CAT, and the enzymes of the ascorbate-glutathione (AsA-GSH) pathway (Ahmad et al., 2010). SOD acts as the first line of defense against elevated ROS levels by converting superoxide anions (O_2^-) into hydrogen peroxide (H_2O_2), which is further broken down into oxygen and water in the plastids, mitochondria, cytoplasm, and peroxisomes. In contrast, peroxidases (POD) directly scavenge H_2O_2 (Budak et al., 2013). The AsA-GSH pathway plays a crucial role in detoxifying ROS and integrates with other defense systems to mitigate oxidative damage caused by abiotic stress (Hasanuzzaman et al., 2019). This pathway involves two key antioxidants, glutathione and ascorbic acid, along with four enzymes: APX, MDHAR, DHAR, and GR. These enzymes contribute to maintaining cellular pools of glutathione and ascorbic acid, thereby protecting plants from abiotic stresses (Pandey et al., 2021). Moreover, these enzymes are integral to the ascorbate-glutathione cycle, which aids in quenching superoxide anions (O^{2-}) and hydrogen peroxide (Farouk, 2011; Wang and Frei, 2011). Subsequently, non-enzymatic mechanisms, including glutathione, ascorbate, tocopherol, carotenoids, alkaloids, non-protein amino acids, and phenolic compounds play a crucial role in preserving the integrity of photosynthetic cell membranes under abiotic stress-induced oxidative stress (Hasanuzzaman et al., 2012). Glutathione is one of the most prevalent non-enzymatic antioxidants and is essential for maintaining redox homeostasis within plant cell compartments (Noctor and Foyer, 1998). In addition, it is vital for plant growth, development, and defense mechanisms against different environmental stresses. Glutathione can directly neutralize ROS or serve as a substrate for enzymes involved in the removal of ROS and toxic compounds. The plant's tolerance to stress is closely related to the redox state of glutathione, which is regulated by the AsA-GSH pathway (Hasanuzzaman et al., 2017a; 2017b).

During stress conditions (salinity and drought), plant cells maintain their water balance through a process known as osmotic adjustment. This adjustment involves the synthesis of osmotically active molecules, including proline, sugar alcohols, glycine betaine, organic acids, soluble sugars, chloride ions, potassium, and calcium. Among these, proline is the most prevalent osmolyte and a key signaling molecule. It is synthesized primarily in the cytosol and plastids while its degradation occurs in the mitochondria (Ashraf et al., 2007). Proline plays multiple roles in plant cells such as stabilizing membranes and proteins buffering cellular redox potential, scavenging free radicals, and mitigating oxidative stress under various abiotic challenges (Hoque et al., 2008). Its accumulation is an early response to water deficit, protecting cells from damage without inhibiting enzymatic activity, even at higher concentrations. The increased accumulation and mobilization of proline during drought stress have been related to enhanced tolerance (Nayyar and Walia, 2003). Similarly, Hong-Bo et al. (2006) identified proline as a critical defense compound against drought.

Salinity and drought stress induces osmotic stress in plants which has a considerable effect on crop productivity due to water limitation (Chun et al., 2018). In response to stress, plant implemented various tolerance mechanisms such as expanding root structures in some species to avoid the high salt concentration in soil surface, and improve the chances of survival in saline conditions, extend roots to get more nutrients, and to increase the water uptake as well as increases biomass, accumulation of osmo-protectants to support the osmotic adjustment, and the expression of different stress-responsive genes (Kulkarni et al., 2017; Ku et al., 2011; Qayyum et al., 2021; Sanders et al., 2012; Reddy et al., 2004).

Water-soluble ascorbic acid and lipid-soluble α-tocopherol function as antioxidants, safeguarding plants from oxidative damage caused by abiotic stress (Gill and Tuteja, 2010; Hollander et al., 2005). In addition, four key enzymes dehydroascorbate reductase, monodehydroascorbate reductase, glutathione reductase, and ascorbate peroxidase, are involved in the ascorbate-glutathione cycle which plays a crucial role in scavenging ROS such as superoxide (O^{2-}) and hydrogen peroxide (Wang and Frei, 2011). Different strategies to mitigate salinity, including the application of biochar (BC), hydrogels (HGs), and organic amendments have been investigated. Organic amendments have been shown to improve nutrient cycling, water-holding capacity, soil porosity, carbon content, soil enzyme activities, organic matter content, and biodiversity in saline soils (Khatun et al., 2019; Dong et al., 2021; Sarma et al., 2017). Prospective remedies to salinity problems are provided by HGs, superabsorbent polymers, and BC amendments (Dhiman et al., 2021). Recent research has emphasized the efficacy of HGs and biochar in alleviating abiotic stressors in plants. The application of hydrogel has been demonstrated to decrease the frequency of irrigation in several crops, providing valuable benefits in terms of time, irrigation expenses, and labor (Kumar et al., 2020).

HGs function as reservoirs for water and nutrients, releasing them gradually in response to plant needs in a controlled manner. This sustained release fosters an optimal environment for root and plant development, which is maintained throughout the growth cycle. HGs are cross-linked macromolecules with a high capacity to absorb and retain significant amounts of water. Through osmosis, HGs absorb water into their hydrophilic matrix, creating an amorphous, gel-like structure that serves as a water reservoir near the rhizosphere (Hanjra and Qureshi, 2010). In agriculture, HGs are particularly useful in arid regions where irrigation water is limited, as they influence soil properties such as water infiltration rates, moisture availability, and aeration (Bakass et al., 2002; Rehman et al., 2011). Studies have shown that soils amended with HGs exhibit improved leaf relative water content and enhanced soil enzyme activities (Baran et al., 2015; El-Hady et al., 2011).

Highly absorbent polymers, such HGs, have been especially designed for application in plant cultivation systems. HGs serve as soil conditioners, improving the availability of macronutrients in the substrate with the aim of maximizing fertilizer effectiveness and fostering enhanced plant growth (Navroski et al., 2015). Moreover, HGs serve to preserve moisture in the plant root zone, facilitating salt dilution and functioning as a water-retaining polymer that reduces salt stress (Lessa et al., 2019;

Neves et al., 2021). HGs can undergo substantial expansion beyond their original dimensions when placed in contact with solvents (Zheng et al., 2018). By using suitable monomers, the swelling process of HGs can be regulated by external factors such as temperature (Zheng and Liu, 2018), salt concentration (Hong et al., 2010), pH (Marcombe et al., 2010), and light (Toh et al., 2014).

Development of environmentally friendly products, such HGs can be achieved by repurposing waste materials (Ahmed et al., 2018; Ibrahim et al., 2015). Experimental results have demonstrated that applying hydrogel (3.0 g L^{-1}) can improve seedling height and total dry mass, even in conditions of moderate and high salinity. These findings indicate that it has the potential to alleviate the negative impacts of saline irrigation water, therefore successfully decreasing salt stress in plants (Sousa et al., 2022).

The potential of HGs to absorb rainwater or irrigation water has garnered significant interest from researchers in the field of agriculture, as it minimizes the need for regular watering. Their utilization ensures the preservation of soil moisture for prolonged durations and controls the retention of nutrients in the soil, therefore promoting plant development and survival (Mohana et al., 2003; Wu et al., 2003). Furthermore, HGs effectively improve ion exchange processes and minimize electrolyte leakage in plants, which are crucial elements for enhancing growth parameters and increasing crop output (Kant et al., 2011; Koupai et al., 2008; Abedi-Koupai et al., 2008; Abou-Baker et al., 2015). Farmers have shown a desire to use HGs but remain apprehensive about possible toxicity and health hazards, especially when HGs are produced from substances like acrylamide, acrylonitrile, or hydroxyethyl methacrylate-based compounds (Klinpituksa et al., 2017).

Typically, HGs are produced by combining synthetic, natural, or hybrid polymers and monomers using several methods such as free radical polymerization, vinyl grafting, ionic contacts, hydrogen bonding, hydrophobic interactions, or physical bonding (Ranjha et al., 2014). Bagasse-derived bio-based HGs present a highly promising alternative for the implementation of saline soil management techniques. The usefulness of bagasse as a hydrogel material is attributed to the presence of structural and functional groups, notably the many hydroxyl (OH) bonds. HGs are renowned for their exceptional swelling characteristics and vast cation exchange capacity, enabling them to efficiently reduce the levels Na^+ and Cl^- ions in soils. In arid soils, these materials have been extensively used to improve water retention, stimulate seed germination, and facilitate plant development (Ibrahim et al., 2015; Das et al., 2021; Liu et al., 2020; Kolambe et al., 2015; Ziogas et al., 2021). The study conducted by Maghchiche et al. (2010) provided evidence that the use of HGs in sandy soils can effectively decrease water loss by 50%. The utilization of HGs as a soil management solution for soils impacted by salt remains insufficiently investigated. Moreover, HGs enhance the physical characteristics of soil by increasing its porosity, thereby reducing the adverse impacts largely linked to conventional irrigation methods, such as soil desiccation following spray irrigation and erosion during gravity irrigation.

Biochar (BC) is a carbonaceous residue generated by the process of pyrolysis of organic substances in an atmosphere with minimal oxygen exposure (Lehmann and Joseph 2015; Ok et al. 2015). Its prospective uses in carbon sequestration, waste management, soil and water remediation, renewable energy production, and amelioration of soil quality and crop productivity have generated significant interest (Azeem et al. 2019; Billa et al., 2019). Moreover, BC has the potential to promote root physiology, thereby facilitating improved absorption of nutrients and water (Jochum et al. 2019; Lin et al. 2020; Niu et al. 2018). Applications of this technology have demonstrated the ability to stimulate plant growth, improve soil fertility (Lehmann 2007), lower soil bulk density (Blanco-Canqui 2017), increase soil water-holding capacity (Basso et al. 2013), decrease nutrient leaching (Ventura et al. 2013), and enhance cation exchange capacity (Nzediegwu et al. 2019). Furthermore, BC has shown effectiveness in environmental cleanup endeavors (Feng et al. 2022).

In their study, Yue et al. (2017) effectively showed that the use of gypsum and BC significantly enhanced the water retention quality of salty soils. Furthermore, Liang et al. (2021) reported that BC considerably enhanced the porosity and plant-available water content of saline-alkali soils, whereas simultaneously decreasing soil bulk density. Studies on the impact of BC addition on saline soils, particularly its interaction with salinity on crop growth and yield metrics, have demonstrated inconsistency and occasionally conflicting results. For example, Akhtar et al. (2015) ascribed the involvement of BC in mitigating salinity stress to its ability to adsorb salt in potatoes. In contrast, Thomas et al. (2013) reported a rise in soil electrical conductivity (EC) when BC was applied to soils modified by salt effects. The anomalies may arise from several aspects associated with both BC and plant features, including the attributes of BC, its source of feedstock, technique and rate of application, possible negative impacts of materials combined with BC, and the salt tolerance of the crops.

The objective of this work is to determine the effectiveness of HGs and BC in mitigating salt stress and their impact on the morphology and physiology of common bean plants. The study seeks to determine the detrimental effects of salt stress on genotypes. Additionally, the research can shed light on bean cultivation with innovative techniques in salt-affected regions.

2. REVIEW OF LITERATURE

Lehmann (2007) emphasized that while conventional renewable energy approaches can reduce fossil fuel CO_2 emissions, they are unable to prevent climate change. BC bioenergy, a process of low-temperature pyrolysis of biomass, is a highly promising approach to both reduce atmospheric CO_2 levels and provide sustainable energy. This process harnesses the off gases generated during the thermal breakdown of wood or grasses to generate heat, power, or biofuels, with BC being a significant secondary product. In comparison to other organic matter forms, BC demonstrates notable environmental advantages, including improved soil cation retention and persistence. Nevertheless, there is ongoing controversy over the precise half-life of BC, which indirectly affects its value in carbon trading. Moreover, the cation retention of fresh BC is lower compared to that of aged BC, and the appropriate circumstances and time for the development of its adsorbing capabilities in BC remain poorly understood.

Kant et al. (2008) examined how varying concentrations of hydrogel (0, 0.5, and 1.0% w/w) affected the physiological characteristics of bean plants exposed to severe salt stress from various sources. The investigation evaluated metrics including dry matter, nutritional composition, chlorophyll concentrations, proline concentration, and ionic equilibrium. The results suggested that the concentration of proline in beans increased as a result of salt stress. The application of a 1.0% hydrogel consistently led to the greatest increases in nitrogen (N) and phosphorus (P) levels in both root and shoot tissues, regardless of the salt treatments. The application of hydrogel to salty soil effectively reduced the negative impacts of high salinity by improving the absorption of nitrogen and phosphorus, decreasing soil electrical conductivity, nitrate levels, proline concentration, and electrolyte diffusion. Furthermore, it enhanced the dry weights (roots and shoots) by promoting the efficient flow of nutrients and water as required. The findings indicate that hydrogel is a very efficient soil conditioner for reducing the negative effects of salt stress and improving the growth of plants in highly saline soils found in arid and semi-arid areas.

Chang et al. (2011) produced ampholytic HGs with pH and salt-responsive characteristics by cross-linking quaternized cellulose (QC) and carboxymethyl cellulose (CMC) with epichlorohydrin (ECH) in an aqueous sodium hydroxide (NaOH) solution. This work aimed to examine the swelling characteristics of QC/CMC hydrogels in relation to the polymer composition, pH level, and salt content. The equilibrium swelling ratio in ultrapure water exhibited substantial variation with polymer composition, rising from 8.6 to 498 g/g as the weight ratio of QC to CMC changed from 3:1 to 1:3 (w/w). The hydrogel Gel32, including a QC to CMC ratio of 3:2, displayed the lowest swelling ratio and maintained electrical neutrality. The H-potential measurements verified the stoichiometry for balanced charge, which aligns with a weight ratio of 1.5 for QC to CMC, which corresponds to Gel32. The HGs exhibited remarkable pH sensitivity throughout a pH distribution spanning from 1 to 13, with significant shrinkage noted at pH 12. The findings showed that CMC predominantly increased swelling as a result of its well-developed water absorption characteristics, whereas QC controlled the charge equilibrium in the QC/CMC system, which contributed to the observed pH sensitivity.

Basso et al. (2013) reported that the addition of BC to sandy loam soil would augment its ability to retain water, and that the extent of BC incorporation would impact the speed at which BC undergoes surface oxidation in the modified soils. To simulate deep banding in rows or uniform topsoil mixing, hardwood fast pyrolysis BC was mixed with the soil at concentrations of 0%, 3%, and 6% (w/w). The resulting mixture was then deposited in columns positioned either in the bottom 11.4 cm or in the top 11.4 cm. Four sets of 18 columns were subjected to incubation at a temperature of 30°C and a relative humidity of 80%. At intervals of 7 days, 150 mL of a 0.001 M calcium chloride solution was introduced to stimulate erosion. Columns were collected at 1, 15, 29, and 91 days after planting. The incorporation of BC resulted in a 23% rise in the gravity-drained water content comparison to the control. The control soils exhibited a progressive increase in bulk density from 1.41 to 1.45 g/cm^3 over time. In contrast, the BC-treated soils maintained a consistent bulk density that was up to 9% lower than the control soils during the whole incubation period. The results indicate that BC enhances the ability of sandy loam soil to retain water and could potentially augment the water storage capacity for agricultural purposes.

Bai et al. (2014) demonstrated that polyacrylamide HGs with a high concentration of lithium chloride can maintain over 70% of their initial water content even in environments with a relative humidity as low as 10% RH. This remarkable water retention capability of these HGs opens up numerous potential applications for their use.

Mehraban and Afrazeh (2014) reported that super absorbent polymers which are hydrophilic gels capable of absorbing and retaining water, then gradually releasing it into the soil, therefore preserving soil moisture levels for prolonged durations. This approach is especially applicable to the growing of mung beans in dry places where limited water availability negatively impacts both growth and productivity. A study undertaken in the Khash region focused on mung beans, a legume crop that thrives in short-season conditions and is abundant in phosphorus and protein. Utilizing a split-plot design within a randomized complete block framework, the study evaluated the impact of different irrigation intervals and polymer application rates on mung bean production and yield components. Testing was conducted at irrigation intervals of 5, 10, and 15 days, with polymer application rates of 0, 15, 30, and 45 kilogrammes per hectare, applied to a depth of 10 cm. The outcomes denoted that the application of 45 kg/ha of the polymer, together with a 10-day irrigation interval, produced the most advantageous outcomes in terms of both yield and yield components.

El-Asmar et al. (2017) emphasized the urgent need of improving water usage efficiency in agriculture, particularly in dry regions. Their study examined the potential of super-absorbent polymers, also known as HGs, to enhance plant survival, productivity, and water use efficiency. Experimental investigations were carried out to evaluate the effects of different rates and techniques of hydrogel application on the performance of plants in clay and sandy clay loam (SCL) soils. This work employed Zea mays and Pinus pinea as representative plants, and applied hydrogel at varying concentrations of 0, 1, 2, 3, and 4 g/kg soil using either banding or mixing methods. Experimental findings showed that the application of hydrogel banding at a concentration of 0.4% in SCL soils resulted in a 25% increase in both fresh and dry biomass of corn, and a 90% extension in the survival of pine seedlings. The application of hydrogel also resulted in a 0.4% increase in evapotranspiration and a 10-13% improvement in water use efficiency in both soil types. In addition, the use of hydrogel in SCL soils enhanced water retention by as much as 33% at a pressure of 100 kPa but had negligible impact on clay soils. The results indicate that the efficiency of HGs in conserving water in agriculture depends on the careful selection of suitable application techniques and enough amounts.

Blanco-Canqui (2017) examines the influence of BC as a soil amendment, with a specific emphasis on its impact on the physical and hydraulic characteristics of the soil. This study emphasizes that BC has the ability to decrease soil bulk density (by 3 to 31%), enhance porosity (by 14 to 64%), and often has no effect on penetration resistance. It improves the structural integrity of wet aggregate by 3 to 226%, boosts the uniformity of soil, and has varying impacts on the stability of dry soil aggregate. Furthermore, BC enhances the availability of water from 4 to 130%. Its impact on saturated hydraulic conductivity varies, declining in soils with a coarse texture and increasing in soils with a fine texture. BC changes water infiltration, soil thermal features, and has a negligible impact on soil water repellency. Minimizing the size of BC particles can improve water retention but may reduce saturation flow. Although BC generally enhances the soil physical environment, the absence of extensive long-term data hinders definitive evaluations.

Nzediegwu et al. (2019) examined the effects of BC produced from plantain peels on the production of potato (Solanum tuberosum L.) when irrigated with wastewater during two consecutive growth seasons, namely 2015 and 2016. The experiment included nine lysimeters (1.0 m × 0.45 m) containing sandy soil with a bulk density of 1.35 Mg m^{-3}. These lysimeters were organized in a completely randomised arrangement, with three replicates. The treatments comprised three groups: (i) wastewater treated with BC, (ii) wastewater desiccated, and (iii) freshwater devoid of BC. A 1% (w/w) application rate of BC was used on the uppermost 0.1 m of soil in 2015. A total of eight irrigations were conducted during a 33-day period, using either freshwater or synthesised wastewater (which is indicative of the wastewater commonly found in developing countries) at 10-day intervals. Vegetative health parameters, such as photosynthetic rate, were evaluated. The potatoes were collected after a period of 120 days, and the fresh weight and count of the tubers were documented. Plant health indices exhibited temporal fluctuations but remained unaltered after the application of BC. Furthermore, there were no notable disparities noted in the overall weight or number of tubers across the different treatments. However, the use of BC greatly enhanced soil pH and cation exchange capacity. The research findings indicate that the application of plantain peel BC as a soil supplement did not have a substantial effect on potato production when operated under wastewater irrigation circumstances.

Nirmala and Guvvali (2019) characterise HGs as very efficient soil conditioners that improve the retention of water and nutrients. Noteworthy examples of commercially available products are Stoosorb, Raindrop, and Agrosorb. Upon the onset of soil drying in the root zone, these HGs operate by facilitating the release of water and nutrients to plants. Hydrogel polymers are artificially produced organic molecules with a large molecular weight that are specifically engineered to absorb water. These polymers exhibit differences in their monomer makeup, water absorption capability, particle size, salinity response, and cost. The utilization of these polymers in soil or synthetic media has various benefits, such as higher water retention capacity, enlarged pore size and quantity, improved soil nutrient reserves, and decreased soil compaction. Recent developments in application technology are expected to expand its use to large-scale commercial agriculture.

Abou-Baker et al. (2020) revealed that HGs had a substantial positive impact on the hydro-physical, chemical, and biological characteristics of soil, exhibiting an EC of 6 dS m^{-1}. In sandy soils, HGs exhibit the ability to decrease irrigation needs, irrigation frequency, and fertilizer leaching, while simultaneously improving soil aggregation, water-holding pores, fine capillary pores, water-holding capacity, irrigation water use efficiency, and nutrient use efficiency. Furthermore, the integration of Agri glass and biofertilizers with HGs generates a novel element that enhances these soil characteristics, thereby enhancing crop yield, especially in challenging circumstances.

Mehmood et al. (2020) investigated the impact of salinity on soybean. The study evaluated the effectiveness of chitosan-modified biochar (CMB) in mitigating salt stress compared to unmodified biochar (BR). Through various analytical techniques—such as Fourier transform infrared spectroscopy (FTIR), scanning electron microscopy (SEM), Brunauer–Emmett–Teller (BET) analysis, elemental analysis, and zeta potential measurements—the researchers observed notable differences between CMB and BR, particularly in Na^+ adsorption kinetics, where pseudo-first and second-order models were most applicable. The findings indicated that salt stress severely hindered soybean growth, root architecture, biomass yield, nutrient uptake, chlorophyll content, and levels of soluble proteins and sugars. However, applying CMB significantly improved these parameters under salt stress conditions. CMB also reduced the salinity-induced increases in Na+, glycine betaine (GB), proline, hydrogen peroxide (H_2O_2), and malondialdehyde (MDA) levels, while enhancing antioxidant activity and boosting the expression of antioxidant enzyme genes (CAT, APX, POD, and SOD) and salt-tolerance genes (GmSALT3 and CHS) already activated by salinity. Overall, the study underscores CMB's critical role in alleviating the hostile effects of salinity on soybean by improving root system architecture, enhancing antioxidant defenses, and upregulating stress-responsive genes, thereby supporting sustainable soybean production under saline conditions.

Feng et al. (2020) underscored the growing importance of sustainable agricultural practices in enhancing water use efficiency, mitigating soil salinity in arid and semi-arid regions, controlling secondary salinization, and improving soil carbon sequestration. To evaluate the efficacy of these green approaches, the researchers performed both laboratory-based soil column simulations and field trials using two amendments: polyacrylamide-based super absorbent polymer (SAP) and corn straw BC, applied at different rates. The laboratory simulations indicated that both SAP and BC significantly reduced soil salinity, with SAP achieving reductions between 9.7% and 26.3%, and BC between 13.5% and 37.2%, contingent upon the application rates. Field experiments demonstrated that the effectiveness of these amendments varied with maize growth stages. SAP was particularly effective in reducing soil salinity during the pre-germination and early jointing stages, while BC provided consistent salinity control throughout the maize growth period. Additionally, both amendments reduced soil electrical conductivity, with BC also enhancing soil levels of Ca^{2+}, Mg^{2+}, and K^{+}. These results highlight that SAP and BC are effective tools for improving soil fertility and managing soil salinity, contributing to environmental sustainability and advanced agricultural practices.

Kumar et al. (2020) reported that more than 60% of India's cultivated land is in dryland regions, with over 30% experiencing insufficient rainfall. HGs have been proposed as a sustainable and environmentally friendly solution to enhance crop productivity under water-limited conditions. These HGs can absorb and retain rainwater and irrigation water, thereby reducing water loss through deep percolation and efficiently utilizing gravitational and capillary water. Their application in soil has been shown to improve growth and yield in various fields, ornamental, and vegetable crops. HGs are typically applied during sowing or near the root zone in nurseries, with an effective rate of just 2.5–5.0 kg/ha, regardless of soil type or climate. In addition to conserving moisture, HGs enhance soil physical characteristics (porosity, water-holding capacity, bulk density), chemical attributes (like cation exchange capacity), and biological factors (such as microbial activity). Since they degrade naturally over time without leaving harmful residues, HGs offer a viable approach for improving agricultural productivity in moisture-stressed environments.

Dhiman et al. (2021) investigated the application of super absorbent polymer (SAP) and a combination of SAP with plantain peel BC to enhance the development and productivity of potatoes (*Solanum tuberosum* L.) and spinach (*Spinacia oleracea* L.) when irrigated with poorly treated wastewater. Given agriculture's status as the primary consumer of freshwater, the utilization of wastewater for irrigation offers a promising remedy, but with the potential drawback of adding pollutants such as heavy metals that can have adverse effects on plants. The objective of the study was to assess the potential of SAP and BC in adsorbing these pollutants and improving agricultural productivity. In lysimeters, potatoes and spinach were cultivated under various treatments including SAP, charcoal, wastewater, and freshwater. Key growth parameters including photosynthesis, stomatal conductance, and yield were actively evaluated. The effects of wastewater irrigation on growth and yield were found to be negligible. Specifically, spinach exhibited a notably greater yield in the SAP+pyrolyzed BC treatment, whereas potato yields were unchanged across all treatments.

The results indicate that these soil amendments can be efficacious in enhancing plant growth through wastewater irrigation, but their influence may differ and be contingent upon the specific crop.

Nassaj-Bokharaei et al. (2021) investigated the application of hydrogel polymers, specifically a composite hydrogel incorporating natural char nanoparticles, to enhance nutrient and water retention in soils. The study evaluated the effects of a starch-based hydrogel-nano natural char composite, applied at three concentrations (0%, 0.3%, and 0.6% w/w), on the growth and nutrition of tomato plants and on soil biological characteristics under varying water-deficit conditions—50% water-holding capacity (WHC) as severe stress, 75% WHC as mild stress, and 85% WHC as non-stress conditions. Results showed that both the concentration of the nanocomposite and the severity of water deficit notably influenced plant growth, nutrient content, and soil microbial characteristics. While drought stress negatively impacted all studied parameters, the hydrogel nanocomposite mitigated these effects. The effectiveness of the nanocomposite varied with concentration and water stress level, with the 0.3% application under non-stress conditions (85% WHC) yielding the most substantial improvements in plant growth (22–45% increase) and nutrient uptake (16–29% increase in P, Fe, and Zn). Additionally, the 0.6% hydrogel nanocomposite under mild stress (75% WHC) enhanced soil respiration (61%) and microbial population size (89%). These findings suggest that the hydrogel-nano natural char composite is a potential amendment for conserving soil moisture, improving tomato growth and nutrient uptake, and supporting soil microbial activity, especially under water-deficient conditions.

Karabay et al. (2021) evaluated the effectiveness of BC in mitigating the adverse effects of salinity stress on salt-sensitive common bean growth. The BC, produced from olive pruning via slow pyrolysis (400°C), was highly alkaline (pH 9.3) with a high CEC, low EC, and a porous structure. Greenhouse-grown common bean plants were subjected to different levels of salt stress (0, 25, 50 mM NaCl) and BC application rates (0, 5, 15 t/ha). Salinity stress decreased shoot growth, biomass, carotenoid and chlorophyll content but BC applications alleviated these effects. Pod setting was also improved under BC treatment. Oxidative stress, measured by increased hydrogen peroxide (H2O2) and cell membrane permeability, was reduced with 5 t/ha BC, while the higher rate (15 t/ha) exacerbated oxidative damage due to elevated soil EC. The study concluded that BC's Na+ adsorption capacity makes it a promising soil amendment to improve common bean growth in saline soils. However, excessive BC can raise soil salinity, suggesting the need for careful management of application rates. Additionally, the study proposes the development of nanocomposite HGs based on BC and functionalized with Fe_3O_4 nanoparticles for water retention, soil amendment, and agricultural support in drought-prone regions.

Malik et al. (2022) emphasized the growing difficulties faced by global agriculture, including urbanization, climate change, sustainable utilization of natural resources, and the buildup of fertilizers. Insufficient water availability, worsened by drought, presents a substantial risk to agriculture, affecting farmers and the socio-economic and environmental equilibrium of countries.

The use of soil amendments, namely superabsorbent polymers (SAPs), presents a promising approach to alleviate the impacts of drought stress. By enhancing water holding capacity, boosting soil porosity (0.26–6.91%), and reducing nitrogen leaching by up to 45%, these environmentally acceptable, non-toxic compounds greatly enhance soil health. SAPs can collect and store water up to several times their own weight, releasing up to 95% of it for plant use. This characteristic makes SAPs very efficient in preserving soil moisture during periods of drought. The analysis highlights the economic advantages of implementing SAPs while recognizing the difficulties related to their implementation in different soil types and drought situations, as well as the rates of adoption among farmers.

Sousa et al. (2022) reported that applying 3.0 g L^{-1} of hydrogel to the substrate enhanced leaf production, even with increasing substrate salinity up to 6.0 dS m^{-1}. Hydrogel concentration improved seedling height, leaf dry mass, and total dry mass as compared to treatments without the water-retaining polymer, effectively mitigating the negative effects of moderate and high salinity. However, the hydrogel's mitigating capacity diminished under more severe salt stress, suggesting a limited effect at higher salinity levels.

Liu et al. (2022) explored the properties, preparation methods, and agricultural applications of superabsorbent hydrogels (SHs), which are three-dimensional cross-linked hydrophilic polymers capable of absorbing and retaining significant amounts of water. Composed of water-affinity monomers, SHs are widely used in fields such as biomedicine, wastewater treatment, hygiene, and slow-release fertilizers (SRFs). Due to their excellent water absorption, water retention capacity, biodegradability and slow-release properties, SHs hold a prominent importance in the SRF market. The study also discusses the use of SHs in double-coated SRFs, their role as nutrient carriers, and models analyzing nutrient release mechanisms. Additionally, the agronomic benefits of SHs, including improved water absorption, enhanced soil water-holding capacity, and increased crop yields, are reviewed. Despite certain limitations, SHs show significant potential for sustainable agriculture in the long term.

Waqar and Bano (2022) investigated the drought stress alleviation effects of HG, BC, poly (acrylic acid), and plant growth-promoting rhizobacteria (PGPR) on soybeans. Before sowing, seeds were soaked in PGPR broth, and BC (5 g/kg soil) and HGs (2 g/kg soil) were mixed into autoclaved soil. After 35 days of growth under optimal conditions, drought stress was introduced on the fourth day. The study documented that drought stress damagingly impacted plant development however, the individual effects of BC, HGs, and PGPR significantly enhanced plant biomass, with BC improving antioxidant enzyme activities (SOD and CAT) and PGPR increasing soil nutrients. PGPR also enhanced hydrogel performance by retaining soil moisture and increasing leaf water content and root fresh weight. The combination of hydrogel and PGPR improved photosynthetic pigments, shoot fresh weight, and soil fertility, while the BC and PGPR combination positively influenced plant osmoregulation and mineral nutrient retention. Overall, BC, HGs, and their combinations with PGPR were effective in mitigating drought stress and promoting plant growth.

Das and Gosh (2022) produced a superabsorbent hydrogel by using charcoal to augment the capacity to retain moisture. Incorporation of BC, which was pyrolyzed at 600°C and defined by its shape and mineral composition, into the hydrogel was achieved by graft-polymerization onto a pine resin backbone. When analyzed using scanning and transmission electron microscopy, the composite material showed notable enhancements in water retention capacity (57-67%), porosity (6-21%), and reductions in bulk density (5-9%). In the range of 37-86 ml/g, the swelling capacity was found to be greater at a pH of 9.2 and lower at pH 4.01. Over a period of 180 days, the material exhibited a weight reduction ranging from 59% to 72%, suggesting its biodegradability with a half-life of 138 days to 162 days. The hydrogel-BC combination exhibits potential as a soil conditioner, controlled-release fertilizer carrier, and moisture retainer, particularly in regions experiencing abiotic stress, therefore promoting sustainable and environmentally non-polluting agriculture.

Hassanin and Samaka (2022) examined the potential of BC and hydrogel amendments to improve the hydro-physical properties of sandy soils in South Mediterranean countries, which are characterized by vast uncultivated sandy lands with poor biochemical, hydrophysical, and nutrient properties. Given the challenges posed by high population density and increased food demand, along with significant crop and orchard residue production, converting these residues into BC has been shown to enhance water retention in sandy soils. Their study investigated the effects of BC (50 g/plant and 100 g/plant), hydrogel (2.7 g/plant), and a combination of BC and hydrogel (2.7 g:50 g) on soil water constants and cowpea fodder crop yield under two irrigation levels (75% and 100%). Results indicated that both BC treatments, especially in combination with hydrogel, improved field capacity and water retention in the soil. Yield variables were higher under the 75% irrigation regime compared to the 100% regime. The study concluded that the effectiveness of BC in enhancing sandy soil properties is influenced by its characteristics and application rates, with BC and hydrogel applications improving soil moisture retention and microporosity, thus increasing crop yields under water-limited conditions.

Tefera et al. (2022) addressed the issue of soil salinity, which affects over 33% of irrigated land globally, by developing a cost-effective and safe hydrogel for soil salinity mitigation. HG was synthesized from liquefied sugarcane bagasse biomass, polyvinyl alcohol, and sodium tetraborate decahydrate. Its effectiveness was evaluated through an incubation experiment at two salinity levels (5 and 10 dS m^{-1}), using four application rates (0%, 1%, 2%, and 3% w/w) with three replications. The study measured pH, electrical conductivity (EC), and concentrations of key ions in porewater and soil. Results demonstrated that the hydrogel significantly reduced porewater EC and soil concentrations of Cl^-, P, Ca^{2+}, and Al^{3+}, with the 3% application rate showing the most substantial effects. However, increased porewater pH and NH_4–N levels were observed at higher salinity levels.

Overall, the hydrogel proved effective in lowering soil salinity and presents potential as a viable amendment for saline soils.

Feng et al. (2022) addressed the issue of soil erosion in karst regions by developing calcium alginate-BC composites (CA-MBs) to enhance nutrient retention and reduce nutrient loss. Two types of composites were synthesized: calcium alginate-rice straw BC (CA-MRB300) and calcium alginate-Eupatorium adenophorum BC (CA-MEB600). Through soil column leaching and pot experiments, the study found that these composites were more effective in retaining nutrients when fertilization was applied compared to non-fertilization. The addition of 0.5 wt% CA-MRB300 and CA-MEB600 significantly decreased nutrient leaching, with CA-MRB300 reducing the cumulative leaching of PO_4^{3-}, NH_4^+, NO_3^-, and K^+ by 48.05%, 28.76%, 36.63%, and 9.08%, respectively. The primary retention mechanisms identified were surface precipitation, ion exchange, and physical adsorption. Furthermore, a 1.5 wt% application of CA-MRB300 improved soybean root length, stem length, and dry weight under non-fertilization conditions, suggesting its potential as a slow-release fertilizer to support soybean growth. This research presents a novel soil amendment aimed at mitigating nutrient loss in karst soils.

Kaur et al. (2023) reviewed the crucial role of HGs in agriculture, focusing on their capacity to retain substantial amounts of water and nutrients within their three-dimensional polymeric networks. This ability allows HGs to improve the local micro-environment around seeds and seedlings, thereby enhancing their growth. The review underscores the advantages of natural biopolymer-based HGs in terms of safety and sustainability. HGs are typically formed through physical interactions or chemical crosslinking of polymer chains, with the degree of crosslinking adjustable to tailor properties such as mechanical strength, porosity, and swelling behaviour for specific agricultural applications. Key applications of HGs include drought resistance for crops, nutrient reservoirs, seed coating agents, and enhanced transplantation success. Their biodegradability and environmental compatibility further support their use in agriculture.

Ahmed et al. (2023) demonstrated that polymers applied at a rate of 2 g/kg exhibited exceptional capacity for retaining both soluble and exchangeable sodium ions. The study found that various natural polymer HGs enhanced tomato fruit production by increasing both the number and fresh weight of the fruits. Additionally, the biopolymer HGs alleviated salinity stress by boosting phenol and flavonoid contents, as well as the activity of antioxidant enzymes such as catalase and peroxidase. They also improved total soluble solids, pH, and juice content, indicating their effectiveness in mitigating the negative impacts of salinity on tomato plants.

Doklega et al. (2024) investigated the impact of irrigation regimes and water-absorbent substances, specifically hydrogel polymers and foliar applications of glutathione (GSH), on common bean plant performance under abiotic stress. The field trials demonstrated that combining hydrogel polymers with GSH significantly enhanced nitrogen (N), phosphorus (P), and potassium (K) accumulation in bean leaves. Notably, the application of GSH and hydrogel polymers under irrigation levels of 100%, 80%, and 60% increased pod yield by 26.5%, 25.23%, and 32.80%, respectively, compared to control treatments. The 100% irrigation combined with hydrogel polymers, with or without GSH, showed the highest improvements in N, P, protein, fiber, carbohydrate percentages, and total soluble dry matter. The study concludes that water-retaining amendments like hydrogel polymers and low-molecular-weight antioxidants such as glutathione can mitigate the adverse effects of elevated ROS, thus enhancing crop productivity and quality. It is recommended that common bean growers use hydrogel polymers (0.24 to 1.20 t ha^{-1}) and apply GSH (1.0 mM) to sustain crop yield and quality under water scarcity conditions.

3. MATERIALS AND METHODS

3.1. Growing conditions

The experiment was carried out during the period of 06/05/2024-05/07/2024 at the growth chamber of the Horticulture Department, Cukurova University, Adana. It was carried out to mitigate the effect of salt stress on common beans by using superabsorbent hydrogels (alginate based) which were prepared after mixing into BC (from apple peels). The hydrogel material was produced by the Romanian project partner and sent to us as part of the TÜBİTAK multinational project, Water4all 223N078. The controlled conditions in the growth chamber were maintained at a temperature of day 25-27°C, night 20-22°C and a humidity of 60-70%, 16 h light and 8 h dark. In this trial, superabsorbent hydrogels (SAH) were used to mitigate salt stress. The experiment layout followed a randomized block design with three replicates per treatment and two plants per replicate. Beans cultivar Bargondia® from Bayer company was used. Six treatments (applied per plant) with sodium chloride (NaCl) salt and HGs along with control were as following: control (T1), salt 50mM (T2), salt 50mM + 0.5g SAH (T3), salt 50mM + 1.5g SAH (T4), salt 50mM + 2.5g SAH (T5) and salt 50mM + 3.5g SAH (T6). In this experiment, each treatment consisted of 3 pots and each pot contained 2 plants. Seeds were initially grown in germination trays using peat moss as the substrate and irrigated with tap water for the first 7 days. Afterward, the seedlings were transplanted into 2 liters capacity polyethylene pots, where SAH was applied to the root zone of the plants (Figure 3.1 and Figure 3.2). The volume ratio of the substrates used per pot were (2L=1.5 potting soil, 1.5 peatmoss (Klasmann Torf TS1 Ltd.), 1 perlite (AgriKal Ltd.): 1.5:1.5:1). Chemical composition of the used growing substrate is presented in table 3.1. Seedlings were irrigated with nutrient solution according to the treatments with salt and without the salt. A nutrient solution for irrigation with an EC value of 3.0 mS/cm and a pH of 6.0-6.5 was used. To induce salinity stress in the plants, 50 mM NaCl was used salty nutrient solution with an EC value of 8 mS/cm and a pH of 6.0-6.5 was applied. Different activities conducted during the experiment are listed in table 3.2. The duration of the experiment was a total of 61 days from the time of seed sowing.

Table 3.1. Analysis results of the growing substrate before planting.

Analysis name	Method	Result	Comments
Saturation	-	54%	Clay loam
Salinity	Saturation Sludge	0.03%	Non-salted
pH	Saturation Sludge	7.50	Slightly alkaline
Lime	Cal symmetrical	27.26%	Very high
Organic Matter	Walkley Black	2.56%	Middle
Phosphorus	Olsen/Spectrophotometers	60 ppm	Very high
Potassium	A. Acetate-AAS	1157.00 ppm	Very high
Calcium	A. Acetate-AAS	4902.00 ppm	High
Magnesium	A. Acetate-AAS	310.00 ppm	Low
Iron	DTPA-AAS	4.44 ppm	Middle
Copper	DTPA-AAS	1.26 ppm	Middle
Manganese	DTPA-AAS	3.04 ppm	Low
Zinc	DTPA-AAS	2.92 ppm	Medium

3.2. Carried out experimental activities

In this experiment various activities were carried out until the final harvesting as in table 3.1.

Table 3.2. Various activities were conducted during the experiment.

Activities	Days after sowing (DAS)
Seedlings transfer	8
1st Irrigation	8
2nd irrigation + 50 mm (beginning of salt application with all subsequent irrigations)	12
3rd, 4th, 5th, 6th, 7th,8th Irrigation	16,18,22,26,29,32
9th Irrigation, enzyme samples collection	34
10th Irrigation, MDA, proline samples collection	36
Stomatal conductance measurement	37
11th Irrigation, plants photography	38
12th Irrigation, SPAD (Chlorophyll) measurements	40
13th Irrigation	41
14th Irrigation, 1st fruit harvest and photography	44
Stomatal conductance measurement	45
15th Irrigation	46
16th Irrigation, old leaves collection and weight	48
17th Irrigation, enzyme, leaf water potential, MDA, proline samples collection	50
Porometer, SPAD, old leaf weight, fruit weight, EC, pH, plants photography	52
2nd fruit harvest and photos, photosynthesis activity, fruits pH, EC, total soluble solids and titrable acidity	53
18th, 19th Irrigation	54, 56
Stomatal conductance measurement	57
Membrane damage, relative water content, old leaves weight	58
20th Irrigation, SPAD, MDA, Proline, osmotic potential, leaf water content	60
Final trial harvest, plant morphology measurements, and sampling for other plant physiological parameters	61

3.3. Plant nutrition

The concentrated solution was prepared separately in two tanks, labeled Stock A and Stock B, using various mineral elements as shown in table 3. 3. The concentrated solution was then diluted based on the requirements for optimal growth conditions of common beans. The specific nutrient prescriptions for common beans are detailed in Tables 3.4.

Table 3.3. Names of the mineral fertilizers used to prepare nutrient solution in this experiment.

Stock A	**Stock B**
Calcium nitrate	Potassium sulphate
Fe – EDDHA	Mono potassium phosphate
	Magnesium sulphate
	Potassium nitrate
	Microelements
	Zinc sulphate
	Boric acid
	Manganese sulphate
	Copper sulphate
	Ammonium molybdate

Table 3.4. The concentration ranges of nutrients used in 100% mineral nutrition control application in the experiment.

Element	**mg/L**
N	150-220
P	30-40
K	270-312
Ca	170-210
Mg	50-65
Fe	3.00-5.00
Zn	0.30-0.55
B	0.70-0.97
Cu	0.20-0.30
Mo	0.10-0.20
Mn	0.55-0.96

3.4. Experimental setup

The seeds were germinated in germination trays using peatmoss as growing substrate in the growth room at favorable conditions and used tap water by the transferring in the pots. Seedlings were transferred when true leaves appeared. At the time of transferring, the superabsorbent hydrogels were applied in the root area of the common beans and irrigated with hydroponically prepared solution according to the prescription of common beans as well as applied salt applications (Table 3.3).

Figure 3.1. Sowing operations; super absorbent hydrogels application and seedling transferring.

Figure 3.2. Experimental setup of common beans under salt stress in pots.

3.5. Treatments

The experiment comprised of control and various treatments of superabsorbent hydrogels (Table 3.5).

Table 3.5. Treatment combinations of the experiment.

Treatments	Doses (g/plant/L growing mix)
T1	Control
T2	Salt 50mM
T3	Salt 50mM + 0.5g SAH
T4	Salt 50mM + 1.5g SAH
T5	Salt 50mM + 2.5g SAH
T6	Salt 50mM + 3.5g SAH

3.6. Measurements

3.6.1. Morphological parameters

Plant height, number of fruits, and root length

The height (Figure 3.3 & 3.6), and root length (Figure 3.5) of bean plants were measured in centimeters using a measuring tape with 1 cm precision. The number of fruits was counted manually for six plants per treatment, and the average was calculated (Figure 3. 4).

Number of leaves and leaf area

The number of leaves was counted manually. Leaf area was determined for 6 plants using a leaf area meter (Li-3100, LICOR, Lincoln, NE, USA), and expressed in cm^2 per plant.

Stem diameter (mm)

Stem diameter was measured for 6 plants using a digital vernier calliper at the root collar, recorded in millimeters (Figure 3.6).

Fresh and dry root weight (g/plant)

After harvesting the plants from the pots, plants were cut from the root collar point and the fresh weight of the green parts was measured with a digital balance. Plant roots were then rinsed in distilled water to remove the growing media particles and other contaminated particles and stored in dry oven for 48 hours at 65ºC to determine dry weight. The readings were made in 6 plants of each treatment and average was taken.

Figure 3.3. Plants under different treatments (a) control (b) salt (c) salt + 0.5g SAH plant^{-1}, (d) salt + 1.5g SAH plant^{-1} (e) salt + 2.5g SAH plant^{-1} (f) salt + 3.5g SAH plant^{-1}.

Figure 3. 4. Common bean pods under salt stress and superabsorbent hydrogel.

Figure 3. 5. Common bean roots under salt stress and superabsorbent hydrogel.

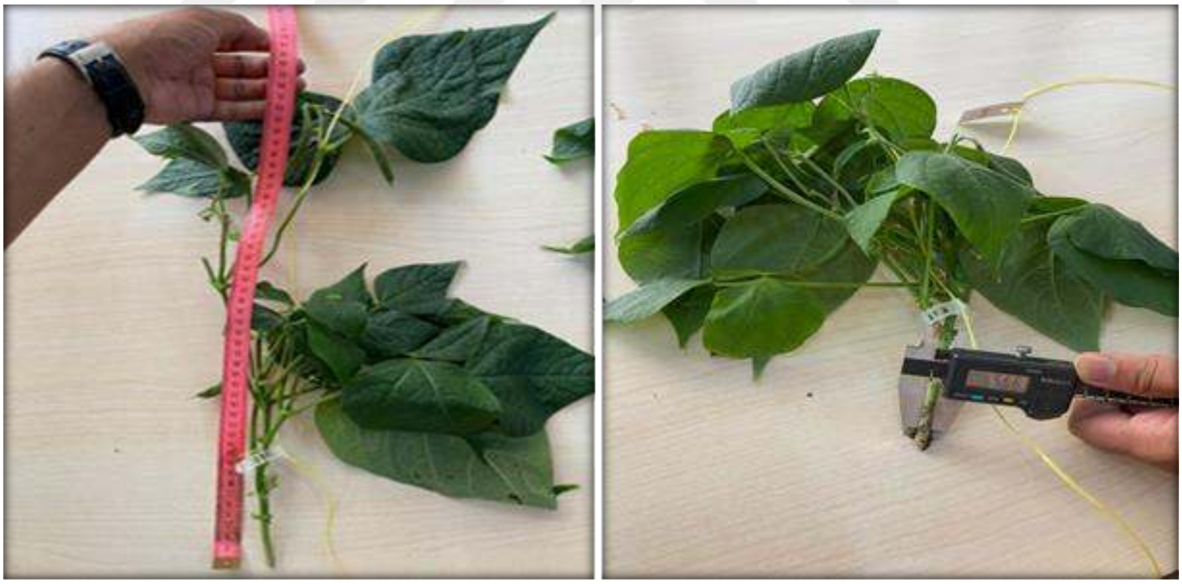

Figure 3. 6. Determination of plant height and number of leaves.

3.6.2. Physiological parameters

Leaf temperature

Leaf temperature was measured with the help of leaf thermometers device (Optris, minisight) in ºC. The readings were taken from six plants per treatment, and statistical analyses were performed on the data (Figure 3.7).

Dry matter content (g/plant)

Plant parts were dried in paper bags in oven at 70ºC for 48 hours and dry weight per plant in grams was determined using digital balance. The readings were taken from six plants per treatment, and statistical analyses were performed on the data.

Photosynthesis rate

The rate of photosynthesis was measured using a portable LI-COR device (LI-6400XT, USA). The readings were taken from six plants per treatment, and statistical analyses were performed on the data (Figure 3.8).

Relative water content (%)

The collected leaf samples' relative water content (RWC) was assessed on the day of harvest. The readings were taken from six plants per treatment. Initially, the leaf samples were submerged in deionized water for 4 h. Subsequently, the turgor weights of the leaf samples were measured following this immersion period. Following these measurements, the leaf samples were dried in an oven at 65°C for 48 h to determine their dry weight (Figure 3.9). The RWC of the leaves was calculated using the following equation

RWC (%) = (FW − DW)/ (TW − DW) × 100
FW: fresh weight, DW: dry weight, TW: turgor weight

Determination of leaf membrane damage index (%)

In case of stress in plant leaves, the membrane damage index (MDI) was calculated by measuring the electrodes released from the cells with an EC meter (Dlugokecka and Kacperska-Palacz, 1978; Fan and Blake, 1994). For this purpose, EC was measured after the leaf discs with a diameter of 1 cm were kept in deionized water for 4 hours, and the EC value of the solution was measured again after the same discs were kept at 100°C for 10 minutes (Figure 3.9). It was calculated as % according to the control using the following formula. The analyses were conducted on six plants per treatment, and statistical analyses were performed on the data (Figure 3. 11).

Membrane Damage Index = (Lt – Lc / 1-Lc) x 100
Lt: EC before autoclaving of leaf in drought stress / EC after autoclaving
Lc: EC before autoclaving of the control sheet / EC after autoclaving

Determination of leaf water potential (MPa)

With a portable Soil moisture brand pressure circle device, the water potential in bar was determined on the 3rd-4th leaves from the main shoot tip and then converted to MPa units

(Akhoundnejad and Dasgan, 2019; Creek, 2019). The readings were taken from six plants per treatment, and statistical analyses were performed on the data (Figure 3. 12).

SPAD (chlorophyll content) and leaf stomatal conductance (mmol m/2 s/1)

Chlorophyll content in the leaves was measured using a SPAD chlorophyll meter (Minolta 502, Osaka, Japan), with readings taken from the same positions on young leaves for consistency. Stomatal conductance was assessed using a Delta T-Devices AP4 portable meter, with multiple readings taken at different times on young leaves. The measurements were taken from six plants for each treatment, and averages were calculated (Figure 3. 10).

Figure 3. 7. Leaf temperature measurement.

Figure 3. 8. Physiological traits measurement (photosynthesis, leaf temperature, etc.).

Figure 3.9. Relative water content determination.

Figure 3.10. Stomatal conductance and chlorophyll content (SPAD) determination.

Figure 3.11. Leaf membrane damage index determination.

Figure 3.12. Leaf water potential determination.

3.6.3. Micro-macro nutrient element analysis in plant leaves and roots

Micro-macro nutrient analysis was made in plant parts to determine the effect of super absorbent HGs applications. The mineral nutrient analysis was conducted by taking one-quarter of six plants from each replication at harvesting, following the method described in Dasgan et al. (2022). The analysis covered macro (N, P, K, Mg, Ca) and micro (Fe, Mn, Cu, Zn) nutrients (Figure 3.13 and 4.14). Common bean leaf samples were cleaned to prevent contamination, washed three times with distilled water and dried at 65ºC for 48 h using the oven dry method. The dried samples were ground to a 40-mesh size using a leaf grinding machine. For the analysis of K, Ca, Mg, Na, Fe, Mn, Zn and Cu, 0.2 g of the ground samples was incinerated at 550ºC for 5 h, and the resulting ash was dissolved in 3.3% (v/v) HCl and then filtered. The prepared samples for K, Ca, Mg and Na were analyzed in the emission mode, while Fe, Mn, Zn and Cu were analyzed in the absorbance mode using an Atomic Absorption Spectrophotometer device (Varian FS220, Las Vegas, NV, USA). Leaf nitrogen and phosphorus were determined using the Kjeldahl and Barton methods, respectively, as described by Dasgan et al. (2023).

Figure 3.13. Nitrogen determination in leaves and roots.

Figure 3.14. Micro (Fe, MN, Zn, Cu) and macro (K, Ca, Mg) nutrient and Na analysis.

3.6.4. Biochemical parameters

Antioxidative enzyme activities

The activity of antioxidant enzymes was assessed by extracting enzymes from 0.5 g of common beans leaf tissue using a mortar and pestle, combined with 5 mL of extraction buffer containing 50 mM potassium-phosphate buffer at pH 7.6 and 0.1mM disodium ethylenediaminetetraacetate. Following centrifugation of the homogenate for 15 min at 15.000 g, the supernatant fraction was utilized for enzyme assays. All enzyme extraction procedures were conducted at 4°C, and activities were determined according to Kusvuran and Dasgan, 2017; Dere et al, 2022; Akhoundnejad and Baran, 2023, İkiz et al., 2024. SOD activity was determined by monitoring the reduction of nitro blue tetrazolium (NBT) induced by superoxide radicals at a wavelength of 560 nm. A unit of SOD activity was defined as the enzyme amount required to inhibit 50% of NBT reduction by photochemical means. CAT activity was determined by monitoring the degradation rate of H_2O_2 at 240 nm. For this analysis, 50 mM phosphate buffer at pH 7.6 containing 0.1 mM EDTA, 0.1 ml of 100 mM H_2O_2, and enzyme extract were added to the reaction medium in a final volume of 1 ml. APX activity was determined by measuring ascorbate consumption at 290 nm. A unit of APX activity was defined as the enzyme amount required to metabolize one mole of ascorbate per minute. GR activity was determined by measuring the absorbance of nicotinamide adenine dinucleotide phosphate (NADPH) at 340 nm and its oxidation rate in the presence of the enzyme. The enzyme volume required to oxidize 1 mol of NADPH per minute was defined as 1 unit of GR activity.

Malondialdehyde and proline analysis (µmol/g FW)

Lipid peroxidation level was calculated based on the MDA (malondialdehyde) level determined using the thiobarbituric acid (TBA) reaction, following the method described by Heath and Packer, (1968) and modified according to Kusvuran and Yilmaz, (2023). Absorbance was measured at 532 nm after centrifugation of the supernatant at 10.000 g for 10 min at 4 °C. Any non-specific absorption at 600 nm was subtracted from the values obtained. The proline content was quantified from aliquots of leaf crude extracts using the method outlined by Magne and Larher, (1992), improved by Dasgan et al. (2009). The analyses were conducted on six plants per treatment, and statistical analyses were performed on the data.

Chloride analysis

0.100 g was weighed from the ground plant sample. 25 ml of pure water was added to it and shaken in the shaker for 45 minutes at 180 rpm. Then, 20 ml of the clear part was taken, and 1 ml of Potassium Chromate (K_2CrO_4) indicator was added to it. The prepared sample was titrated with Silver Nitrate ($AgNO_3$) as reported by Dasgan and Ekici (2004). The analyses were conducted on six plants per treatment, and statistical analyses were performed on the data.

Leaf osmotic potential (MPa)

The method followed was based on Akhoundnejad and Dasgan (2019). The osmometer was calibrated between 12:00-1:00. Samples were taken from the fourth leaf from the tip of experimental plants, stored at -20ºC, and then taken to the lab. One gram of leaf tissue was mixed with 19 ml of distilled water, and after filtration through a 0.45 µm filter, 50 µl of the liquid sample was measured using a Gonatec 030 model osmometer. The values were recorded in MPa. Readings were taken from six plants per treatment, and statistical analyses were performed on the data (Figure 3. 15).

Figure 3.15. Leaf osmotic potential determination.

Total Sugar content analysis (mg/100g DW)

Young leaves were collected from six plants and leaf samples were dried at 65°C (Figure 3.16). the dry samples were grind, and 0.125g sample was weighed and total sugar content were measured by using method of Antron explained by Kaplankiran (1984) and Bolat and Sahin (2016).

Figure 3.16. Total sugar content determination.

Measurements of total soluble solids (TSS), pH and electrical conductivity (EC) in beans pods

The juice from the beans was utilized to measure the TSS (Total Soluble Solids), pH and EC (Electrical Conductivity) using a digital refractometer (Atago PR-101, Tokyo, Japan) and a pH and EC meter (WTW pH/Cond 3320, Weilheim, Germany). The analyses were conducted on six plants per treatment, and statistical analyses were performed on the data.

Vitamin C (L-Ascorbic Acid) content in fruit (mg/100g)

Vitamin C was determined by the modified method of Elgailani et al. (2017). Common bean fruit was blended in juicer machine for extraction. Added 5ml extract in 45ml 0.4% oxalic acid and then filtered. Take 1ml from filtered added in 9ml 2,6 (dichloro phenolindo-phenol sodium salt) and mixed. Lastly, all the samples were read at 520nm in UV spectrophotometer. The analyses were conducted on six plants per treatment, and statistical analyses were performed on the data.

Determination of total phenolic substance

Total phenolic content of bean fruits was determined by modifying the spectrophotometric method with the fruit juice obtained from the bean plant and the readings were calculated by using the absorbance value read at a wavelength of 765 nm in the spectrophotometer and the calibration curve prepared with gallic acid (Spanos and Wrolstad, 1990).

Measurement of Total Phenolic (mg GAE/100 g FW) and Flavonoid Substances in fruits

Total phenolics were determined in bean pods by modifying the spectrophotometric method described by Spanos and Wrolstad (1990). The readings were calculated using the absorbances at a wavelength of 765 nm in a spectrophotometer (UV-1700 PharmoSpec Shimadzu, Kyoto, Japan), and the calibration curve was prepared with gallic acid. Total flavonoids were analyzed using a spectrophotometer at 415 nm, following the method by Quettier-Deleu et al., 2000. The total amount of flavonoid substances was calculated using the calibration curve based on rutin.

Drained solution analysis

During the experiment the drained water EC and pH were determined to observe the effect of super absorbent hydrogels against the salt stress. Drained water was collected in 20ml volume bottles, and measured the EC, and the pH of drained water (Figure 3.17).

Figure 3.17. Electrical conductivity and pH determination of drained water.

3.7. Statistical analysis of data

The variance analysis of the trial data was performed using the JMP statistical package program (Version 13, Statistical Software, 2013). For the parameters that were found to be statistically significant at the $p<0.05$ level, the differences between the applications were examined with the Least Significant Difference (LSD) multiple comparison test and evaluations were made accordingly.

4. RESULTS AND DISCUSSIONS

4.1. Morphological traits

The application of superabsorbent hydrogels (SAH) significantly improved plant height compared to treatments without SAH in salt-stressed conditions. Among all the salt-stressed applications, the highest plant height was statistically observed in the T3 treatment was 38% more than T2. T4 and T5 treatments showed statistically similar results, while the lowest plant height was recorded in the T2 treatment, with a measurement of 30.00cm. Shoot fresh weight was 14% improved in T3 compared to non-treated SAH salt application.

Statistically significant, the higher shoot dry matter 70%, 64% and, 40% were in T3, T4, and T5, respectively, recorded more than salt (T2) application. After the control treatment, the highest stem diameter was recorded in the T4 treatment (5.24 cm), followed by T3 (5.19 cm) and T5 (5.09 cm) (Table 4.1). Salt stress negatively impacted the growth and biomass yield of soybean. Nevertheless, the application of chitosan-modified BC under saline conditions significantly improved growth parameters (Mehmood et al., 2020). Similarly, Turan (2019) reported that the combined use of BC and chitosan enhanced dry biomass production in plants subjected to stress.
Abd El-Mageed et al. (2020) and Hakim et al. (2014) reported that the modified BC in vegetable plants significantly improves plant growth under saline conditions. It was reported HGs improve water uptake decreases abiotic stresses in result increase plant height (Danilova and Tabynbayeya. 2019). The incorporation of Superabsorbent polymers into the soil also conferred a beneficial effect on the biomass generation of quinoa. The observed effect can be attributed to an augmentation in the water-holding capacity, resulting in a notable increase in water retention within the rhizosphere of the plant and, moreover, a decrease in oxidative stress (Nazarli et al., 2010; Islam et al., 2011). Many species have a decrease in biomass production when exposed to hyperosmotic salinity, mostly because of the ionic and osmotic stressors (Munns and Tester, 2008). Akhtar et al. (2015) reported a reduction in the fresh weight of the leaf, root, and plant caused by salt. The study conducted by Tomadoni et al. (2020) yielded evidence supporting the use of alginate-based HGs in horticultural systems. Specifically, lettuce plants cultivated in substrate containing 5% HGs showed an 80% increase in fresh weight compared to the control substrate after 7 days under drought conditions. Abd El-Badea et al. (2011) reported that, the application of soil conditioner veterra hydrogel (VH) and irrigation every 26 days with 150 kg N fed^{-1} positively affected on potato tuber quality attributes like total dry matter (22.71 to 22.83%), starch content (14.78 to 15.70%).

A reduction in dry shoot biomass and the beneficial impact of BC application on shoot biomass under salt stress are consistent with findings from previous research (Akhtar et al., 2015b, 2015c; Agbna et al., 2017; Usman et al., 2016). Treatments incorporating BC resulted in significantly larger stem diameters compared to the control treatment. Agbna et al. (2017) similarly reported increased stem diameter in BC-amended treatments relative to the non-BC control under salt stress.

In addition, studies validate that the salt-induced reduction in leaf, stem, root, and plant fresh weight can be mitigated with the use of BC and superabsorbent polymers (Akhtar et al., 2015).

Table 4.1. Effect of superabsorbent hydrogels on some morphological traits of common bean under salt stress.

Treatment	Plant height (cm)	Shoot fresh weight (g)	Shoot dry matter ratio (%)	Stem diameter (mm)
Control	40.00 a	68.66 a	10.79 a	5.95 a
Salt	30.00 c	41.00 bc	6.12 c	4.84 d
Salt + 0.5g SAH	41.33 a	46.82 b	10.44 a	5.19 bc
Salt + 1.5g SAH	38.50 ab	41.22 bc	10.09 a	5.24 b
Salt + 2.5g SAH	38.50 ab	37.92 cd	8.61 b	5.09 bc
Salt + 3.5g SAH	34.33 bc	32.72 d	8.05 b	5.02 c
LSD (0.05)	4.364	6.125	1.029	0.177
P-value	0.0017*	0.0001*	0.0001*	0.0001*

The control group had the highest root length (49.00 cm), while the salt treatment significantly reduced it to 21.66 cm. The addition of SAH (at different concentrations) to the salt treatment increased the root length by 85%, 89%, 89%, 85% in T3, T4, T5, and T6, respectively, compared to the salt (T2) application. fresh weight was increased by 17% in T3, and 21% in T4 application compared to T2. The control plants had the highest root dry matter ratio (12.64 a), while salt stress significantly reduced it to 9.98% (c), demonstrating the negative impact of salt on root biomass. Applying SAH at 0.5g, 1.5g, and 2.5g significantly improved the root dry matter ratio (11.52–11.69 b). it's a close to control application. However, the 3.5g SAH treatment resulted in a ratio similar to the salt treatment (10.11 c), showing diminished effectiveness at higher doses. The superabsorbent hydrogels (SAH) treatments at moderate doses (0.5g, 1.5g, 2.5g) effectively mitigated the salt stress effect, improving root dry matter ratio, while the highest SAH dose (3.5g) was less effective (Table 4.2).

Salinity inhibits root extension by directly inhibiting the cell wall extensibility (Bressan et al., 1990). Caines and Shennan. (1999) observed that root length was lower in higher salinity treatments. The application of BC has been shown to significantly enhance root growth, potentially by improving soil properties and reducing the uptake of Na^+ through the roots. Previous research on common beans has demonstrated that BC application notably promotes root development (Luo et al., 2017; Farhangi Abriz and Torabian, 2017, 2018). Farhangi-Abriz and Torabian (2018) observed that BC addition reduced Na^+ content in the roots under salt stress, which is beneficial for root growth. As a result, the application of BC increases root growth, thereby improving overall plant growth and yield under saline conditions. Modified BC has also been shown to significantly enhance plant growth in vegetables, such as peppers, under saline conditions (Abd El-Mageed et al., 2020). Additionally, while salt stress can impair root architecture, chitosan-modified BC has been found to significantly improve morphological and physiological parameters of plants under such stress (Mehmood et al., 2020). Turan (2019) demonstrated that both BC and chitosan contribute to increased dry biomass of plants under abiotic stress conditions. Moreover, research on the particle size of growth mediums has confirmed that they significantly enhance root fresh weight and length (Tzortzakis and Massa, 2022).

Table 4.2. Effect of superabsorbent hydrogels on some root traits of common bean under salt stress

Treatment	**Root length (cm)**	**Root fresh weight (g)**	**Root dry matter ratio (%)**
Control	49.00 a	5.81	12.64 a
Salt	21.66 c	3.38	9.98 c
Salt + 0.5g SAH	40.00 b	3.94	11.52 b
Salt + 1.5g SAH	41.00 b	4.10	11.69 b
Salt + 2.5g SAH	41.00 b	3.43	11.67 b
Salt + 3.5g SAH	40.00 b	3.87	10.11 c
LSD (0.05)	3.556	NS	0.618
P-value	0.0001*	0.1552	0.0001*

The highest leaf area was recorded in the T3, T2, and T6 treatments, after the control group. The use of HGs led to significant improvements in mitigating salt stress. The T3 treatment (0.5g SAH + salt) among all salt-stressed applications produced the highest number of leaves, with a count of 14. After this, T2, T4, T5, and T6 treatments exhibited statistically similar and lower leaf numbers. Shoot fresh weight significantly increases 92% in T3, 65 in T4, 52% in T5, and 31% in T6 (Table 4.3). Abd El-Mageed et al. (2020) reported that the modified BC in pepper plants significantly improves plant growth. BC has been shown to positively impact leaf area under saline conditions (Akhtar et al., 2015c). Sweet Pepper plants exhibited increased leaf area when treated with hydrogel (Singh, 2012). Most studies highlight the benefits of BC in terms of biodegradability, reduced phytotoxicity, and enhanced plant growth performance (Farzi et al., 2017, Mazloom et al., 2019). In addition, the physicochemical properties of zeolite-amended hydrogel substrates have been reported to improve water retention and positively influence various plant growth parameters, such as leaf number, height, leaf area, flowers, and branches (Danilova and Tabynbayev, 2019).

Table 4.3. Effect of superabsorbent hydrogels on some morphological traits of common bean under salt stress

Treatment	Leaf area (cm^2/Plant)	Number of leaves/plants	Shoot fresh weight (g/Plant)
Control	2773.10 a	20.00 a	55.12 a
Salt	1106.06 c	11.66 c	16.69 e
Salt + 0.5g SAH	1348.07 b	14.00 b	32.17 b
Salt + 1.5g SAH	1193.86 bc	12.00 c	27.54 c
Salt + 2.5g SAH	1162.bc	12.00 c	25.50 c
Salt + 3.5g SAH	1062.99 c	11.50 c	21.92 d
LSD (0.05)	198.75	1.463	2.941
P-value	0.0001*	0.0001*	0.0001*

4.2. Physiological traits

Relative water content in plant leaves was higher in the control group compared to other treatments. However, T5 and T6 showed a significant increase in leaf relative water content compared to the other treatments, with the lowest values recorded in the salt (T2) treatment. Water potential in leaves was significantly higher in the control, while the lowest was measured in the salt-stressed plants. T3 and T4 also showed significantly higher water potential than the salt treatment, with values of -0.17 and -0.20 MPa, respectively. Stomatal conductance was highest in the control group, while among the salt treatments, T3(0.5g SAH) had the highest conductance. In contrast, T2, T4, T5, and T6 exhibited statistically lower stomatal conductance under salt stress. The highest leaf osmotic potential was recorded in the control group, with statistically similar values observed in T2, T3, T4, and T5. The lowest osmotic potential was measured in the T6 treatment. Regarding membrane damage in leaves, SAH significantly reduced the extent of damage, with reductions of 28%, 20%, 21%, and 14% in T4, T6, T5, and T3, respectively, compared to the salt (T2) treatment (Table 4.4).

Application of BC significantly improved leaf relative water content with Hydrogel BC under abiotic stressed vegetables such as potato (Akhtar et al., 2015). Applications of superabsorbent polymer improved relative water content in the leaves and enhanced the soil capacity for water retention which resulted in enhanced growth (Yu et al., 2019; Waqar et al., 2022). The BC and superabsorbent polymers application significantly causes an increase in stomatal gas exchange in the plant under 300mM salinity stress (Derbali et al., 2023). Hyperosmotic salinity decreases biomass production, predominantly due to the ionic and osmotic stresses (Munns and Tester. 2008). BC plays a crucial role in improving plant resilience when grown in saline conditions by enabling a higher root surface area and reduced drainage of water (Taban and Movahedi, 2006). The major photosynthetic mechanisms can be affected by abiotic stresses, such as photosynthetic pigments, photosystems, electron transport systems, gas-exchange processes and enzymes involved in carbon metabolism (Ashraf and Harris. 2013). The BC induced increase in the chlorophyll content leaves was accompanied by an increase in CO_2 fixation (from 6.36 to 12.51 CO_2 µmol m^{-2} s^{-1}) (Speratti et al., 2018; Sun et al; 2017; Akhtar et al, 2015). HGs augment the chlorophyll concentration, leaf transpiration, and stomatal conductance in plants, therefore exerting a beneficial effect on their ability to fix CO2. Application of SAH enhances leaf conductance, stomatal transpiration, and reduces leaf membrane damage in common bean plants during severe abiotic stress (Heidari and Golpayegani, 2012; Kenawy et al., 2022). Conversely, a rise in the salinity of irrigation water results in a substantial decrease in stomatal conductance and the rate of photosynthesis, together with higher leaf temperatures and electrolyte leakage (Parkash and Singh, 2020). BC application help to increase g_S and photosynthesis rate and decrease leaf temperature and electrolyte leakage in tissue of leaves (Parkash and Singh. 2020). The high salinity levels, osmotic stress in the root zone increases, which results in a reduction in transpiration rate (Testi et al., 2008). Higher leaf temperature inhibits the activity of photosystem II, inactivates Rubisco, and disintegrates plasmalemma (Camejo et al., 2005). Electrolyte leakage occurs in response to stress-induced injuries to tissues (Bajji et al., 2002; Blum and Ebercon, 1981; Levitt and Levitt, 1987). Electrolyte leakage is an indirect method to measure the integrity and stability of the cell membrane. The cell membrane acts as a barrier, and its disintegration results in the death of the cell (Mcneil and Steinhardt, 1997; Hameed et al., 2012). Hyperosmotic salinity reduces biomass production due to the ionic and osmotic stresses (Munns and Tester. 2008).

Table 4.4. Effect of superabsorbent hydrogel on physiological traits of common bean under salt stress

Treatments	**Leaf relative water content (%)**	**Leaf water potential (MPa)**	**Stomatal conductance (mmol H_2O /m^2/s^1)**	**Leaf osmotic potential**	**Membrane damage index (%)**
Control	78.14 a	-0.13 a	368.33 a	-0.215 a	-
Salt	67.02 d	-0.41 e	134.50 c	-0.264 b	66.24 a
Salt + 0.5gSAH	68.13 cd	-0.17 b	175.00 b	-0,274 b	47.50 c
Salt + 1.5g SAH	72.75 bc	-0.20 c	134.50 c	-0.274 b	56.89 b
Salt + 2.5g SAH	73.19 b	-0.25 d	117.50 c	-0.269 b	52.35 bc
Salt + 3.5g SAH	76.95 ab	-0.22 cd	117.50 c	-0.294 c	52.09 bc
LSD (0.05)	4.95	0.025	20.75	0.012	7.720
P-value	0.0021*	0.0001*	0.0001*	0.0001*	0.0047*

The photosynthesis rate in common beans was found to be statistically significant, with the highest rate observed in the control application. When comparing all treatments to the salt application without SAH (T2), the addition of SAH (T3-T6) notably increased the photosynthesis rate by 111%, 90%, 48%, and 38% in T4, T3, T5, and T6, respectively, compared to the salt (T2) group (Figure 4.1). Thereafter, excessive amounts of salt enter the shoot and eventually rise to toxic levels in the older transpiring leaves; this toxicity causes premature senescence, reducing the photosynthetic leaf area of the plant to a level that cannot sustain growth (Munns and Gilliham, 2015).

In their study, Ning et al. (2018) showed that soybean plants subjected to saline stress need an enhanced synthesis of photosynthetic products in their roots in order to efficiently assimilate adequate moisture and nutrients for proper growth. Key elements of photosynthetic processes, such as photosynthetic pigments, photosystems, electron transport chains, gas-exchange processes, and enzymes involved in carbon metabolism, are essential for efficient photosynthetic activity and can be negatively impacted by abiotic stresses like salinity (Ashraf and Harris, 2013). BC significantly improves stomatal conductance and photosynthetic rate, while also decreasing leaf temperature and electrolyte leakage in leaf tissues (Parkash and Singh, 2020). Elevated leaf temperatures can suppress the function of photosystem II, render Rubisco inactive, and compromise the structural integrity of the plasmalemma (Camejo et al., 2005).

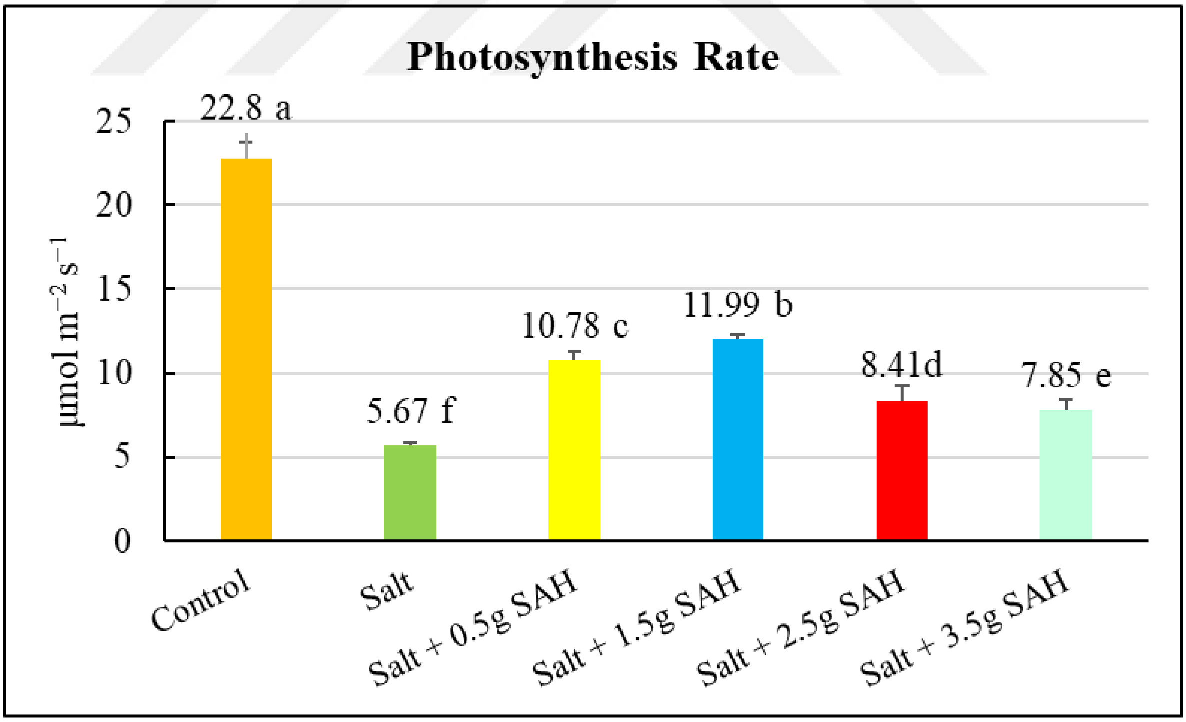

Figure 4.1. Effect of superabsorbent hydrogel on photosynthesis rate of common bean under salt stress.

4.3. Nutrient element analysis in plant leaves and roots

In terms of nitrogen content in leaves of common beans, T3 showed a significant increase compared to T2, with the highest nitrogen content observed in the control treatment. The highest phosphorus content was found in T2, thereafter in T3 application. T4 and T6 were statistically similar in phosphorus content. Magnesium content was significantly higher in T5, showing a 35% increase compared to the control. Other treatments, including T6, T2, T4, and T3, exhibited increases of 32%, 28%, 25%, and 21%, respectively, over the control. Potassium plays a vital role to cope salt stress in plants which was significantly increased in SAH applications: 35%, 28%, 15%, 4% was increased in T5, T4, T6, and T4 application, respectively, compared to untreated SAH salt application (T2). Calcium level was decreased non-significant across all treatments compare to control (Table 4.5). Salt-stress decreased the soybean nutrients acquisition while chitosan modified BC with salt-stress notably enhanced the growth parameters (Mehmood et al., 2020).

Table 4.5. Effect of superabsorbent hydrogel on leaf nutrients concentration (%) of common bean under salt stress.

Treatments (Leaf)	Nitrogen	Phosphorous	Magnesium	Potassium	Calcium
Control	4.52 a	1.20 bc	1.04 c	2.09 d	6.93 a
Salt	3.82 bc	1.40 a	1.34 ab	3.18 c	6.22 b
Salt + 0.5g SAH	3.84 b	1.34 b	1.26 b	3.31 c	6.18 b
Salt + 1.5g SAH	3.74 cd	1.29 c	1.30 ab	4.08 a	5.70 bc
Salt + 2.5g SAH	3.33 e	1.19 d	1.41 a	4.32 a	5.45 c
Salt + 3.5g SAH	3.72 d	1.27 c	1.38 ab	3.68 b	5.38 c
LSD (0.05)	0.095	0.050	0.142	3.729	0.618
P-value	0.0001*	0.0001*	0.0024*	0.0001*	0.0019*

The micronutrient concentrations (manganese, copper, iron, and zinc) in common bean leaves are under salt stress and treatments with SAH. Salt stress increased manganese (120.16), Iron (207.33), and zinc (34.41) compared to the control, while copper levels remained unchanged were statistically non-significant. SAH treatments maintained high manganese levels (120.30–122.85) in T3-T5), with the 3.5g SAH treatment slightly reducing manganese (108.36). The manganese was slightly increase by 0.12%, 2.03%, and 2.24% in T3, T4, and T5 application, respectively. Iron significantly reduced by 8% and 9% in T3 and T4 application, respectively due to SAH compared to salt application. Zinc levels varied significantly with the 1.5g and 3.5g SAH treatments showing a -17% and -13% reduction than salt application. Copper levels were not significantly affected in all applications (Table 4.6).

The application of SAH in saline soils can be advantageous, as saline conditions often reduce phosphorus availability (Rezapour et al., 2021; Ding et al., 2020). HGs have the potential to absorb phosphorus, thereby enhancing their availability to plants over time (Ding et al., 2020). Shi et al. (2010) found that the application of hydrogel increased potassium ion (K^+) concentrations in soil, as hydrogel consists of a potassium-based polymer. In addition, Saruchi et al. (2015) reported significant increases in potassium, phosphorus, and organic carbon concentrations due to hydrogel application. Potassium plays a crucial role in reducing water loss and preventing crop wilting by regulating stomatal openings (Saruchi et al., 2014; 2015). In our experimental results, SAH applications also increased the K content in the leaves This finding is consistent with the literature.

Table 4.6. Effect of superabsorbent hydrogel on leaf nutrients concentration (ppm) of common bean under salt stress.

Treatments (Leaf)	**Manganese**	**Copper**	**Iron**	**Zinc**
Control	51.70 c	18.60	127.95 c	33.46 a
Salt	120.16 a	18.63	207.33 a	34.41 a
Salt + 0.5g SAH	120.30 a	18.65	189. 95 b	35.20 a
Salt + 1.5g SAH	122.60 a	15.35	187.25 b	28.23 b
Salt + 2.5g SAH	122.85 a	16.80	214.85 a	33.39 a
Salt + 3.5g SAH	108.36 b	15.40	204.10 a	29.68 b
LSD (0.05)	9.950	NS	12.187	3.417
P-value	0.0001*	0.1099	0.0001*	0.0058*

The highest nitrogen content in common bean roots was significantly recorded in T4 and T5, with increases of 7.56% and 5.97%, respectively, compared to the control. The highest phosphorus content across all treatments was observed in T4, measuring 1.57. Magnesium content was 3% higher in SAH application (T3) application than T2. The potassium concentration was statistically less in all salt applications in comparison to control. BC amendments improve soil water-holding capacity and increase the availability of essential nutrients such as N and P, which can lead to improved plant yield and increased fresh weight (Yu et al., 2019; Van Zwieten et al., 2010).

Table 4.7. Effect of superabsorbent hydrogel on root nutrients (%) uptake of common bean under salt stress.

Treatments (Roots)	**Nitrogen**	**Phosphorous**	**Magnesium**	**Potassium**
Control	2.51 b	1.48 abc	0.95 a	1.05 a
Salt	2.39 c	1.55 ab	0.82 bc	0.44 b
Salt + 0.5g SAH	2.31 cd	1.44 bc	0.85 b	0.43 b
Salt + 1.5g SAH	2.70 a	1.57 a	0.82 bc	0.40 b
Salt + 2.5g SAH	2.66 a	1.37 c	0.74 d	0.41 b
Salt + 3.5g SAH	2.24 d	1.45 bc	0.79 c	0.35 b
LSD (0.05)	0.096	0.117	0.034	0.111
P-value	0.0001*	0.0319*	0.0001*	0.0001*

Salt stress significantly increased manganese concentration (32.70) compared to the control (19.80). SAH treatments further increased the manganese, 47% with 1.5g SAH showing the highest value (72.15). All SAH treatments significantly improved manganese uptake compared to salt stress alone. Copper level was (27.15) compared to the control (16.86). The 2.5g SAH treatment maintained a high copper concentration (28.63 was more than 5% in T2 comparison), like the salt treatment, while other SAH treatments significantly reduced copper levels (17.35–22.75), closer to control levels. Salt stress reduced iron levels (453.10) compared to the control (542.95). All SAH treatments restored or increased iron content, with 1.5g and 0.5g SAH showing the highest levels (657.25 and 626.95, respectively), statistically different from salt treatment. T3, and T4 showed a significant increase in roots iron uptake by 38%, and 45%, respectively. Salinity significantly increased zinc levels (165.45) compared to the control (106.95). The 1.5g and 3.5g SAH treatments further increased zinc (176.57 and 175.17), increase in 6.7% and 5.8% indicating a significant improvement in zinc uptake compared to salt alone (Table 4.8). BC can adsorb and retain micro-nutrients to improve soil fertility which are leach in the soil, such as N, P, K etc. (Noreen et al., 2020). The application of hydrogel at a concentration of 10 dS/m resulted in an average reduction of phosphorus concentration by 9% relative to the control group (Tefera et al., 2022). Additionally, HGs have been demonstrated to significantly decrease phosphorus concentration in the soil (Mazloom et al., 2020).

Table 4.8. Effect of superabsorbent hydrogel on root nutrients (ppm) uptake of common bean under salt stress

Treatments (Roots)	Manganese	Copper	Iron	Zinc
Control	19.80 d	16.86 c	542.95 b	106.95 d
Salt	32.70 c	27.15 a	453.10 d	165.45 b
Salt + 0.5g SAH	48.30 b	17.35 c	626.95 a	133.86 c
Salt + 1.5g SAH	72.15 a	18.00 c	657.25 a	176.57 a
Salt + 2.5g SAH	31.85 c	28.63 a	442.15 d	168.13 ab
Salt + 3.5g SAH	49.00 b	22.75 b	511.20 c	175.17 a
LSD (0.05)	3.556	3.124	31.316	9.157
P-value	0.0001*	0.0001*	0.0001*	0.0001*

The control plants had a low chloride (Cl) content (2.10) in leaves, while salt stress increased it to 9.36 a, indicating a significant accumulation of chloride in the leaves due to salt stress. SAH treatments at 0.5g and 1.5g reduced chloride content compared to the salt treatment (7.49 and 7.96, respectively), showing that moderate. SAH doses helped lower chloride accumulation in the leaves. However, higher SAH doses (2.5g and 3.5g) resulted in chloride levels similar to the salt treatment (8.89 and 8.77), suggesting that at higher concentrations, SAH is less effective in mitigating chloride buildup. In a nutshell, the SAH reduced chloride accumulation by 19%, 14%, 5%, and 6% in T3, T4, T5, and T6 in comparison with salt application with 0g SHG (Figure 4.2). Excessive chloride accumulation in leaves due to salt stress can lead to ionic toxicity, which affects plant metabolism, reduces photosynthesis, and hinders growth. The SAH treatments, particularly at lower doses (0.5g and 1.5g), significantly reduce chloride buildup, helping alleviate some of the negative effects of salt stress.

The control treatment had the lowest chloride content (1.42) in roots, while salt stress significantly increased chloride accumulation in the roots (2.33), demonstrating the detrimental effect of salinity on chloride uptake in the roots. The addition of SAH at 0.5g and 1.5g moderately reduced chloride content compared to the salt treatment (1.98 and 1.88, respectively), indicating that these doses of SAH help mitigate chloride intake under salt stress. Higher doses of SAH (2.5g and 3.5g) brought the chloride content closer to control levels (1.51 and 1.61), suggesting that higher doses of SAH are more effective at reducing chloride accumulation in the roots than lower doses. Higher doses of SAH decrease the chloride intake by 15%, 19%, 35%, and 30% in T3, T4, T5, and T6 applications, respectively (Figure 4.2). Salt stress significantly increases chloride uptake in roots, which can lead to ionic toxicity, impairing root function and overall plant health. The SAH treatments, particularly at higher doses (2.5g and 3.5g), effectively reduce chloride accumulation in the roots, offering better protection against the harmful effects of salinity compared to lower doses. This highlights the potential of SAH in mitigating salt-induced chloride toxicity in roots.

Sodium (Na) levels in plant reduced with 0.5g, 1.5g, 2.5g, and 3.5g of SAH application was -50.76%, -40.15%, -32.58%, -23.48% compared to the salt treatment(T2). The SAH effectively reduced the accumulation of Na in the plant. These SAH treatments reduce sodium levels compared to the salt-only treatment. Among these, 0.5g of SAH appears to be the most effective in mitigating sodium accumulation by 5.66% in comparison to salt application in roots. The increase in root sodium with the salt treatment confirms the expected response to salinity stress. The application of SAH reduced sodium levels compared to salt alone, with the 0.5g dose showing the most pronounced reduction. However, higher doses of SAH (2.5g and 3.5g) do not improve and may exacerbate sodium accumulation. Hydrogels enhance the potassium (K^+) availability to the plant roots in result potassium tried to maintain the Na balance in plant roots. Whatever, the increase in potassium in plant roots maintained the Na accumulation in plant roots.

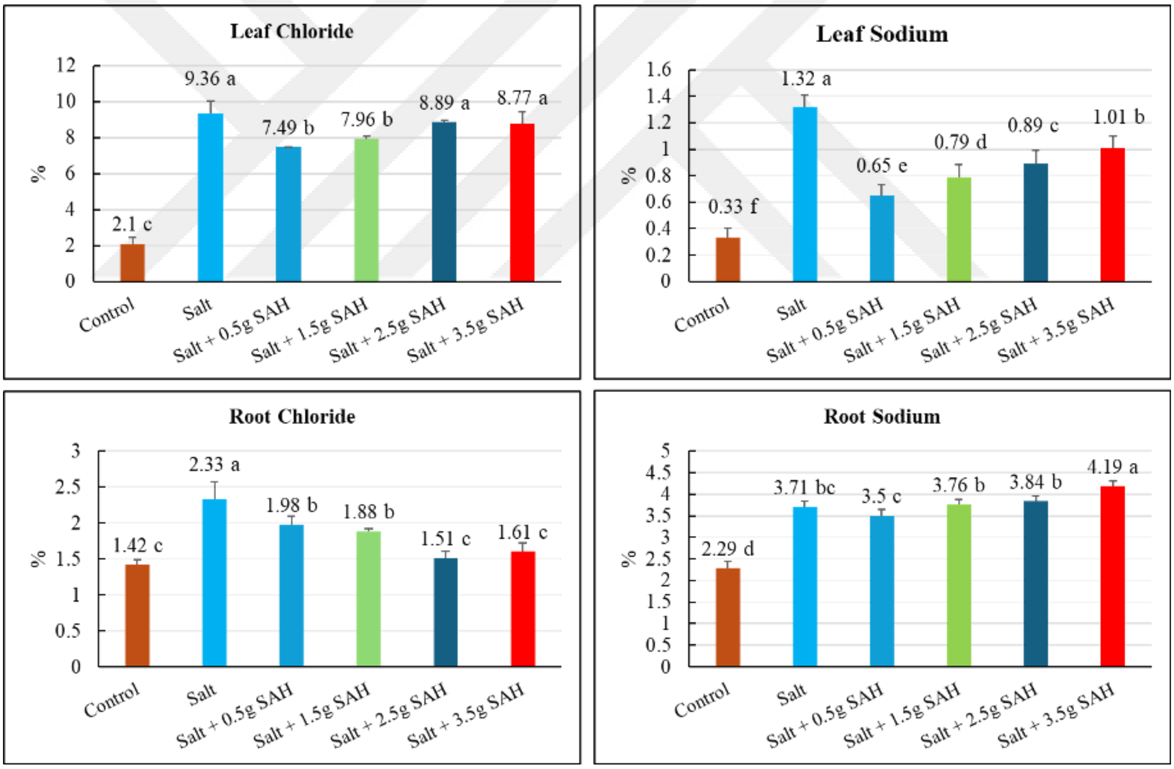

Figure 4.2. Effect of superabsorbent hydrogel on physiological responses of common beans under salt stress.

Hydrogel application has been demonstrated to decrease concentrations of Cl^-, P, and Ca^{2+} in soil, as well as to significantly mitigate soil salinity (Tefera et al., 2022). Wang et al. (2004) found that hydrogel substantially reduces chloride concentration in soil. Chloride is a micronutrient vital for plant growth, playing a key role in stomatal function and photosynthesis. However, excessive chloride levels can be detrimental, potentially causing leaf burn or scorch and leading to chloride accumulation in leaf tissue (Libutti et al., 2019; Bayer and Njue, 2016; McCauley et al., 2019). Consequently, the reduction of Cl- following hydrogel application may enhance soil fertility and support plant health.

Shi et al. (2010) observed that the application of hydrogel in saline soils led to a reduction in Cl^- and Na^+ concentrations, while K^+ levels increased. Particularly, the Na^+ concentration in hydrogel-treated samples decreased by an average of 16% compared to untreated controls. This reduction in Na^+ concentration is beneficial for soil health and plant growth, as higher Na^+ levels can be detrimental. Plants typically experience stress when Na^+ concentrations exceed 230 mg L^{-1} or 70 mg L^{-1} in soil water (Foundation, 2007). Wei et al. (2019) demonstrated that a hydrogel composed of potassium acrylate, minerals, and acrylamide effectively improves soil conditions under saline stress. In addition, chitosan-modified BC (CMB) has been shown to significantly affect soil salinity by adsorbing Na^+. Hammer et al. (2015) found that BC application decreased Na^+ uptake in lettuce under salt stress and Kim et al. (2016) reported similar results in salt-stressed maize. BC not only improved root growth by enhancing soil properties but also reduced Na^+ uptake through the roots. In studies involving common beans, BC application significantly promoted root growth (Luo et al., 2017; Farhangi-Abriz and Torabian, 2017, 2018) and decreased Na^+ content in roots under salt stress (Farhangi-Abriz and Torabian, 2018). This reduction in Na^+ content is advantageous for root development, leading to improved plant growth and yield under saline conditions.

4.4. Biochemical traits

Stress antioxidants activities (non-enzymatic)

Lipid peroxidation was assessed based on the malondialdehyde (MDA) levels in plants subjected to salt stress. The highest MDA levels were observed in the salt treatment (5.38), followed by Salt + 1.5g SAH (4.12), which was statistically similar to the Salt + 0.5g, 2.5g, and 3.5g hydrogel treatments (Table 4.9). Lowest MDA level was measured in the control group. Additionally, plants showed a positive physiological response under salt stress conditions, with HGs support increasing proline production in all treatments. Notably, T3 showed a 396% increase in proline, statistically was higher than all other treatments, while T4 (237%), T5 (258%), and T6 (267%) also recorded significant increases compared to the control (Table 4.9). Moreover, chitosan modified BC reduced the salinity-induced increase in the Na+, glycine betaine (GB), proline, hydrogen peroxide (H2O2), and malondialdehyde (MDA) levels in plants (Mehmood et al., 2020). The chitosan modified BC increases the soluble protein content under salt-stress significantly (Mehmood et al., 2020).

Proline is a prevalent osmolyte synthesized by plants in response to salinity (Dar et al., 2016). This low-molecular-weight compound assists plants in managing osmotic stress (Jogawat, 2019). Proline and similar osmolytes are crucial for osmotic adjustment and function in guard cells by scavenging ROS (Pinheiro et al., 2004). Recently, Derbali et al. (2023) observed that amendments with BC and superabsorbent polymers (SAP) resulted in a notable reduction in MDA and proline levels under 300 mM NaCl salinity. Furthermore, these amendments significantly mitigated the increase in MDA and hydrogen peroxide induced by the NaCl treatment. The authors proposed that BC and SAP could potentially enhance or support the antioxidant system, thereby aiding in the regulation of ROS levels within plant tissues (Schutzendubel and Polle, 2002).

Table 4.9. Effect of superabsorbent hydrogel on stress antioxidant production of common bean under salt stress

Treatments	**Lipid peroxidation (µmol/g FW)**	**Proline (nmol Proline / g FW)**
Control	3.12 c	273.10 c
Salt	5.38 a	895.66 b
Salt + 0.5g SAH	3.22 bc	1356.24 a
Salt + 1.5g SAH	4.12 b	921.26 b
Salt + 2.5g SAH	3.78 bc	979.46 b
Salt + 3.5g SAH	3.25 bc	1004.10 b
LSD (0.05)	0.199	222.16
P-value	0.0035*	0.0001*

Stress antioxidants activities (enzymatic)

Antioxidant ascorbate peroxidase production was significantly improved in SAH applications such as T3, T4, and T5 were 4%, 418%, and 678%, respectively, more than T2 (salt) applications. Antioxidant superoxide dismutase (SOD) enzyme activity, which combats ROS play a vital role in stress conditions, was also found significant. T4 exhibited the highest enzyme activity at 538, followed by T3 at 507, T5 at 461 were 22%, 15%, 5%, respectively, more than salt (T2) group (Figure 4.3). The antioxidant activity induced by salinity was considerably enhanced by the application of chitosan-modified BC, which notably augmented the expression of four antioxidant enzyme genes (CAT, APX, POD, and SOD) (Mehmood et al., 2020). Moreover, in conditions of hyperosmotic salinity (300 mM NaCl), the activities of ROS defense enzymes such as SOD, APX, and guaiacol peroxidase (GPOX) were raised when BC and superabsorbent polymers were used (Derbali et al., 2023). In sandy soils, the application of hydrogel improved various enzymatic activities (Branch. 2009) including dehydrogenase, acid phosphatase, urease, alkaline phosphatase, and protease (Langaroodi et al., 2013; Islam et al., 2011).

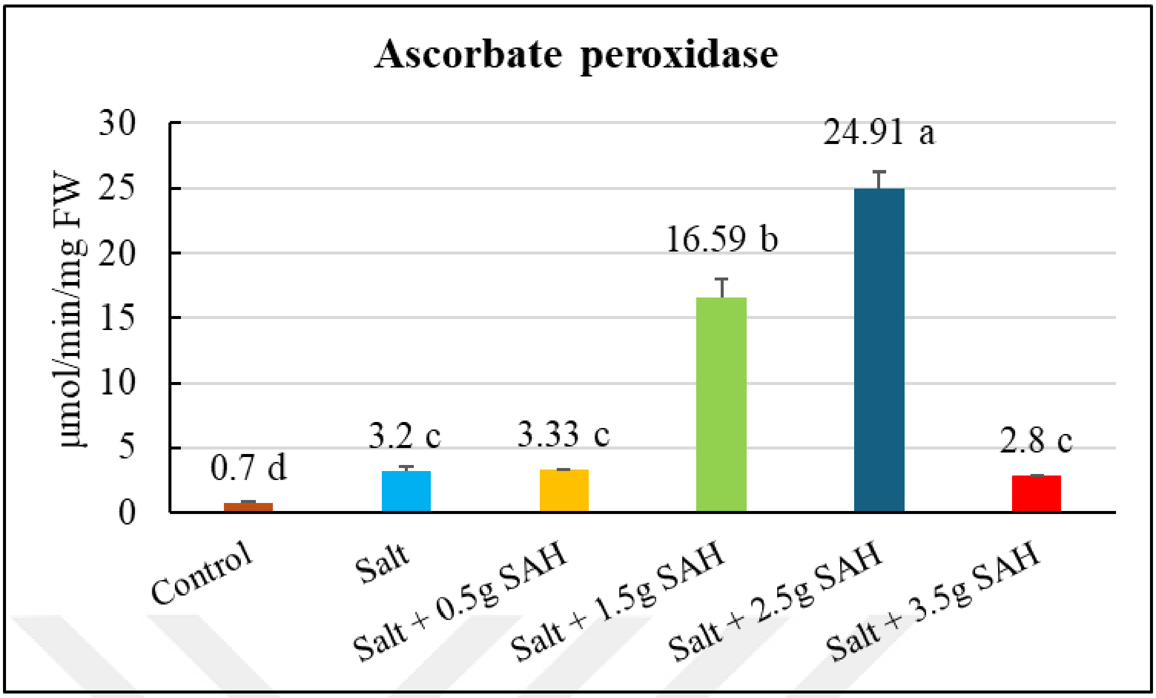

Figure 4.3. Effect of superabsorbent hydrogel on enzymatic activities of common bean under salt stress.

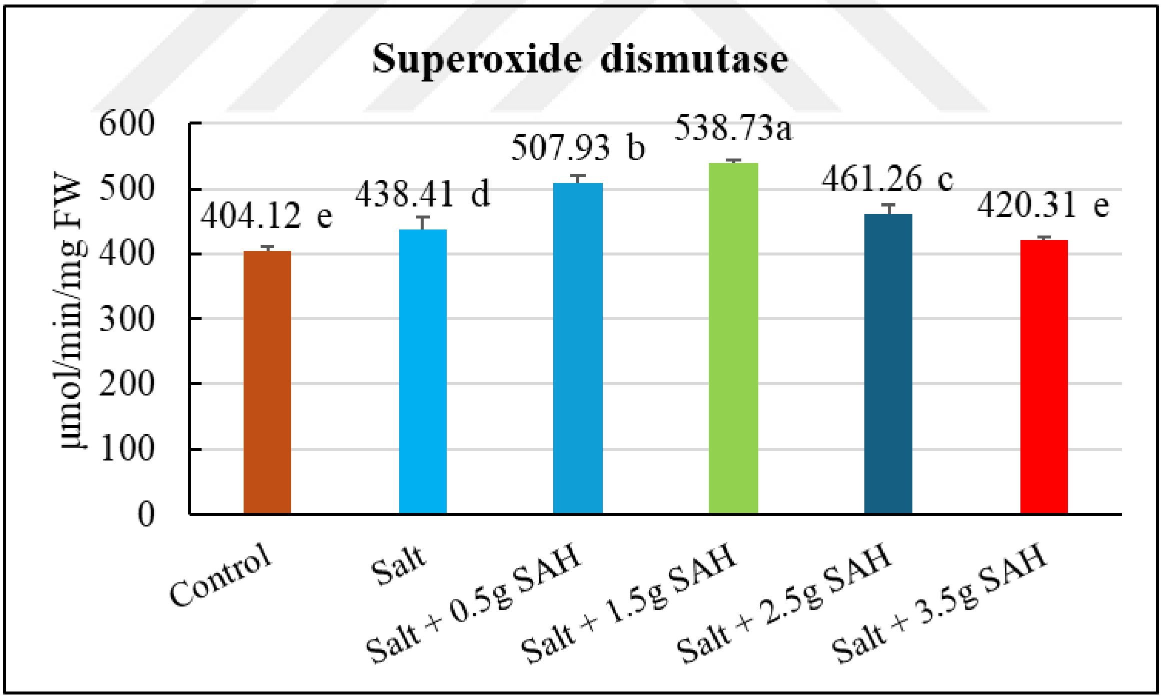

Figure 4.4. Effect of superabsorbent hydrogel on enzymatic activities of common bean under salt stress.

Sugar contents in leaves

Control and salt treatments have similar sugar content (1.35 b and 1.37 b, respectively), indicating that salt stress alone does not significantly alter sugar levels compared to the control. Adding SAH at 0.5g and 1.5g concentrations significantly increases sugar content to 1.78 and 1.64, respectively. These results suggest that low to moderate levels of SAH can enhance sugar accumulation in the presence of salt stress. Higher concentrations of SAH (2.5g and 3.5g) result in sugar content similar to or lower than the control (1.31 and 1.18, respectively) (Table 4.10), indicating a possible adverse effect or diminishing return on sugar accumulation at these higher SAH doses. The chitosan modified BC increases the sugar contents under salt-stress significantly (Mehmood et al., 2020). Superabsorbent hydrogels significantly support the plants to reduce the stress in plant, and in result less sugar production (Kenawy et al., 2022).

Table 4.10. Effect of superabsorbent hydrogel on total sugar content of common bean under salt stress.

Treatments	Total sugar content
Control	1.35 b
Salt	1.37 b
Salt + 0.5g SAH	1.78 a
Salt + 1.5g SAH	1.64 a
Salt + 2.5g SAH	1.31 b
Salt + 3.5g SAH	1.18 b
LSD (0.05)	0.243
P-value	0.0023*

Salt stress increased leaf temperature (25.00) compared to the control (23.26). The leaf temperature measured in common beans was statistically high in T2 (salt) application. Furthermore, the statistically measured same in all applications T1, T3-6. All SAH treatments reduced leaf temperature to levels similar to the control, with values ranging between 23.55°C and 23.95°C, indicating that SAH helps mitigate the heat stress caused by salt. Salt stress reduced chlorophyll content (40.76) compared to the control (43.36).

With increasing SAH concentrations, SPAD values further declined, from 38.30 (0.5g SAH) to 34.56 (3.5g SAH) (Table 4.11). This shows that although SAH treatments lower the chlorophyll content, they remain statistically different from both control and salt treatments. The decrease in chlorophyll levels in salt stressed plants has been considered as a typical symptom of oxidative stress and was attributed to the inhibition of chlorophyll synthesis, together with the activation of its degradation by the enzyme chlorophyllase (Santos. 2004

The application of BC and superabsorbent polymers markedly enhances chlorophyll content in plants subjected to 300 mM salinity stress (Derbali et al., 2023). The reduction in chlorophyll content under 300 mM NaCl conditions has been attributed to increased chlorophyllase enzyme activity and limited nitrogen uptake (Reddy and Vora, 1986; Van et al., 2001).

Table 4.11. Effect of superabsorbent hydrogel on leaf temperature of common bean under salt stress.

Treatments	Leaf temperature (^{O}C)	SPAD
Control	23.26 b	43.36 a
Salt	25.00 a	40.76 b
Salt + 0.5g SAH	23.95 b	38.30 c
Salt + 1.5g SAH	23.55 b	37.80 c
Salt + 2.5g SAH	23.90 b	36.36 cd
Salt + 3.5g SAH	23.93 b	34.56 d
LSD (0.05)	0.773	2.224
P-value	0.0094*	0.0001*

The highest number of fruits was statistically observed in the T1 treatment. T2, T3, T5, and T6 treatments showed statistically similar results, while the lowest fruit count was noted in the T4 (salt+ 3SAH) treatment. The fruit weight was statistically highest in T1 and similar across T2 to T6 treatments (Table 4.12). Jalilian and Mohsennia (2013) found that applying hydrogel at a rate of 200 kg/ha significantly improved yield and various agronomic traits in peanuts grown in sandy soil under hot and arid conditions.

Table 4.12. Effect of superabsorbent hydrogels on some fruit traits of common bean.

Treatments	No. of fruits/plant	Fruit weight (g/ plant)
Control	16.00 a	76,99 a
Salt	9.00 b	34.22 b
Salt + 0.5g SAH	8.66 b	38.95 b
Salt + 1.5g SAH	7.00 c	32.65 b
Salt + 2.5g SAH	9.00 b	32.99 b
Salt + 3.5g SAH	9.33 b	33.58 b
LSD (0.05)	1.627	6.560
P-value	0.0001*	0.0001*

4.5. Fruit quality traits

The data shows that TSS (% plant) increases significantly in the SAH treatments compared to the control, with the highest values in the Salt + 2.5g SAH (9.43%) and Salt + 3.5g SAH (9.16%) treatments, compared to 6.66% in the control. Titrable acidity is highest in the control (0.97) and significantly decreases in SAH treatments, especially with 2.5g+SAH (0.14) and highest is with 0.5g+SAH (0.40). The pH also increases notably with SAH, reaching 8.08 in the 2.5g SAH treatment, compared to 6.43 in the control. Lastly, EC (dS m^{-1}) shows a varied response, with the highest value (7.63) in Salt + 1.5g SAH, while the control has a moderate EC of 6.61 (Figure 4.4). Overall, the SAH treatments improve TSS and reduce acidity compared to both control and salt-alone treatments.

The total phenol content significantly increased due to SAH treatments, with values of 53.9%, 52.5%, and 40.7% observed in T6, T5, and T4 applications, respectively, compared to the control. Additionally, the highest flavonoid content was recorded in the T5, T4, and T6 applications, with values of 145.60, 131.15, and 112.93 mg RU/100g. This represents an increase of 138%, 114%, and 84% in T5, T4, and T6 applications, respectively, compared to the control. Vitamin C content was statistically highest in the T6 application, showing a 27% increase over the control. Additionally, Increases were also measured in T5 (24%), T4 (18%), T2 (13%), and T3 (3%) applications when compared to the control (Figure 4.5).

It was reported that the HGs played a significant role in flavonoid production under abiotic conditions (Abd El-Aziz et al., 2022) Ascorbic acid, total soluble solids (TSS), pH, lycopene and juice % increased by using of eco-friendly hydrogel. An increase in ascorbic acid content was recorded in tomatoes due to the application of hydrophilic polymer (Sendur-Kumaran. 2016). Increase pH value in tomato due to use of hydrophilic polymer was noticed. These metabolites share in the original composition of TSS (Sendur-Kumaran. 2016). Lowering in the pH value of fruits under drought stress was because of a change in the acid content (Anthon et al., 2011). Abd El-Badea et al. (2011) reported that, the application of soil conditioner veterra hydrogel (VH) and irrigation every 26 days with 150 kg N fed^{-1} positively affected on potato tuber quality attributes like total dry matter (22.71 to 22.83%), starch content (14.78 to 15.70%).

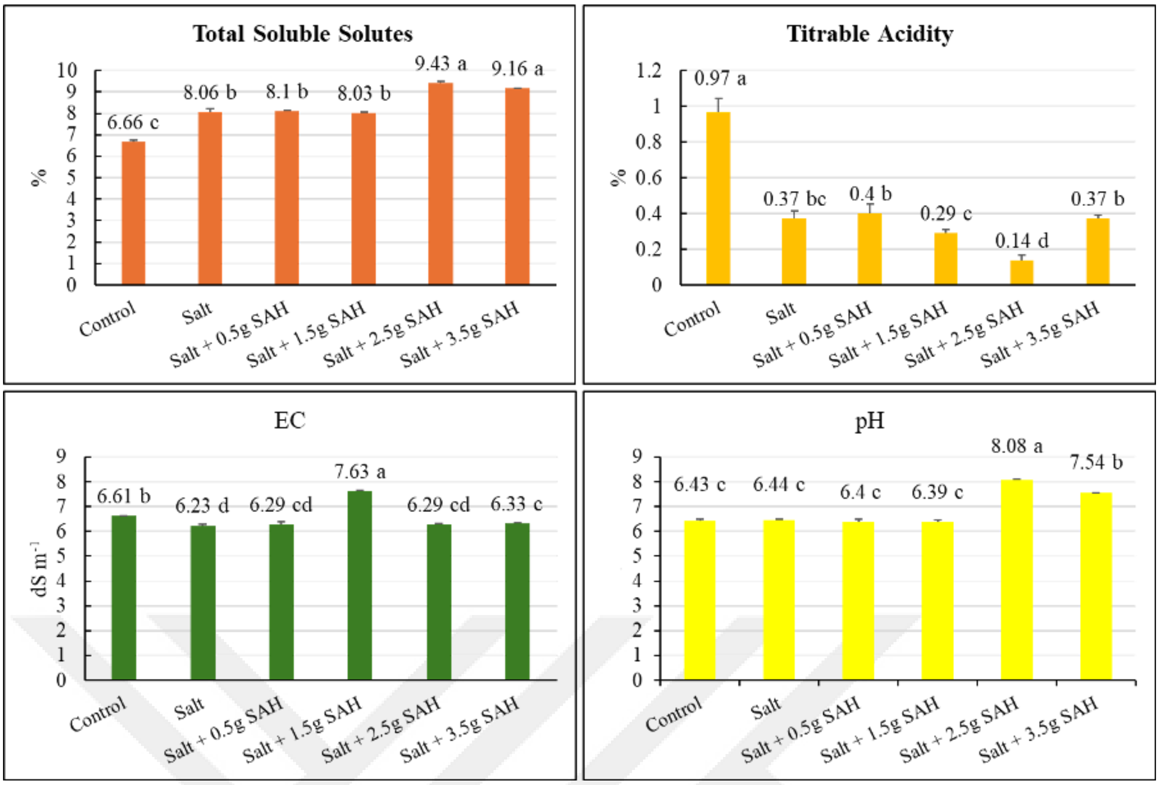

Figure 4.5. Effect of superabsorbent hydrogel on fruit quality traits of common bean under salt stress.

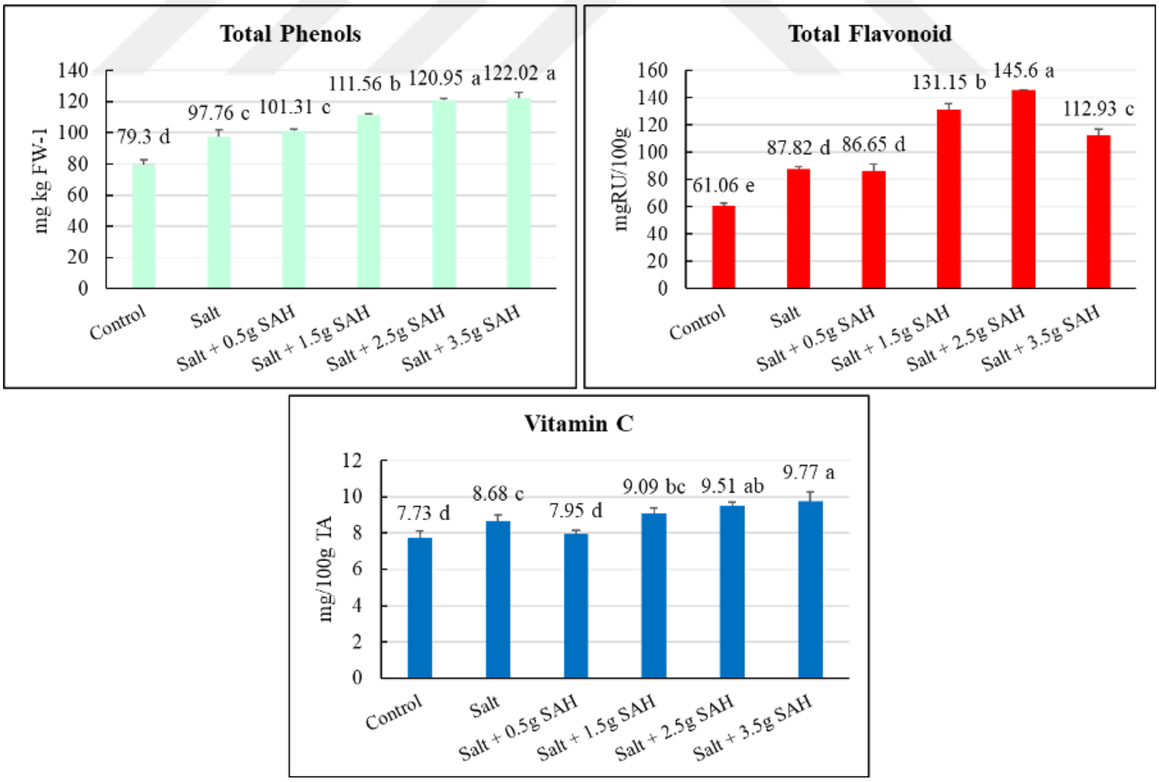

Figure 4.6. Effect of superabsorbent hydrogel on fruit quality traits of common bean under salt stress.

Drained water analysis

EC was lower in most of the applied treatments of SAH, compared to salt. pH values were lower in most of SAH treatments compared to control. however, T3 showed no variation in pH.

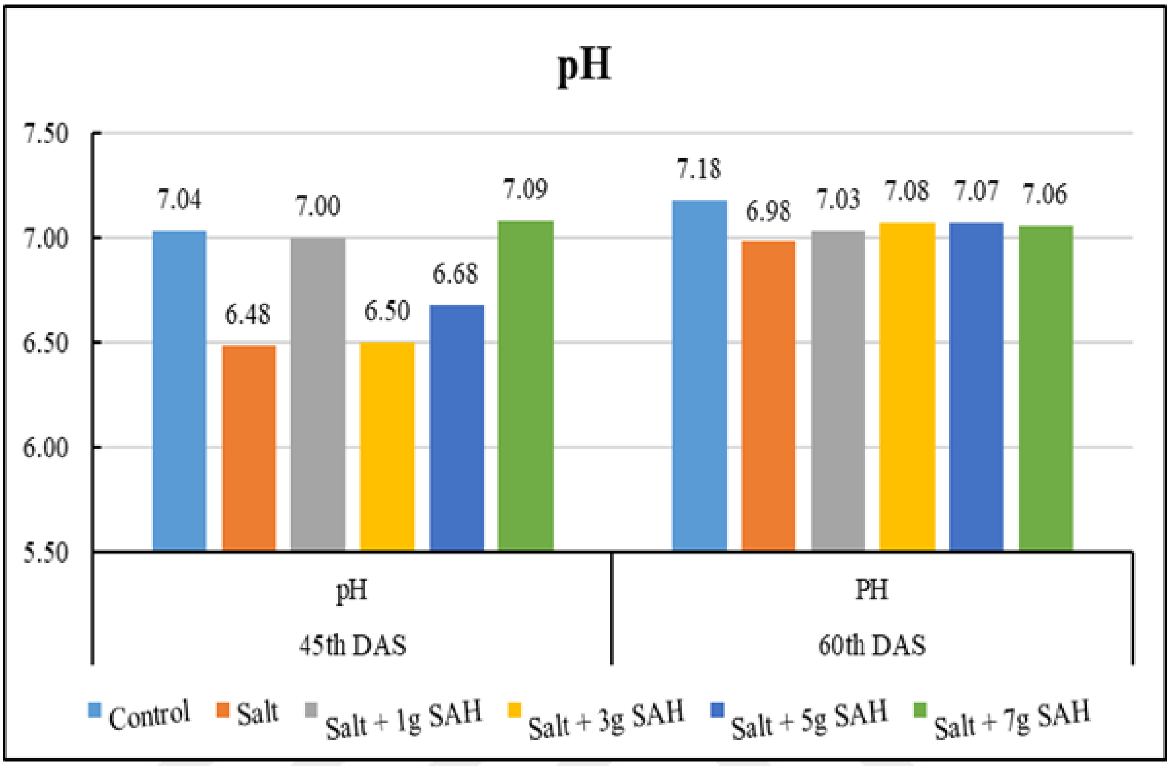

Figure 4.7. Drained water pH.

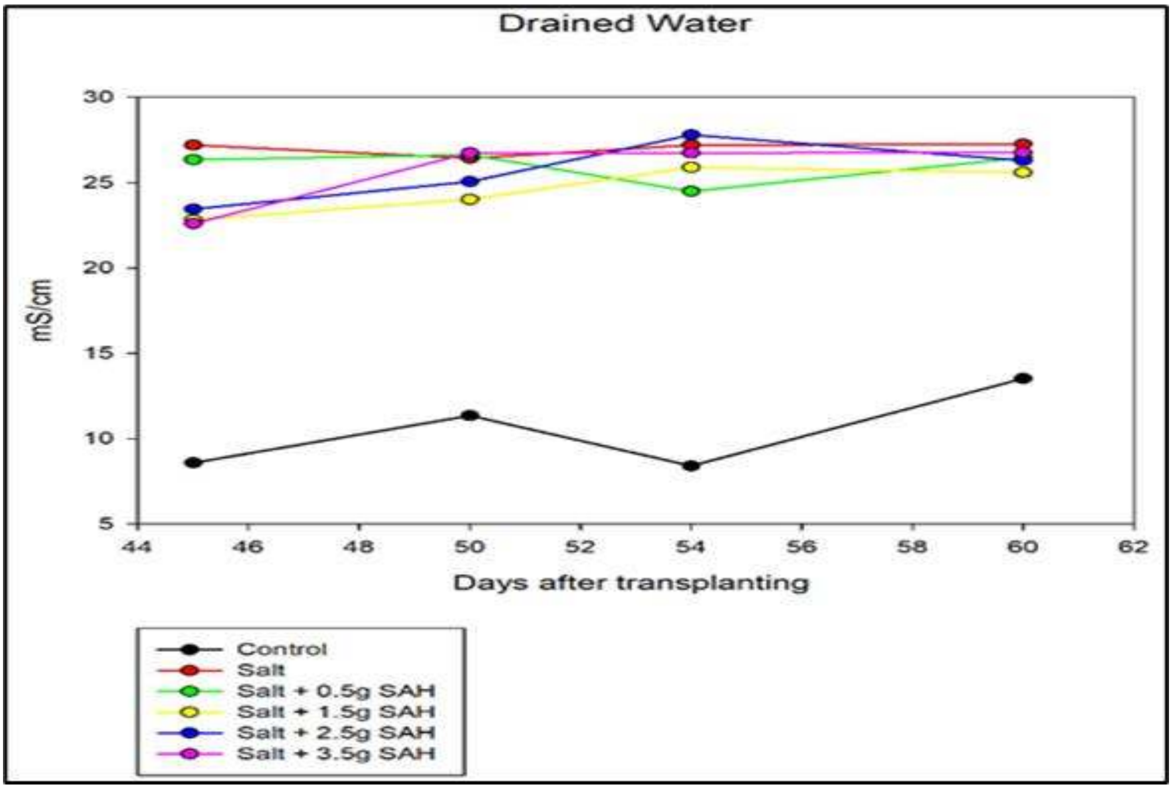

Figure 4.8. Drained water EC.

5. CONCLUSIONS

Salinity poses a major challenge to agricultural production globally, adversely affecting the growth, development, and yield of crops. In this study, salt stress significantly impaired the morphological, physiological, biochemical, and fruit traits of common bean plants. However, the application of a superabsorbent hydrogel (SAH), composed of alginate-based hydrogel and biochar derived from fruit peels, effectively alleviated these negative effects. Notably, the use of 0.5 g SAH showed considerable improvement in plant morphology and physiology under salt stress conditions. While increasing SAH doses resulted in sodium accumulation in the bean roots, which correlated with a reduced mitigation of salt damage at higher doses, the application of SAH positively influenced the plant's water status and key biochemical parameters, such as enzyme activity, proline content, and MDA levels. These findings suggest that SAH can enhance the plant's resilience to salt stress. Future studies should explore additional physiological and biochemical parameters in the roots to further understand the potential of SAH in mitigating salt stress in bean crops.

Some recommendations and future directions

1. **Optimization of SAH application**: Further research should focus on optimizing the concentration and application frequency of SAH to determine the most effective dosage and method for various soil and environmental conditions.
2. **Long-term studies:** Conduct long-term field studies to evaluate the sustained effectiveness of SAH under different climatic conditions and its impact on long-term soil health and productivity.
3. **Mechanistic understanding**: Investigate the underlying mechanisms by which SAH interacts with plant physiological processes and soil properties to better understand its role in mitigating salt stress.
4. **Comparison with other amendments**: Compare the efficacy of SAH with other soil amendments and stress mitigation strategies to establish a broader context for its application in sustainable agriculture.
5. **Scalability and Economic Feasibility**: Assess the scalability and economic feasibility of SAH application for large-scale agricultural production to ensure its practical viability for farmers.
6. **Diverse crop studies**: Extend research to other crops to evaluate the generalizability of SAH's benefits across different plant species and stress conditions.

By addressing these areas, future research can enhance the practical application of SAH and contribute to developing more resilient agricultural practices in the face of increasing salinity and other environmental stresses.

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CURRICULUM VITAE

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PUBLICATIONS

Dasgan, H. Y., Yilmaz, D., Zikaria, K., Ikiz, B., & Gruda, N. S. (2023). Enhancing the yield, quality and antioxidant content of lettuce through innovative and eco-friendly biofertilizer practices in hydroponics. *Horticulturae*, *9*(12), 1274.

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