



Article Growth and Yield Response and Water Use Efficiency of Cotton under Film-Mulched Drip Irrigation to Magnetized Ionized Water and *Bacillus subtilis* in Saline Soil in Xinjiang

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Abstract: In irrigated agriculture, the combination of multiple regulation measures is an effective method to improve saline soil and promote crop growth. Magnetized ionized water irrigation is considered a promising irrigation water activation technique, while the use of Bacillus subtilis for soil amelioration is environmentally friendly. In this study, magnetized ionized water irrigation and B. subtilis were used to promote cotton growth under film-mulched drip irrigation (FMDI) in saline soil. A two-year field experiment was conducted to investigate the effects of differing B. subtilis amounts under two irrigation water types (non-magnetized-ionized water (NMIW) and magnetized ionized water (MIW)) on the growth (plant height, leaf area index, shoot dry matter and chlorophyll content) and the yield of cotton, as well as the soil water content, salts accumulation, water use efficiency (WUE) and irrigation water use efficiency (IWUE) under FMDI in a saline soil in southern Xinjiang. Five amounts of *B. subtilis* (0, 15, 30, 45 and 60 kg ha⁻¹) under NMIW (designated as B0, B1, B2, B3 and B4) and MIW (designated as M, MB1, MB2, MB3 and MB4) were applied to the field experiments. The results showed that MIW and B. subtilis increased soil water content and reduced salts accumulation in the 0-40 cm soil layers compared with B0. Moreover, the two measures significantly (p < 0.01) increased cotton plant height, leaf area index, shoot dry matter and chlorophyll content compared with B0. Seed cotton yield, WUE and IWUE were also observed to significantly increase (p < 0.05). Compared with the NMIW treatments, the MIW treatments increased seed cotton yield by 2.1–12.2%, increased WUE by 0.2–9.0%, and increased IWUE by 2.1–12.2%. Under MIW, with the B. subtilis amount as an independent variable, quadratic function relationships with seed cotton yield, WUE and IWUE were established. By taking the first derivative of the quadratic function, the highest seed cotton yield, WUE and IWUE were obtained with the B. subtilis amounts of 51.8, 55.0 and $51.4 \text{ kg} \text{ ha}^{-1}$, respectively. Based on comprehensive consideration of seed cotton yield, WUE, IWUE and salts accumulation in soil, 51.4 kg ha⁻¹ of *B. subtilis* under MIW treatment is recommended for cotton cultivated under FMDI in a saline soil of southern Xinjiang, China.

Keywords: magnetized ionized water; *Bacillus subtilis*; yield; water use efficiency; saline soil; salts accumulation

1. Introduction

Soil salinization is an important factor restricting agricultural production in arid and semi-arid regions. There are approximately 11 million ha of saline soil worldwide, and the total area is increasing at a rate of 10% each year [1]. Moreover, projections estimate that more than 50% of arable land will be salinized by 2050 [2]. Thus, urgent solutions are needed to enhance crop yield and water use efficiency (WUE) in response to the growing threat of soil salinization worldwide [3]. The technology of film-mulched drip irrigation (FMDI) has been proven to save water and promote leaching of salts in cotton crops cultivated in northwest China [4–6]. Xinjiang, located in the northwest of China, has a vast distribution of saline soil, with salinized cultivated land covering 2.33 million ha,



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Copyright: © 2023 by the authors. Licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (https:// creativecommons.org/licenses/by/ 4.0/). accounting for 38% of the cultivated land and causing crop yield losses of over 15% [7]. As a strongly salt tolerant crop and an important economic crop, cotton is widely planted in Xinjiang. In 2022, nearly 2.5×10^6 ha of cotton was planted in Xinjiang, accounting for approximately 83.2% of the planting area and 90.2% of the cotton yield in China (http://www.stats.gov.cn/sj/zxfb/202302/t20230203_1901689.html (accessed on 30 May 2023)). However, cotton cultivation faces numerous soil-related problems, such as the inadequate leaching of salts and poor soil water holding capacity in the root zone [8]. Thus, determining how to improve saline soil with environmentally friendly techniques whilst maintaining high efficiency is key to ensuring the normal growth and high yield and WUE of cotton in Xinjiang.

In order to improve the leaching of salts and increase soil water content, activated water technology for agricultural irrigation, including magnetized ionized water (MIW), has attracted an increasing amount of attention as it is an environmentally friendly and highly efficient technique [9,10]. In particular, under MIW, the magnetic field increases the average distance between water molecules and weakens (or even breaks) some hydrogen bonds, which reduces the size of the large associated water molecular clusters [11,12]; after the treatment of irrigation water by ionized technology, the salt ions in the water remain positively charged. The positive ions repel each other, preventing the binding of the water and salts [13]. Magnetized technology and ionized technology for irrigation reduce the surface tension and viscosity coefficients of water and increase the diffusion coefficient, thus effectively enhancing the diffusion ability and permeability of water and leaching of salts in saline soil areas [14-19]. Moreover, magnetized treatment or ionized treatment is also beneficial for the planting, physiology and yield of crops in saline soil [20]. Meysam et al. [21] reported that the growth parameters (i.e., germination rate, seedling height and weight, etc.) of maize and soil salinity leaching improved under magnetic water treatment compared with no treatment. Alkassab et al. [22] also found that magnetized water irrigation was associated with a statistically significant increase in plant height and leaf area. Wei et al. [13] reported improvements in plant height, leaf area index and shoot dry matter resulting from the ionized treatment. Numerous scholars have also demonstrated that the application of magnetized treatment or ionized treatment for irrigation can enhance crop yield and WUE [13,23–25]. Water serves as the medium, solvent, or catalyst facilitating the transportation of various substances within soil [20]. Under MIW, the physicochemical properties of irrigation water undergo a transformation, leading to increased efficiency of soil substance transportation, such as soil water diffusion ability and leaching of salts, which has positive effects on the growth characteristics of crop. Such enhancements establish the groundwork for the effective remediation of saline soil with other improvement measures.

Plant growth promoting rhizobacteria have attracted much attention as a non-polluting and effective approach to improve saline soil. Compared with other saline soil amendment methods such as gypsum and biochar, using plant growth promoting rhizobacteria to improve saline soil is considered to be a safe and environmentally friendly measure [26]. The bacteria *B. subtilis* can effectively fulfill its function under long-term stress conditions due to the formation of long-living and stress-tolerant spores [27]. During the growth and metabolism processes, *B. subtilis* can induce plants to produce plant hormones such as Indole acetic acid (IAA) to promote plant growth [28,29] and increase crop yield [30–33]. Moreover, *B. subtilis* can improve the soil structure by secreting extracellular polymeric substances, which can form hydrogen bonds with soil water to keep more water in the soil [34]. *B. subtilis* can reduce soil particles together to form stable large aggregates [35,36]. Using *B. subtilis* can reduce soil bulk density and increase soil porosity, which consequently enhances soil permeability, water infiltration and leaching of salts [37].

Despite the proven beneficial effects of using either MIW or *B. subtilis* on plant growth and soil, the extent of these effects and their underlying mechanisms remain unclear. Furthermore, research on the coupling effects of MIW and *B. subtilis* on plant growth, soil water content, salts accumulation, WUE and IWUE in the field is limited. The current study implemented water conservation and biological measures to improve the two most

important components of cotton growth, water and soil [38], in order to enhance cotton growth in saline soil. The aims of the study are to: (i) examine the performances of MIW and *B. subtilis* on cotton growth and yield; (ii) investigate the effects of MIW and *B. subtilis* on soil water content and salts accumulation; and (iii) determine the optimal applied amount of *B. subtilis* to maximize seed cotton yield and WUE and IWUE under MIW treatment in a saline soil of southern Xinjiang, China.

2. Materials and Methods

2.1. Experimental Site

A two-year field experiment was conducted during 2021 and 2022 at the seventh company of the eighth regiment within the first division of Xinjiang Production and Construction Corps (N 40°37'32.26", E 80°52'57.78") in Alar City, Xinjiang Province, northwest China. Alar is in a warm temperate zone with an extreme continental arid desert climate and large temperature differences between day and night. This region is rich in light resources, with an annual average solar radiation of 133.7–146.3 kcal cm⁻² and an annual average 2556.3–2991.8 h of sunshine. The city has a 207 d frost-free period and an average annual potential evaporation of 1876.6-2558.9 mm (http://www.ale.gov.cn/ssgk/qhtj (accessed on 30 May 2023)). In 2021 and 2022, the daily average air temperature during the cotton growing period (April to September) was 20.9 and 21.0 °C, with a total precipitation of 40.4 and 35.0 mm, respectively (Figure 1). Table 1 reports the physical and chemical properties of a 0-100 cm soil profile in the region. The soil profile (0-40 cm) average electrical conductivity of 1:5 extracts for soil and water (by weight) was 3.5 dS m^{-1} ; the exchangeable sodium percentage (ESP) was 6.3% before sowing. Thus, the soil saline class of the experimental site was slightly to moderately saline [39]. The average values of soil properties for 0–100 cm were determined before sowing: bulk density ($\overline{\rho}$) of 1.5 g cm⁻³; pH of 7.9 of 1:5 extracts for soil and water (by weight); total organic matter of 10.5 g kg^{-1} ; alkali-hydrolyzable nitrogen of 78.8 mg kg⁻¹; available phosphorus of 32.0 mg kg⁻¹; and available potassium of 144.1 mg kg⁻¹. The groundwater depth was below 5 m, and the salinity of canal water used for irrigation was 0.4 g L^{-1} .



Figure 1. Daily air temperature and precipitation during the cotton growing seasons in 2021 and 2022.

2.2. Magnetized Ionized System for Irrigation Water Treatment

The canal water originated from meltwater in the Tianshan Mountains. After being filtered and pressurized in the pump room, it entered the field through buried pipelines. The outlet of the buried pipeline in the field was a 90 mm PVC pipe. A magnetized ionized device was connected to the 90 mm water pipeline. The magnetized ionized system (Xi'an Wangkaiyue Metal Products Co., LTD) consisted of a magnetized ionized device, ground electrode and conductor (Figure 2). The magnetizer uses a permanent magnet ring with a

magnetic field intensity of 3000 Gs. The grounding resistance of the experimental device was 5 Ω and the ground bolt was connected to the ground electrode via a wire. Under the action of the magnetic field, the average distance between water molecules increases, and some hydrogen bonds weaken or even break. With the application of the ionized technology, as water passes through the system, it is enriched with electrons, and these electrons are then channeled underground through grounded wires [40,41].

Soil Layer	Particle Composition			Soil	Soil Bulk Tensity		Soil Water Content (cm ³ cm ⁻³)			
(cm) -	Clay (%)	Silt (%)	Sand (%)	lexture	$(g cm^{-3})$	$\theta_{\rm WP}$ $\theta_{\rm FC}$	$\theta_{\rm S}$			
0–20	12.18	79.11	8.60	Silty loam	1.46	0.042	0.31	0.41		
20-40	12.66	81.93	5.42	Silty loam	1.52	0.042	0.33	0.41		
40-60	11.45	85.44	3.10	Silty loam	1.54	0.045	0.32	0.41		
60-80	11.94	84.30	3.75	Silty loam	1.53	0.046	0.33	0.42		
80-100	8.68	81.65	9.67	Šilty	1.55	0.060	0.34	0.40		

Table 1. Physical and chemical properties of soil at the experimental site.

 θ_{WP} is the permanent wilting point; θ_{FC} is the field capacity; and θ_s is the saturated soil water content.



Figure 2. Magnetized ionized system of the experiments.

The use of canal water for irrigation directly without passing through the magnetized ionized system is referred to as non-magnetized-ionized water (NMIW), and canal water passing through the magnetized ionized system is referred to as magnetized ionized water (MIW) (Figure 3). Magnetization and ionization altered the water surface tension from 66.8 to 64.0 mN m⁻¹ in this study.

2.3. Experimental Design and Treatments

Cotton (*Gossypium hirsutum* L. Tahe No.2) was sown on 9 April 2021 and 10 April 2022; and harvested on 10 October 2021 and 30 September 2022. The planting density was 1.80×10^5 plants per hectare, with a plant and row spacing of 10 cm. The cotton was grown under drip irrigation with a plastic film-mulched system (Figures 4 and 5). The drip system was installed with a row configuration of 10 cm + 60 cm + 10 cm + 60 cm + 10 cm (outside narrow, wide, inside narrow, wide, outside narrow). Two driplines were

installed per six rows under a 1.5 m wide film. The distance between two films was 50 cm. Irrigation was performed using a 1.6 cm diameter drip line. The average emitter spacing was 30 cm, and the discharge rate for each drip emitter was 2.0 L h⁻¹. Table 2 reports the cotton developmental stages in 2021 and 2022. Figure 6 reports the irrigation schedule of the cotton under FMDI in this study. The first irrigations during the cotton growth stage in the cotton field were performed on 12 June 2021 and 13 June 2022. The irrigation amounts recorded by a water meter were 501.60 and 518.85 mm in 2021 and 2022, respectively. Urea (N \geq 46%) and potassium dihydrogen phosphate (KH₂PO₄ \geq 99.5%) fertilizers were applied eight times during the cotton growth stage. Differential pressure fertilizer barrels with a 15 L capacity were used to apply the fertilizers. One barrel was placed in each experimental plot.



Figure 3. Schematic diagram of the drip irrigation system arrangement of the experiments.



Figure 4. Positions of row spacing, driplines, plastic film and sampling points.

Table 2. Cotton developmental stages in 2021 and 2022.

Cotton Crowth Stagos	Year					
Cotton Growin Stages —	2021	2022				
Sowing	9 April	10 April				
Emergence	20 April	22 April				
Budding	12 June	10 June				
Flowering	7 July	3 July				
Boll	6 August	6 August				
Boll-opening	7 September	5 September				
Harvest	10 October	30 September				



Figure 5. Experimental site.



Figure 6. Irrigation schedules in 2021 and 2022.

We considered the effects of the *B. subtilis* amounts under two types of irrigation water (NMIW and MIW) on the soil water, salts distribution, cotton growth and yield, and water use efficiency. The five amounts of *B. subtilis* (0, 15, 30, 45 and 60 kg ha⁻¹) under NMIW were denoted as B0, B1, B2, B3 and B4, and as M, MB1, MB2, MB3 and MB4 under MIW (Table 3). The experiment treatments of B0, M, MB1, MB2, MB3 and MB4 were conducted in 2021 and all the 10 treatments were conducted in 2022. Each experimental plot was 2 m wide and 30 m long. All the treatments were replicated three times in a randomized block design. The wettable powder of *B. subtilis* with 20 billion live spores per gram was produced by Shandong Lvlong Biotechnology Co., Ltd. (Weifang City, Shandong Province, China). *B. subtilis* was applied to the soil through the 15 L capacity pressure fertilizer barrels at the first cotton irrigation event (12 June 2021 and 13 June 2022).

Treatment	Irrigation Water Type and B. subtilis Amounts	Treatment	Irrigation Water Type and B. subtilis Amounts
B0	NMIW + 0 t ha ^{-1} B. subtilis	Μ	MIW + 0 t ha ^{-1} B. subtilis
B1	NMIW + 15 t ha ^{-1} B. subtilis	MB1	MIW + 15 t ha ^{-1} B. subtilis
B2	NMIW + 30 t ha ^{-1} B. subtilis	MB2	MIW + 30 t ha ^{-1} B. subtilis
B3	NMIW + 45 t ha ^{-1} B. subtilis	MB3	MIW + 45 t ha ^{-1} B. subtilis
B4	NMIW + 60 t ha ^{-1} B. subtilis	MB4	MIW + 60 t ha ^{-1} B. subtilis

Table 3. Experimental design and treatments.

2.4. Measurements and Calculations

2.4.1. Cotton Height (H) and Leaf Area Index (LAI)

Cotton height and leaf area were measured every seven days after the first irrigation. For the determination of plant height and leaf area, representative plants with uniform growth potential were selected as the research object in each experimental plot. Each experimental plot was marked with three plants in the inside narrow row and three plants in the outside narrow row. The length and width of each leaf on the plants were determined with a tape measure to calculate the leaf area [42]. The green leaf area of the plant was calculated as follows:

$$LA = \sum_{i}^{n} Ai \times Bi \times 0.703$$
 (1)

where LA is the leaf area of per plant (cm²); A_i (cm) and B_i (cm) are the length and width of cotton leaf *i*, respectively; *n* is the total number of leaves per cotton plant; and 0.703 is the correction factor for cotton [43]. The LAI was then determined as follows [44]:

$$LAI = \frac{LA}{S_0}$$
(2)

where S_0 is the occupied land area (cm²).

2.4.2. Modified Logistic Growth Model

To quantify the change process of cotton plant height (H) and leaf area index (LAI) with time under different treatments, a modified logistic model was used to describe the quantitative relationship between H and the day (t, d) after the first irrigation during the cotton growth stage (Equation (3)), as well as the quantitative relationship between LAI and t (Equation (4)):

$$H = \frac{H_m}{1 + e^{a_1 + b_1 t}}$$
(3)

$$LAI = \frac{LAI_m}{1 + e^{a_2 + b_2 t + ct^2}}$$

$$\tag{4}$$

where H_m and LAI_m are the theoretically achievable maximum plant height (cm) and the theoretically achievable maximum LAI of the cotton, respectively; and a_1 , b_1 , a_2 , b_2 and c are the shape parameters in the modified logistic model.

2.4.3. Shoot Dry Matter

At the budding stage (1 June 2021 and 1 July 2022) and boll stage (30 August 2021 and 30 August 2022), six plants (three inside narrow and three outside narrow) were randomly selected from each plot. The leaves, stem, flowers and bolls of the plants were placed in an oven at 105 °C for 30 min, dried at 75 °C to a constant weight, and then subsequently weighed. The leaves and stem were considered as the nutritive organs; and the buds, flowers and bolls as the reproductive organs. The proportion of the cotton nutritive organs

dry matter (NOP) and reproductive organs dry matter (ROP) to the total shoot dry matter (nutritive organs and reproductive organs) was calculated as follows:

$$NOP = \frac{Nutritive \text{ organs dry matter}}{Total \text{ shoot dry matter}}$$
(5)

$$ROP = \frac{\text{Reproductive organs dry matter}}{\text{Total shoot dry matter}}$$
(6)

where NOP is the proportion of cotton nutritive organs to the total shoot dry matter (%); and ROP is the proportion of cotton reproductive organs to the total shoot dry matter (%).

2.4.4. Relative Chlorophyll Content

The SPAD value represents the leaf greenness or the photosynthetic pigment content [45]. SPAD values were measured at the flag leaves with a SPAD 502 Plus system (Konica Minolta, Japan) between 11:00 a.m. and 13:00 p.m. (GMT + 8) on sunny days. Measurements were performed every seven days after the first irrigation. The average values of 20 randomly selected leaves and the fourth leaf from the top leaves were determined.

2.4.5. Soil Water Content and Salinity

Soil samples were collected to determine soil water content and salinity at 10 cm intervals from 0 to 40 cm and at 20 cm intervals from 40 to 100 cm using an auger with a 5 cm diameter in the middle of the wide, narrow, and bared strips (Figure 4). The samples were collected at the late main growth stages for cotton one day prior to irrigation and harvest. All the auger holes were refilled with soil to minimize the experimental error after each of the samples was collected. The soil samples were weighed, dried in a fan assisted oven at 105 °C for 24 h, and reweighed to determine the gravimetric soil water content. The volumetric soil water content was then obtained by multiplying the gravimetric soil water content by the corresponding soil bulk density. A DDSJ-308A conductivity meter (Shanghai Yidian Scientific Instrument Co., Ltd., Shanghai, China) was used to measure the electrical conductivity of a 1:5 soil water extract (EC_{1:5}) under 25°C. The value of EC_{1:5} (dS m⁻¹) for each soil sample can be converted into salts content in soil (SC, g kg⁻¹) based on a linear relationship (SC = $0.99 \times EC_{1:5}$, R² = 0.9992, n = 30).

2.4.6. Crop Evapotranspiration Calculations

Crop evapotranspiration (ET_c , mm) was determined via the water balance equation as follows [46–49]:

$$ET_c = \Delta W + P + I - D \tag{7}$$

where ΔW is the change of soil water storage in the 0–100 cm soil layer from the pre-sowing to post-harvesting of cotton (mm); *P* is the precipitation during the growing period (mm); *I* is the irrigation amount (mm); and *D* is the drainage amount (mm), respectively. By experiment and calculation [3], *D* was negligible in this study.

 ΔW was calculated as:

$$\Delta W = 1000 \left(\frac{1}{3} \Delta \theta_{\text{inside narrow}} + \frac{4}{21} \Delta \theta_{\text{wide}} + \frac{1}{3} \Delta \theta_{\text{outside narrow}} + \frac{1}{7} \Delta \theta_{\text{bare}} \right)$$
(8)

where $\Delta \theta_{\text{inside narrow}}$, $\Delta \theta_{\text{wide}}$, $\Delta \theta_{\text{outside narrow}}$ and $\Delta \theta_{\text{bare}}$ are the differences in the volumetric soil water content for the inside narrow, wide, outside narrow and bare soil strips, respectively, from the pre-sowing to post-harvesting of cotton in the 0–100 cm soil profile (cm³ cm⁻³). Weights of 1/3, 4/21, 1/3 and 1/7 were assigned as proportions of the strip widths at different locations.

2.4.7. Water Use Efficiency (WUE) and Irrigation Water Use Efficiency (IWUE) WUE and IWUE were calculated as follows [50]:

$$WUE = \frac{Y}{ET_c}$$
(9)

$$IWUE = \frac{Y}{I}$$
(10)

where Y is the seed cotton yield (kg ha⁻¹).

2.4.8. Salts Accumulation Calculation

Salts accumulation in soil was calculated as the difference between the content of salinity from the pre-sowing to post-harvesting of cotton. The change in soil salinity storage in the 0–100 cm soil layer (ΔS , g m⁻²) was calculated as:

$$\Delta = 10\overline{\rho} \left(\frac{1}{3} \Delta SC_{\text{inside narrow}} + \frac{4}{21} \Delta SC_{\text{wide}} + \frac{1}{3} \Delta SC_{\text{outside narrow}} + \frac{1}{7} \Delta SC_{\text{bare}} \right)$$
(11)

where ΔS denotes the changes in soil salinity from the pre-sowing to post-harvesting of cotton; and $\Delta SC_{\text{inside narrow}}$, ΔSC_{wide} , $\Delta SC_{\text{outside narrow}}$ and ΔSC_{bare} are the changes in volumetric soil salinity for the inside narrow, wide, outside narrow and bare soil strips, respectively, from the pre-sowing to post-harvesting of cotton (g m⁻²). A positive value of ΔS denotes the accumulation of salts in the soil, while a negative value indicates the occurrence of desalination.

2.4.9. Data Analysis

SAS 9.2 statistical software (SAS Institute Inc., Cary, NC, USA) was employed to perform the analysis of variance (ANOVA). All indicators are reported as the average of three replicates. The significant differences among all treatments were determined by the least significant difference (LSD) at p < 0.05 level. Microsoft Excel 2020 (Microsoft Corporation, Redmond, WA, USA) and Origin 2019b (OriginLab Corporation, Northampton, MA, USA) were used to analyze the data and plot the figures, respectively.

3. Results

3.1. Plant Height and Leaf Area Index of Cotton

3.1.1. Plant Height of Cotton

As the MIW and *B. subtilis* treatments were applied to the cotton during the first irrigation, the growth of cotton after the first irrigation was analyzed. Figure 7 shows the growth process of cotton height (H) for 1–49 days after the first irrigation. During this period, the H of different treatments gradually increased and tended to stabilize. During the 1–21-day period (budding stage), the average growth rate of H for each treatment in 2021 and 2022 was 0.91–1.90 cm d⁻¹, and during the 22–49-day period (budding stage to flowering stage), the corresponding range was 0.38–0.56 cm d⁻¹. Compared with B0, M increased the average growth rate of H during the 1–21-day period, with an increase of 28.3% in 2021 and 54.1% in 2022. Similarly, compared with the treatment without *B. subtilis*, the treatment with *B. subtilis* also increased the average growth rate of H during the 1–21-day period, with an increase of 16.4–55.2% in 2021 and 23.2–115.3% in 2022.

This study then analyzed H on the 49th day after the first irrigation. The effects of two irrigation water types (NMIW and MIW) on H were significant in the saline soil (p < 0.01). Compared with B0, M had a significant impact on H (p < 0.01), inducing an increase of 7.6% in 2021 and 9.1% in 2022. In addition, the amount of *B. subtilis* significantly impacted H in saline soil (p < 0.01), and H increased with the increasing *B. subtilis* amounts. The H of B4 and MB4 was maximized with the largest amount of *B. subtilis* in the NMIW and MIW treatments, respectively. The MIW treatment increased the H by 8.7–11.2%, compared

with the NMIW treatment. Under NMIW, the H of B2, B3 and B4 significantly increased by 9.6–20.3% (p < 0.01) in 2022, compared with B0. Under MIW, the H of all *B. subtilis* amounts significantly increased by 10.3–23.2% in 2021 and 14.9–30.7% in 2022 (p < 0.01), compared with B0.



Figure 7. Effects of *B. subtilis* amounts and irrigation water type on the plant height of cotton in the 2021–2022 growing seasons. (**a–c**) show plant height in 2021, and plant height under NMIW and MIW treatments in 2022, respectively. B0, B1, B2, B3, and B4 denote *B. subtilis* amounts of 0, 15, 30, 45, and 60 kg ha⁻¹, respectively, under the NMIW treatment. M, MB1, MB2, MB3, and MB4 denote *B. subtilis* amounts of 0, 15, 30, 45, and 60 kg ha⁻¹, respectively, under the NMIW treatment. Data represent the mean values of three replicates. Errors bars indicate standard errors. Different letters above the bars indicate significant differences among treatments at p < 0.01.

3.1.2. Leaf Area Index of Cotton

Figure 8 shows the leaf area index (LAI) growth process of cotton for the period of 1–49 days after the first irrigation. The LAI of different treatments gradually increased with time and tended to stabilize. In the 1–21-day period (budding stage), the average growth rate of LAI for each treatment in 2021 and 2022 was $0.02-0.09 d^{-1}$, while in the 22–49-day period (budding stage to flowering stage), the corresponding value was $0.02-0.05 d^{-1}$. Compared with B0, M increased the average growth rate of LAI during the 1–21-day period by 64.8% in 2021 and 19.7% in 2022. Similarly, compared with the treatment without *B. subtilis*, the treatment with *B. subtilis* also increased the average growth rate of LAI during the 1–21-day period by 44.6–287.6% in 2021 and 7.0–204.4% in 2022.



Figure 8. Effects of *B. subtilis* amounts and irrigation water type on the leaf area index of cotton in 2021–2022 growing seasons. (**a–c**) represent cotton leaf area index in 2021, and cotton leaf area index under the NMIW and MIW treatments in 2022, respectively. B0, B1, B2, B3, and B4 denote *B. subtilis* amounts of 0, 15, 30, 45, and 60 kg ha⁻¹, respectively, under the NMIW treatment. M, MB1, MB2, MB3, and MB4 denote *B. subtilis* amounts of 0, 15, 30, 45, and 60 kg ha⁻¹, respectively, under the NMIW treatment. M, MB1, MB2, MB3, and MB4 denote *B. subtilis* amounts of 0, 15, 30, 45, and 60 kg ha⁻¹, respectively, under the MIW treatment. Data are reported as the mean values of three replicates. Errors bars indicate standard errors. Different letters above the bars indicate significant differences among treatments at *p* < 0.01.

This study then analyzed the LAI on the 49th day after the first irrigation. Significant effects were exerted by irrigation water type and the *B. subtilis* amounts on LAI in the saline soil (p < 0.01). The LAI increased with the increasing *B. subtilis* amounts across both NMIW and MIW, with treatments B4 and MB4 maximizing LAI. Compared with B0, M resulted in a significant difference in LAI (p < 0.01), with an increase of 12.0% in 2021 and 14.4% in 2022. The MIW treatment increased the LAI by 7.9–14.4% compared with the NMIW treatment. Under NMIW, all *B. subtilis* amounts led to significant differences in LAI (p < 0.01) compared with B0 in 2022, with increases of 15.9–39.1%. Under MIW, all *B. subtilis* amounts resulted in significant differences in LAI (p < 0.01) compared with B0, with increases of 25.2–53.1% in 2021 and 30.4–52.6% in 2022 (p < 0.01).

3.1.3. Modeling Cotton Plant Height and Leaf Area Index

The logistic model is typically used to characterize the growth of crop height and LAI [51]. The effects of irrigation water type and B. subtilis amounts on the H and LAI of cotton following the first irrigation are presented in Figures 7 and 8, respectively. To reflect the differences between treatments, the days after the first irrigation during the growing season (t) was used as the independent variable. In general, the growth processes of H and LAI with t conformed to the regularity of the logistic model. Therefore, the logistic model was employed to characterize the relationships between (H and t) and (LAI and t). Tables 4 and 5 report the logistic model parameters fitted by H and LAI with different B. subtilis amounts under MIW. The logistic model was able to effectively describe the changes in H and LAI with t (R² > 0.96, RMSE_H < 3.00 cm and RMSE_{LAI} < 0.15). The shape parameters a_1, a_2 and c increased with the increasing B. subtilis amounts, while b_1 and b_2 decreased with increasing *B. subtilis* amounts. The theoretical maximum H and LAI of cotton (H_m in Table 4 and LAI_m in Table 5) were used to establish the relationships with the B. subtilis amount (Figure 9). Figure 9 presents these relationships under the MIW treatment, revealing that B. subtilis amounts less than 60 kg ha⁻¹ exerted a promoting effect on the nutritive organs (stem and leaves) of cotton.

Table 4. Logistic model parameters fitted for plant height with different *B. subtilis* amounts under magnetized ionized water.

Traction			2021					2022		
Ireatment	H _m	<i>a</i> ₁	b_1	R ²	RMSE _H	H _m	<i>a</i> ₁	<i>b</i> ₁	R ²	RMSE _H
М	73.09	0.46	-0.08	0.98	1.87	71.42	0.25	-0.05	0.96	2.90
MB1	76.20	0.52	-0.09	0.98	2.41	76.49	0.24	-0.08	0.98	2.06
MB2	78.31	0.60	-0.10	0.98	2.49	79.88	0.28	-0.09	0.99	0.91
MB3	80.79	0.65	-0.10	0.99	2.96	82.07	0.37	-0.10	0.99	1.08
MB4	83.66	0.71	-0.11	0.98	2.98	84.32	0.39	-0.11	0.99	1.14

M, MB1, MB2, MB3, and MB4 denote *B. subtilis* amounts of 0, 15, 30, 45, and 60 kg ha⁻¹ under the MIW treatment, respectively; H_m is the theoretical maximum value of plant height, cm; a_1 and b_1 are the shape parameters of the logistic model.

Table 5. 1	Logistic mod	el parameters	fitted for	leaf area	index with	different	B. subtilis	amounts	under
magnetiz	zed ionized w	vater.							

Transformer			20	21					20	22		
Ireatment	LAIm	<i>a</i> ₂	b_2	с	R ²	RMSELA	AI LAI _m	<i>a</i> ₂	b_2	С	R ²	RMSELAI
М	3.056	0.047	0.024	-0.003	0.99	0.09	2.972	0.024	-0.006	-0.003	0.98	0.05
MB1	3.458	0.305	0.003	-0.002	0.98	0.14	3.374	0.254	0.001	-0.003	0.98	0.04
MB2	3.688	0.515	-0.029	-0.001	0.99	0.05	3.658	0.369	-0.021	-0.002	0.99	0.04
MB3	4.081	0.683	-0.043	-0.001	0.99	0.06	3.950	0.526	-0.052	-0.001	0.99	0.05
MB4	4.142	0.753	-0.060	-0.001	0.99	0.07	4.098	0.634	-0.088	-0.001	0.99	0.03

M, MB1, MB2, MB3, and MB4 indicate *B. subtilis* amounts of 0, 15, 30, 45, and 60 kg ha⁻¹ under the MIW treatment, respectively; LAI_m is the theoretical maximum value of the leaf area index; and a_2 , b_2 and c are the shape parameters of logistic model.



Figure 9. Relationship between the *B. subtilis* amount under magnetized ionized water and the theoretical maximum values of plant height (cm) and leaf area index in 2021 and 2022. H_m and LAI_m are theoretical maximum values of plant height (cm) and leaf area index; *MB* denotes the *B. subtilis* amount under the MIW treatment (kg ha⁻¹).

3.2. Shoot Dry Matter of Cotton

B. subtilis amounts were observed to exert an extremely significant impact on the shoot dry matter of cotton (p < 0.01), yet irrigation water type did not have a significant effect (Figure 10). Compared with the shoot dry matter of B0, M increased by 3.3% and 2.1% at the budding stage in 2021 and 2022; and 0.8% and 10.9% at the boll stage in 2021 and 2022, respectively. The MIW treatment increased the shoot dry matter by 2.1–25.0% at the budding stage and by 0.5–18.6% at the boll stage compared with the NMIW treatment. Under NMIW, B3 and B4 resulted in significant differences in the shoot dry matter at the budding stage compared with B0 (p < 0.01) in 2022, with increases of 29.5–37.4%. Moreover, B2, B3 and B4 led to significant differences at the boll stage compared with B0 (p < 0.01) in 2022, with increases of 28.4–30.8%. Under MIW, MB3 and MB4 significantly increased shoot dry matter at budding compared with B0 (p < 0.01) by 27.8–31.0%. A similar trend was observed with MB2, MB3 and MB4 at the boll stage (p < 0.01) compared with B0, with increases of 16.5–20.6% in 2021. MB2, MB3 and MB4 also significantly increased the shoot dry matter (p < 0.01) compared with B0 by 37.1–49.3% at the budding stage and 29.0–53.5% at the boll stage in 2022.

Shoot dry matter is one of the important factors affecting crop yield. The proportion of biomass allocation varied with the cotton growth stages. The distribution and transportation of nutrients were centered on the vegetative organs in the early stage and gradually transferred to the reproductive organs after the budding stage. Coordinated biomass allocation is the key to increasing seed cotton yield [52]. To explore the effects of *B. subtilis* amounts on the reproductive and nutritive growth of cotton, quadratic functions were employed to characterize the *B. subtilis* amounts and ROP (proportion of cotton reproductive organs to total shoot dry matter) at the boll stage under MIW, and the *B. subtilis* amounts and NOP (proportion of cotton nutritive organs to total shoot dry matter) at the boll stage under MIW (Figure 11). Compared with B0, the MIW treatments and *B. subtilis* amounts increased the proportion of reproductive organs at the boll stage. This aided in increasing cotton yield. By taking the first derivative of the quadratic function (Figure 11), when the *B. subtilis* amount reached 62.5 kg ha⁻¹, the ROP and NOP reached maximum values of 37.3% and 62.7%, respectively, in 2021; when the *B. subtilis* amount reached 39.2 kg ha⁻¹, the ROP and NOP reached maximum values of 56.7% and 43.3%, respectively, in 2022.



Figure 10. Effects of *B. subtilis* amounts and irrigation water type on the cotton shoot dry matter in 2021 and 2022. (**a**–**c**) represent the cotton shoot dry matter in 2021, cotton shoot dry matter under NMIW in 2022 and cotton shoot dry matter under MIW in 2022, respectively. B0, B1, B2, B3, and B4 denote *B. subtilis* amounts of 0, 15, 30, 45, and 60 kg ha⁻¹, respectively, under the NMIW treatment. M, MB1, MB2, MB3, and MB4 denote *B. subtilis* amounts of 0, 15, 30, 45, and 60 kg ha⁻¹, respectively, under the MIW treatment. Data are reported as the mean the values of three replicates. Errors bars indicate standard errors. Different letters above the bars indicate significant differences among treatments at *p* < 0.01.



Figure 11. Relationship between the *B. subtilis* amount under MIW and ROP, and NOP at the boll stage in 2021 and 2022. ROP_{2021} , NOP_{2021} , ROP_{2022} and NOP_{2022} are the proportion of cotton reproductive organs and the proportion of cotton nutritive organs to the total shoot dry matter at the boll stage in 2021 and 2022, respectively (%). *MB* is the *B. subtilis* amount under MIW (kg ha⁻¹).

3.3. Chlorophyll Content (SPAD) of Cotton

The SPAD value is an indicator of plant physiological activity and is commonly used to indicate nitrogen levels in plants. No significant differences were observed in the SPAD values under the B0 and M treatments. However, greater SPAD values were determined under the MIW treatments compared with the NMIW treatments. The SPAD value initially increased and subsequently decreased with cotton growth, reaching a maximum value at 21 days after the first irrigation (at the end of the budding stage) (Figure 12). The MIW treatment increased the SPAD by 0.7–7.7%, compared with the NMIW treatment, at 21 days after the first irrigation. The *B. subtilis* amounts had a significant effect on the SPAD

value (p < 0.01). In particular, compared with B0, B3 induced significant differences in the SPAD value at 21 days after the first irrigation (p < 0.01), with increases of 4.6% in 2022. Moreover, under MB2, MB3 and MB4, there were also significant increases in the SPAD values at 21 days after the first irrigation compared with B0 (p < 0.01) (6.2–10.6% in 2021 and 6.8–11.6% in 2022).



Figure 12. Effects of *B. subtilis* amounts and irrigation water type on the leaf SPAD values in the 2021–2022 cotton growing seasons. (**a–c**) represent leaf SPAD values in 2021, leaf SPAD values under NMIW in 2022 and leaf SPAD values under MIW in 2022, respectively. B0, B1, B2, B3, and B4 denote *B. subtilis* amounts of 0, 15, 30, 45, and 60 kg ha⁻¹, respectively, under the NMIW treatment. M, MB1, MB2, MB3, and MB4 denote *B. subtilis* amounts of 0, 15, 30, 45, and 60 kg ha⁻¹, respectively, under the MIW treatment. Data denote the mean values of three replicates. Errors bars denote standard errors. Different letters above bars indicate significant differences among treatments at *p* < 0.01.

3.4. Soil Water Content and Water Consumption of Cotton

3.4.1. Soil Water Content

The boll stage is a key period for seed cotton yield formation as well as the critical period of water need in cotton crops. During this period, cotton needs to consume more water to form yield [53]. Therefore, the amount of water in the soil directly affects the cotton yield. Figure 13 depicts the changes in soil water content at the 0–100 cm depth during the boll stage. Cotton roots are mainly located within the top 0-40 cm of soil under drip irrigation [43]. Therefore, the top 0–40 cm soil layer was considered as the main root zone in the cotton field. The MIW treatment increased the soil water content by 2.7–28.4% compared with the NMIW treatment in the 0-40 cm soil layer. Soil in the MIW treatment displayed a good water diffusion and infiltration ability, which was reflected by the higher water content of M in the 0–40 cm soil layer compared with that of B0, increasing by 9.5% in 2021 and 12.1% in 2022. The adequate application of B. subtilis can improve the soil water content. Compared with the soil water content in the 0–40 cm depth of B0, that for B3, B4, MB3, and MB4 increased by 14.6–20.0% and 11.4–27.2% in 2021 and 2022, respectively. However, the soil water content in the 60–100 cm soil layer of B3, B4, MB3, and MB4 was lower than that of B0, with decreases of 8.6–11.5% and 1.9–6.3% in 2021 and 2022, respectively. The distribution of soil water in the root zone at different cotton growth stages (from budding to harvest) were similar to that at the boll stage (Figure 13). Thus, this study considers the boll stage as an example to explore the effect of irrigation water type and B. subtilis amounts on the soil water distribution.

100

(a)



(b)

Figure 13. Effects of *B. subtilis* amounts and irrigation water type on the soil water content during the boll stage in 2021 and 2022. (**a**–**c**) represent soil water content in 2021, soil water content under NMIW in 2022 and soil water content under MIW in 2022, respectively. B0, B1, B2, B3, and B4 denote *B. subtilis* amounts of 0, 15, 30, 45, and 60 kg ha⁻¹, respectively, under the NMIW treatment. M, MB1, MB2, MB3, and MB4 denote *B. subtilis* amounts of 0, 15, 30, 45, and 60 kg ha⁻¹, respectively, under the NMIW treatment.

100

(c)

3.4.2. Cotton Water Consumption

100

The changes in soil water storage (ΔW) and water consumption (ET_c) during the entire growth period of the cotton were calculated by Equations (7) and (8), respectively. The soil water storage and water consumption of cotton under different treatments in 2021 and 2022 are presented in Figure 14. Approximately one and a half months after the last irrigation, the cotton was harvested (from 22 August to 10 October 2021, and from 13 August to 30 September 2022). The irrigation amount in the whole cotton growth period of each treatment was less than the water consumption of cotton. The growing cotton consumed both irrigation water and soil water stored in the root zone. Under the NMIW treatment, ΔW increased with the increase in *B. subtilis* amounts; under the MIW treatment, ΔW exhibited an initial upward trend followed by a downward trend with the increase in *B. subtilis* amounts. Compared with B0, the ΔW of M increased by 42.7% in 2021 and 64.9% in 2022, while that of B1 to B4 under NMIW increased by 7.2–116.3% in 2022, and that of MB1 to MB4 under MIW increased by 16.8–76.1% in 2021 and by 162.1–274.6% in 2022.



Figure 14. Effects of *B. subtilis* amounts and irrigation water type on the water consumption (ET_c) and soil water storage changes (ΔW) in the cotton growing season of 2021–2022. (**a**–**c**) represent ET_c and ΔW in 2021, ET_c and ΔW under NMIW in 2022, and ET_c and ΔW under MIW in 2022, respectively. The irrigation amount was 501.6 mm in 2021 and 518.85 mm in 2022. The precipitation was 40.4 mm in 2021 and 35.0 mm in 2022. B0, B1, B2, B3, and B4 denote *B. subtilis* amounts of 0, 15, 30, 45, and 60 kg ha⁻¹, respectively, under the NMIW treatment. M, MB1, MB2, MB3, and MB4 denote *B. subtilis* amounts of 0, 15, 30, 45, and 60 kg ha⁻¹, respectively, under the MIW treatment.

Vigorously growing cotton plants require an ample amount of soil water to support their growth, physiological functions, and yield formation. As a result, there was an increase in both soil water storage changes and water consumption. In this study, based on the measurements of H, LAI, shoot dry matter and SPAD, the MIW treatment and *B. subtilis* amounts promoted the growth and development of cotton (particularly in the MB3 and MB4 treatments) and increased ETc. The trend of change in ETc was similar to the trend of change in ΔW . The MIW treatment increased the ETc by 1.2–3.4% compared with the NMIW treatment. Compared with B0, the ETc of M, MB3 and MB4 increased by 1.3%, 2.4% and 0.5% in 2021; and by 1.2%, 5.2% and 3.8% in 2022.

3.5. Salts Change in Soil

The changes in salts from the pre-sowing to post-harvesting stages were analyzed in the 0–40 cm and 0–100 cm soil layer (Figure 15). In the 0–40 cm soil layer, M reduced salts accumulation compared with B0, with a 3.1% reduction in 2021 and a 5.5% reduction in 2022. The *B. subtilis* amounts under the NMIW treatment reduced salts accumulation by 4.2–10.6% in 2022. Greater reductions in salts accumulation were observed with the *B. subtilis* amounts under the MIW treatment, with decreases of 9.5–12.9% in 2021 and 6.4–12.6% in 2022, respectively. Among them, the B3 treatment induced a soil desalination in the 0–40 cm soil layer, with a reduction in salts of 10.6% in 2022. Under the MIW treatment, MB3 and MB4 also induced soil desalination in the 0–40 cm soil layer, with reductions of 12.9% and 11.9% in 2021; and 12.6% and 12.0% in 2022. The results indicate that applying *B. subtilis* under the MIW treatment can lead to a greater desalting effect. In the 0–100 cm soil layer, all the treatments exhibited salts accumulation, whereas the M, *B. subtilis*, and *B. subtilis* combined with MIW treatments could reduce salts accumulation by 3.7% in 2021 and 2.3% in 2022; by 2.3–5.4% in 2022; and by 5.8–6.2% in 2021 and 6.5–7.4% in 2022, respectively.



Figure 15. Effects of *B. subtilis* amounts and irrigation water type on the salts change in soil for the 2021–2022 cotton growing season. (**a**–**c**) represent salts change in soil in 2021, salts change in soil under NMIW in 2022 and salts change in soil under MIW in 2022, respectively. B0, B1, B2, B3, and B4 denote *Bacillus subtilis* amounts of 0, 15, 30, 45, and 60 kg ha⁻¹, respectively, under the NMIW treatment. M, MB1, MB2, MB3, and MB4 denote *B. subtilis* amounts of 0, 15, 30, 45, and 60 kg ha⁻¹, respectively, under the MIW treatment.

3.6. Seed Cotton Yield, WUE and IWUE

3.6.1. Seed Cotton Yield and Yield Composition

Table 6 shows the seed cotton yield and yield composition of cotton. The treatment M raised seed cotton yield by 4.0% in 2021 and 6.5% in 2022 compared with B0, yet these increases were not significant. With the exception of B0 and M in 2021 and B2 in 2022, no differences were observed in the cotton single boll weight. This indicates that increasing the number of cotton bolls is key to increasing seed cotton yield. The MIW treatment increased seed cotton yield by 2.1–12.2% compared with NMIW treatment. In 2021 and 2022, B3

and MB3 maximized seed cotton yield under NMIW and MIW, respectively. Compared with B0, treatments B3 and B4 exerted significant differences in the boll number per plant and seed cotton yield (p < 0.05) under NMIW in 2022, increasing the boll number per plant by 19.7% and 15.7%, and seed cotton yield by 16.5% and 15.9%, respectively. All the *B. subtilis* amounts had significant differences in the boll number per plant and seed cotton yield (p < 0.05) under MIW, increasing the boll number per plant by 6.5–16.1% in 2021 and 14.2–20.9% in 2022, and seed cotton yield by 9.8–21.6% in 2021 and 13.1–20.6% in 2022. Thus, applying *B. subtilis* under MIW treatment induced more significant differences (p < 0.05) with B0 compared with other treatments.

Table 6. Seed cotton yield, boll number per plant, single boll weight and water use efficiency under different treatments across the cotton growing seasons in 2021 and 2022.

Year	Treatment	Seed Cotton Yield (kg ha ⁻¹)	Boll Number Per Plant	Single Boll Weight (g)	WUE (kg ha ⁻¹ mm ⁻¹)	IWUE (kg ha ⁻¹ mm ⁻¹)
	B0	$6304.82 \text{ e} \pm 48.70$	$4.48~\mathrm{c}\pm0.10$	$6.17\mathrm{b}\pm0.12$	$11.27 \text{ c} \pm 0.09$	$12.57 \text{ e} \pm 0.05$
	М	$6558.18~{ m de}\pm23.89$	$4.67~\mathrm{bc}\pm0.12$	$6.15b\pm0.12$	$11.57~\mathrm{c}\pm0.04$	$13.07~\mathrm{de}\pm0.05$
0001	MB1	$6922.32 \text{ cd} \pm 290.34$	$4.87~b\pm0.15$	$6.23~\mathrm{ab}\pm0.12$	$12.31~\mathrm{b}\pm0.52$	$13.80~\mathrm{cd}\pm0.58$
2021	MB2	7270.68 bc \pm 210.82	$4.77~b\pm0.12$	$6.68~\mathrm{a}\pm0.10$	12.79 a \pm 0.37	$14.49\mathrm{bc}\pm0.42$
	MB3	7666.45 a \pm 140.59	$5.20~\mathrm{a}\pm0.17$	$6.47~\mathrm{ab}\pm0.55$	13.39 a \pm 0.25	$15.28~\mathrm{a}\pm0.28$
	MB4	7470.14 ab \pm 145.11	$4.83~b\pm0.15$	$6.78~a\pm0.32$	$13.28~\mathrm{a}\pm0.26$	14.89 ab \pm 0.29
	B0	$6597.05 c \pm 600.98$	$4.78~\mathrm{c}\pm0.18$	$5.74~\mathrm{ab}\pm0.34$	$11.68b\pm1.08$	$12.71 \text{ c} \pm 1.16$
	М	$7026.08 \text{ bc} \pm 166.52$	$5.12~\mathrm{bc}\pm0.14$	5.71 ab \pm 0.09	12.30 ab \pm 0.29	$13.54 \text{ bc} \pm 0.32$
	B1	$6651.05\ c\pm 500.051$	$4.88~\mathrm{c}\pm0.53$	5.69 ab \pm 0.17	$11.76~\mathrm{b}\pm0.88$	$12.82 \text{ c} \pm 0.96$
	B2	7290.00 abc \pm 439.80	5.39 ab \pm 0.19	$5.64b\pm0.21$	12.92 ab \pm 0.80	$14.05~\mathrm{abc}\pm0.85$
2022	B3	7685.96 ab \pm 108.00	$5.72~\mathrm{a}\pm0.05$	5.71 ab \pm 0.10	13.36 a \pm 0.19	14.81 ab \pm 0.21
2022	B4	7646.94 ab \pm 180.67	$5.53~\mathrm{ab}\pm0.19$	5.75 ab \pm 0.21	13.23 a \pm 0.31	$14.74~\mathrm{ab}\pm0.35$
	MB1	7461.05 ab \pm 544.56	$5.17\mathrm{bc}\pm0.37$	$6.03~\mathrm{a}\pm0.54$	12.82 ab \pm 0.94	14.38 ab \pm 1.05
	MB2	7707.02 ab \pm 122.85	$5.46~\mathrm{ab}\pm0.17$	$5.87~\mathrm{ab}\pm0.18$	13.22 a \pm 0.21	$14.85~\mathrm{ab}\pm0.24$
	MB3	7952.99 a \pm 275.79	$5.78~\mathrm{a}\pm0.11$	5.75 ab \pm 0.14	13.39 a \pm 0.46	15.33 a \pm 0.53
	MB4	7808.94 a \pm 414.03	$5.69~a\pm0.34$	$5.71~\mathrm{ab}\pm0.15$	$13.32~\mathrm{a}\pm0.71$	$15.05~\mathrm{a}\pm0.80$

B0, B1, B2, B3, and B4 denote *B. subtilis* amounts of 0, 15, 30, 45, and 60 kg ha⁻¹, respectively, under the NMIW treatment. M, MB1, MB2, MB3, and MB4 denote *B. subtilis* amounts of 0, 15, 30, 45, and 60 kg ha⁻¹, respectively, under the MIW treatment. Different letters above the bars indicate sig-nificant differences among treatments at p < 0.05.

3.6.2. WUE and IWUE

Compared with B0, M increased WUE by 2.7% in 2021 and 5.3% in 2022, yet these differences were not significant. The MIW treatment increased WUE by 0.2–9.0% compared with the NMIW treatment. B3 and B4 led to significant differences in WUE (p < 0.05) in 2022, with increases of 14.4% and 13.3% compared with B0. Under the MIW treatment, MB2, MB3 and MB4 significantly increased WUE (p < 0.05) by 9.2–18.8% in 2021 and 9.8–14.6% in 2022 compared with B0. Among them, MB3 maximized WUE in 2021 and 2022.

Compared with B0, M increased IWUE by 4.0% in 2021 and 6.5% in 2022, yet these differences were not significant. The MIW treatment increased IWUE by 2.1–12.2% compared with the NMIW treatment. B3 and B4 significantly increased IWUE (p < 0.05) by 16.5% and 16.0% in 2022 compared with B0. Under MIW, all *B. subtilis* amounts induced significant differences in IWUE (p < 0.05), increasing WUE by 9.2–18.8% in 2021 and 9.8–14.6% in 2022, and increasing IWUE by 9.8–21.6% in 2021 and 13.1–20.6% in 2022 compared with B0. Among them, the IWUE was maximized under MB3 in 2021 and 2022.

3.6.3. Effect of B. subtilis Amount on Seed Cotton Yield, WUE and IWUE

The quadratic relationships between seed cotton yield and *B. subtilis* amount, WUE and *B. subtilis* amount, and IWUE and *B. subtilis* amount under the MIW treatment are presented in Figure 16. The coefficients of determination (R^2) for seed cotton yield, WUE and IWUE were 0.72, 0.85 and 0.85, respectively. Based on the production function of

B. subtilis, the highest yield, WUE and IWUE were obtained with *B. subtilis* amounts of 51.8, 55.0 and 51.4 kg ha⁻¹, respectively. At the *B. subtilis* amounts of 51.4–55.0 kg ha⁻¹, there were minimal differences in seed cotton yield (7707.91 to 7711.56 kg ha⁻¹), WUE (13.27 to 13.28 kg ha⁻¹ mm⁻¹) and IWUE (15.09 to 15.10 kg ha⁻¹ mm⁻¹). Based on comprehensive consideration of seed cotton yield and WUE and IWUE, and salts accumulation in soil, the minimal *B. subtilis* amount of 51.4 kg ha⁻¹ was determined as optimal for film-mulched drip irrigation under the MIW treatment for cotton cultivated under FMDI in the saline soil of southern Xinjiang, China.



Figure 16. Relationship between *B. subtilis* amount and seed cotton yield, WUE and IWUE under the MIW treatment. Y is seed cotton yield (kg ha⁻¹); WUE is cotton water use efficiency (kg ha⁻¹ mm⁻¹); IWUE is cotton irrigation water use efficiency (kg ha⁻¹ mm⁻¹); and *MB* is the *B. subtilis* amount under magnetized ionized water irrigation (kg ha⁻¹).

B0, B1, B2, B3, and B4 denote *B. subtilis* amounts of 0, 15, 30, 45, and 60 kg ha⁻¹, respectively, under the NMIW treatment. M, MB1, MB2, MB3, and MB4 denote *B. subtilis* amounts of 0, 15, 30, 45, and 60 kg ha⁻¹, respectively, under the MIW treatment. Different letters above the bars indicate significant differences among treatments at p < 0.05.

4. Discussion

4.1. Effects of Magnetized Ionized Water Treatment on Soil Water and Salt Conditions and Cotton Growth

Cotton is a salt-tolerant crop, yet lower soil water content and/or higher salts content can still limit the growth and yield of cotton [54]. Based on the average of the two years of experimental results, this study concluded that the MIW treatment was able to increase water content by 10.8% and reduce salts accumulation by 4.3% in the cotton's main root zone soil (0-40 cm). This may be attributed to the capability of the magnetized and ionized technology to reduce the water surface tension coefficient [55], viscosity coefficient [56] and the binding degree [20], thus enhancing water diffusion and permeability, and promoting leaching of salts [9]. Therefore, changes in the physical and chemical properties of MIW could increase soil water content and reduce salts accumulation in soil. Khoshravesh et al. [57] reported significant effects of magnetized irrigation water on soil water content (p < 0.01), which was greater than that under non-magnetized water. Mostafazadeh et al. [58] found that the soil water content of a magnetized treatment increased by 7.5% and an average reduction of 33.6% in soil sodium compared with the control treatment. Wang et al. [41] observed that ionized technology could increase soil water content by 18.9% and promote leaching of salts by 8.5%. The results of this study, as well as previous research, prove that the MIW treatment is effective in regulating soil water content and salt conditions.

The MIW treatment induced an increase in the soil water content and reduced salts accumulation in the soil, which promoted the growth and yield of cotton in the saline soil. Zhang et al. [59] employed magnetic treatment to alleviate salt stress in cotton seed germination, and the results showed that cotton seed maximum relative water absorption and germination increased by 19.8% and 41.9%, respectively, compared with a non-magnetized

water treatment. Ma et al. [60] reported significant increases in plant height, biomass and the chlorophyll content of rice under MIW by 24.3%, 30.4% and 9%, respectively, compared with NMIW, relieving the inhibition due to salt stress. Patil et al. [61] found that magnetized water promoted banana seed germination, flowering, crop growth, fruit growth and yield, avoiding salts accumulation in nearby plants. Wei et al. [13] demonstrated that plant height, leaf area index, shoot dry matter, chlorophyll content and ET_c of cotton increased by 8.0%, 13.3%, 14.7%, 19.2% and 6%, respectively, as a result of an ionized water treatment in saline soil. This study also showed that the MIW treatment could promote cotton growth (i.e., H, LAI, shoot dry matter and SPAD of cotton), water consumption, yield, and WUE and IWUE. This is in agreement with previous research results.

4.2. Effects of B. subtilis Amount on Soil Water and Salt Conditions and Cotton Growth

The dispersion and expansion of the soil aggregate structure in saline soil cause the blocking of water-conducting pores [62], which reduces the soil water content and leaching of salts. This is a key reason explaining the inhibition of crop growth in saline soil. In order to address the effects of saline stress, the promotion of crop growth by *B. subtilis* is twofold [63,64]. Firstly, B. subtilis can alleviate the adverse effects of soil water and salt stress on crops. B. subtilis can biosynthesize extracellular polymeric substances (EPS), which have the ability to promote the formation of soil aggregate structures and increase the stability of the aggregation structure in saline soil [34]. Zhou et al. [65] reported that the addition of *B. subtilis* increased the soil water stable aggregate content, with a maximum increase of 17.6%, and provided good soil water and salt conditions. EPS possess a large water holding capacity and can influence the soil water retention characteristics and hydraulic conductivity by modifying the structural and physicochemical properties of the rhizosphere soil [66]. Previous research has shown that the ESP biosynthesized by *B. subtilis* increased soil water content [67–70]. This study demonstrated that applying *B. subtilis* improved the soil water retention capacity. This was revealed by the higher soil water contents of B3, B4, MB3 and MB4 in the 0–40 cm soil layer than that of B0, and lower contents in the 60–100 cm soil layer. The application of *B. subtilis* was beneficial to the formation of soil aggregates and improved the soil pore structure, which subsequently promoted leaching of salts. Sun et al. and Hou et al. found that applied *B. subtilis* effectively reduced the salts content in soil [71,72]. This study also showed that the application of *B. subtilis* could reduce salts accumulation in soil during the cotton growth period, particularly under MIW. The salts accumulation in the 0–100 cm soil was closed as the *B. subtilis* amount increased under MIW, yet MB3 and MB4 exhibited desalination in the 0–40 cm soil in 2021 and 2022. This indicates that applied *B. subtilis* can result in the salts in soil leaching from the upper layer (0–40 cm) of the soil to the deeper layer (60–100 cm). It is noteworthy that MIW treatment could increase soil water content and reduce salts accumulation in upper layer soil (0–40 cm), compared with NMIW treatment. This might be attributed to the changes in the physicochemical properties of irrigation water under the MIW treatment, which enhanced the substance transportation capability of the irrigation water, such as soil water diffusion ability and leaching of salts, thereby providing a better growing environment for cotton. This study also noted that the soil water holding and salt leaching capacities of B4 and MB4 were inferior to those of B3 and MB3. This may be due to an excessive amount of B. subtilis and its secretions blocking soil capillaries, which in turn, affects the capillary connectivity of the soil. The investigation of this phenomenon is reserved for future research.

Additionally, *B. subtilis* can produce phytohormones, such as Indole acetic acid (IAA), to promote plant growth [73,74]. Based on a two-year field experiment, Lu et al. [75] found that applied *B. subtilis* can significantly improve the plant height and yield of rice (p < 0.05). Zhao et al. [76] reported that, compared with a control treatment, the treatment of *B. subtilis* increased seed cotton yield by 6.1%. Moreover, the application of *B. subtilis* resulted in the plant fresh weight, dry weight, and number of branches increasing by 28.2%, 18.6%, and 5.1%, respectively. Lyu et al. [77] demonstrated an increase in cannabis flower fresh

weight by 5.1% for plants inoculated with *B. subtilis*. Our results indicated that moderate applications of *B. subtilis* promoted crop growth (height, LAI, shoot dry matter and SPAD), seed cotton yield, WUE, and IWUE. In order to quantify the change in cotton plant height and LAI with time after the first irrigation under different *B. subtilis* amounts, a logistic model was used to describe the growth process of cotton. Based on this, the logistic growth models of H and LAI under the MIW treatment were established. H_m and LAI_m increased with the *B. subtilis* amounts for the range of 0–60 kg ha⁻¹. Cotton descends from perennial ancestors, and as a result, excessive water or nutrients leads to rank growth, namely an increase in stem length, internode length, and leaf number at the expense of reproductive growth [78]. We also found that excessive amounts of *B. subtilis* resulted in predominantly nutritive growth and reduced seed cotton yield (Figures 9, 11 and 16). The application of *B. subtilis* played an important role in improving cotton growth, WUE, IWUE, seed cotton yield, soil water and salt conditions, and seed cotton yield. An optimal application amount of *B. subtilis* was proposed to account for these components.

5. Conclusions

The MIW treatment and application of *B. subtilis* were more effective than B0 in improving soil water and salts accumulation and promoting cotton growth. Under the MIW treatment, WUE, IWUE and seed cotton yield were noticeably affected by *B. subtilis* amounts in a saline soil of southern Xinjiang during the growing seasons in 2021 and 2022. Compared with the B0 treatment, the application of *B. subtilis* under the MIW treatment led to a higher seed cotton yield (9.8–21.6%), WUE (9.2–18.8%), and IWUE (9.8–21.6%). MIW treatment increased seed cotton yield by 2.1–12.2%, WUE by 0.2–9.0%, and IWUE by 2.1–12.2%, compared with the NMIW treatment. Moreover, the *B. subtilis* treatments significantly increased H, LAI and the boll number per cotton plant (p < 0.05). The production functions of seed cotton yield, WUE and IWUE were analyzed for differing amounts of *B. subtilis* (Figure 16) under the MIW treatment. The *B. subtilis* amount of 51.4 kg ha⁻¹ under the MIW treatment could optimize yield, WUE and IWUE, promote the growth of cotton, improve soil water content and reduce salts accumulation, compared with B0 and MB4. Finally, 51.4 kg ha⁻¹ of *B. subtilis* under MIW treatment is recommended for cotton cultivated under FMDI in a saline soil in southern Xinjiang, China.

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