

Article

Control of Fungal Diseases and Increase in Yields of a Cultivated Jujube Fruit (*Zizyphus jujuba* Miller var. *inermis* Rehder) Orchard by Employing *Lysobacter antibioticus* HS124

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Received: 7 November 2019; Accepted: 13 December 2019; Published: 15 December 2019



Abstract: The objective of this study is to investigate the inhibitory effects of *Lysobacter antibioticus* HS124 on fungal phytopathogens causing gray mold rot, stem rot, and anthracnose. Another objective of this study is to promote the yield of fruit in jujube farms. *L. antibioticus* HS124 produces chitinase, a lytic enzyme with the potential to reduce mycelial growth of fungal phytopathogens involving hyphal alterations with swelling and bulbous structures, by 20.6 to 27.3%. Inoculation with *L. antibioticus* HS124 decreased the appearance of fungal diseases in jujube farms and increased the fruit yield by decreasing fruit wilting and dropping. In addition, *L. antibioticus* HS124 produced the phytohormone auxin to promote vegetative growth, thereby increasing the fruit size. The yield of jujube fruits after *L. antibioticus* HS124 inoculation was increased by 6284.67 g/branch, which was 2.9-fold higher than that of the control. Auxin also stimulated fine root development and nutrient uptake in jujube trees. The concentrations of minerals, such as K, Ca, Mg, and P in jujube fruits after *L. antibioticus* HS124 inoculation were significantly increased (1.4- to 2.0-fold greater than the concentrations in the control). These results revealed that *L. antibioticus* HS124 could not only control fungal diseases but also promote fruit yield in jujube farms.

Keywords: auxin; biocontrol; chitinase; fruit; fungal pathogen; jujube; *Lysobacter antibioticus* HS124; mineral concentration; production; Rhamnaceae

1. Introduction

Jujube (*Zizyphus jujuba* Miller var. *inermis* Rehder) is an elliptical or spherical-shaped fruit [1]. Approximately 170 species of jujube have been cultivated in China over the past 5000 years [1,2]. They are mainly cultivated in central and southern China and southern and eastern Europe where the climate is warm [3]. Jujube fruit is mostly consumed fresh. Its dried form is also consumed in various food products, such as bread, cake, candy, powder, and juice [4]. Jujube fruit is rich in fiber, minerals, phenolic compounds, and vitamins [1,5]. It can strengthen the cardiovascular system with its antioxidant capacity [6,7]. Eating one jujube fruit per day provides an adult with the dose of vitamins B and C recommended by the Food and Agriculture Organization of the United Nations (FAO)/World Health Organization (WHO) [1,8]. In addition, jujube seeds are known to contain saponins with pharmacological properties, such as antibacterial, insecticidal, anti-inflammatory,

and immunity-enhancing activities [9,10]. In ancient China and Korea, jujube fruit was commonly used for health promotion and as a medicinal ingredient [10–13].

A high incidence of fruit fungal pathogens has been reported in intensive cultivation of jujube because of its high sugar content of approximately 5.4 to 10.5 g/100 g and its high moisture content of over 80% [1,14]. Fungal diseases in jujube fruits and trees include gray mold rot, stem rot, and anthracnose [15–18], which can reduce jujube production. Fungi and spores are mainly present in the soil and atmosphere. Rain, increases the atmospheric humidity, resulting in the cracking of jujube fruits. Thus, the interior part of the jujube is exposed to the outer environment where fungi and spores are present. Generally, it is difficult to control fungal diseases and fungicides can be used to control the fungal diseases in jujube farms [19,20]. However, the continuous use of fungicides results in an increased number of fungi with acquired resistance. Fungicide-resistant fungi require higher doses and increased fungicidal application frequency, eventually necessitating the development of new fungicides [20]. Although intensive farming methods ensure high yields and quality, they also require the use of excessive chemicals, such as fungicides, pesticides, and fertilizers [19]. Excessive use of chemicals can lead to environmental pollution, such as the eutrophication of water quality and salt accumulation in the soil [21], the destruction of soil microorganisms, and the inhibition of plant growth [22,23]. Recently, increased focus has been directed toward environmentally friendly practices in fruit cultivation systems. However, the cultivation of fruit tree orchards still suffers from fungal diseases because of the lack of knowledge regarding biological control methods.

Recent changes in awareness about the environment and changes in consumer trends favoring environmentally friendly crop products have prompted a demand to transform the existing farming methods [24,25]. Among various other eco-friendly cultivation methods, environmentally friendly cultivation methods that employ biological control by using microorganisms can promote crop production. Thus, they are gaining popularity because of their high potential value [25–28]. Plant growth promoting rhizobacteria (PGPR) play a significant role in reduction of infection by plant pathogenic fungi leading to be a promising alternative to control plant diseases. PGPR are known to produce lytic enzymes, preventing infection by fungal diseases [27–30]. However, the biocontrol efficacy of PGPR on diseases associated with jujube fruit is not clearly investigated. PGPR provide plants with phytohormones [24,31], especially auxin, which can loosen the cell wall of root tissue cells, allowing cells to absorb additional nutrients, thereby promoting the growth of root hair and lateral roots [24,25,27,28,31,32]. In addition, auxin helps establish symbiosis with rhizobia or mycorrhiza and facilitates the uptake of nutrients from soils, promoting plant production [24,25,27,28,31,32]. In particular, after the flower is fertilized, auxin acts on the division and expansion of cells, promoting vegetative growth and increasing fruit size and yield [31,33,34]. *Lysobacter* species are isolated from rhizosphere soil. Genus *Lysobacter* is famous for their positive effects on plant health [30,35]. *Lysobacter antibioticus* strains can produce a variety of bioactive compounds, including lytic enzymes and antimicrobial compounds, that can effectively inhibit the growth of phytopathogenic fungi [30]. However, the effects of bioenhancers derived from *L. antibioticus* strains on the production and nutrient uptake in fruit trees remain poorly understood.

Fruit trees have been studied with respect to many bacterial species that can act as PGPR. These bacterial species have been reported to successfully provide biological control of diseases and can also improve fruit production. However, there are only a few studies on the simultaneous use of PGPR for the biological control of disease and the promotion of fruit production [26,36]. Currently, the increasing demand for fruit tree production, along with a significant reduction in the use of synthetic chemical fertilizers and fungicides, is a major challenge. Despite fungicide treatment for field survey areas of cultivated jujube orchards, several other flowers and fruits have been found to be contaminated with fungal diseases, including gray mold rot, stem rot, and anthracnose (Figure 1c). In Korea, fungal diseases caused by *Botrytis cinerea*, *Botryosphaeria dothidea*, and *Colletotrichum gloeosporioides* are major diseases in jujube fruit. These causative agents are potential fungal pathogens in jujube farms [15–18]. To improve the production of jujube fruit, it is crucial to consider the capacity of beneficial

microbes on fungal diseases and the production of jujube fruits in cultivated orchards. Therefore, the aim of this study is to observe the biocontrol capacities of fungal plant pathogens such as *B. cinerea*, *B. dothidea*, and *C. gloeosporioides*. Another objective of this study was to promote fruit yield in cultivated jujube orchards through the performance of antagonistic bacteria, such as *L. antibioticus* HS124.



Figure 1. Locations of the study sites (a). Cultivated jujube orchard at the experimental site (b), gray mold rot by *B. cinerea* (left), stem rot by *B. dothidea* (center), and anthracnose by *C. gloeosporioides* (right) (c).

2. Materials and Methods

2.1. Bacterial Culture

The antagonistic bacterial strain *L. antibioticus* HS124 was isolated from field rhizosphere soil in Naju City, Korea [30]. It was subcultured in casein-yeast (CY) agar medium (Pancreatic digest of casein (Neogen, Lansing, MI, USA), 3 g/L; $\text{CaCl}_2 \cdot 2\text{H}_2\text{O}$ (Daejung chemicals, Siheung, Korea), 1.36 g/L; yeast extract (Daejung chemicals, Siheung, Korea), 1 g/L; agar (Daejung chemicals, Siheung, Korea), 20 g/L; and distilled water, 1 L). The CY medium composition and preparation for inoculation of *L. antibioticus* HS124 followed instructions of the Korean Agriculture Culture Collection (KACC, Suwon, Korea). It was incubated at 30 °C for three days. A single colony from fresh culture medium was pre-inoculated into CY broth (Pancreatic digest of casein, 3 g/L; $\text{CaCl}_2 \cdot 2\text{H}_2\text{O}$, 1.36 g/L; yeast extract, 1 g/L; and distilled water, 1 L) and cultured for three days. The pre-inoculated broth culture of strain HS124 was also checked with an ultraviolet (UV) spectrometer (Shimadzu, Kyoto, Japan) at 600 nm. Its optical density (OD) value was 1.71 [37]. Then, 100 μL of the pre-inoculated *L. antibioticus* HS124 culture (10^7 CFU/mL) was inoculated into 100 mL of CY broth and incubated at 30 °C for 10 days with shaking (130 rpm). Samples were collected on each inoculation day and spread onto CY agar medium using the serial dilution technique. The viable bacterial cells were counted as colony forming unit (CFU) on each inoculation day and the growth pattern of *L. antibioticus* HS124 was examined [30].

2.2. Chitinase Activity

To examine the chitinase activity based on incubation time, *L. antibioticus* HS124 was cultured in chitin–potato–dextrose (CPD) broth (Colloidal chitin [38], 10%; yeast extract, 0.05%; and potato dextrose broth (Daejung chemicals, Siheung, Korea), 1.2%) at 30 °C for 10 days in a shaking incubator at 130 rpm [39]. Samples were taken at 2-day intervals and centrifuged at 12,000 rpm for 10 min. The resulting supernatant was used to investigate chitinase activity on each sampling day.

Chitinase activity was measured by the Lingappa and Lockwood method [40]. Briefly, a reaction mixture containing 50 µL of *L. antibioticus* HS124 supernatant, 450 µL of 0.2 M sodium acetate buffer (pH 5.0), and 500 µL of 0.5% colloidal chitin was incubated at 37 °C. Subsequently, 200 µL of 1 N NaOH (Yakuri pure chemicals, Kyoto, Japan) was added to terminate the reaction. The mixture was centrifuged at 12,000 rpm for 10 min at 4 °C. Next, 750 µL of the supernatant was mixed with 1 mL of Schales' reagent and 250 µL of distilled water and incubated at 100 °C boiling water for 15 min. The amount of reducing sugar was then quantitatively analyzed at 420 nm using a UV-spectrometer. The assay was repeated three times. One unit of chitinase enzyme activity was determined as the quantity of enzyme releasing 1 µmol of *N*-acetyl-glucosamine per hour at 37 °C.

2.3. Antifungal Activity of *L. antibioticus* HS124 toward Fungal Phytopathogens

The phytopathogenic fungi used in this study, *B. cinerea* (KACC 41008), *B. dothidea* (KACC 45481), and *C. gloeosporioides* (KACC 40897), were provided by the KACC. These three phytopathogenic fungi were cultured in potato dextrose agar (PDA) medium at 25 °C for seven days. The antagonistic activities of *L. antibioticus* HS124 against three phytopathogenic fungi were determined by the dual culture method. One-loopful of *L. antibioticus* HS124 colonies was streaked on one side of a CY agar plate and incubated at 30 °C for three days [30]. Then, a 5-mm plug of each phytopathogenic fungus from a 7-day-old culture plate was placed on the other side of the same plate at a distance of 4 cm and the plates were incubated at 25 °C. Depending on the growth rate of each phytopathogenic fungus, the incubation days were different: *B. cinerea*, four days; *B. dothidea*, five days; and *C. gloeosporioides*, eight days. A plate without inoculation of *L. antibioticus* HS124 was used as a control. The experiment was repeated three times with three replications. The inhibition of fungal growth by the HS124 strain was determined with the following formula: inhibition (%) = $[(\alpha - \beta)/\alpha] \times 100$, where α was the radial growth of phytopathogenic fungus on the control plate and β was the radial growth of phytopathogenic fungus on the dual culture plate [27,28,30].

A small piece of mycelium at the boundary of the fungal colony inhabited by *L. antibioticus* HS124 was taken and observed for hyphal deformation and degradation caused by *L. antibioticus* HS124 under a light microscope at 200× magnification (Olympus BX41TF, Tokyo, Japan).

2.4. Indole-3-Acetic Acid (IAA) Production by *L. antibioticus* HS124

Quantitative analyses of IAA production by *L. antibioticus* HS124 were performed using a UV spectrometric method. Briefly, *L. antibioticus* HS124 was cultured in a medium containing 0.1 g/L crab shell powder (Purne, Jangseong, Korea), 0.2 g/L Na₂HPO₄ (Daejung chemicals, Siheung, Korea), 0.1 g/L KH₂PO₄ (Daejung chemicals, Siheung, Korea), 0.5 g/L NaCl (Daejung chemicals, Siheung, Korea), 0.1 g/L NH₄Cl (Yakuri pure chemicals, Kyoto, Japan), 0.05 g/L MgSO₄ 7H₂O (Shimakyu's pure chemicals, Osaka, Japan), 0.05 g/L CaCl₂ 2H₂O, 0.01 g/L yeast extract, and 0.1 g/L L-tryptophan (Junsei chemical, Tokyo, Japan). The culture was incubated at 30 °C in a shaking incubator (140 rpm). Samples were taken every two days five times from the day of inoculation. Quantitative measurement of the HS124 strain was performed according to Salkowski's method [41]. Briefly, the samples were centrifuged at 12,000 rpm for 10 min at 4 °C and 1 mL of the resulting supernatant was mixed with 2 mL of Salkowski's reagent. Subsequently, the reaction mixture was incubated at room temperature under dark conditions for 25 min. The IAA concentration of each sample was measured at 530 nm using a UV-spectrometer.

2.5. Study Area and Field Experimental Conditions

The experimental sites of a cultivated jujube orchard were located (35°84'69" N, 128°80'40" E) in Gyeongsan City, Gyeongbuk Province, Korea (Figure 1a). The soils at the study sites were fluvial deposits. The major soil type of the study area was sandy loam. The experimental sites in Gyeongsan City had a temperate climate with a mean temperature of 14.6 °C in 2016. The precipitation on-site was 1227 mm, approximately 57% of which fell between July 2016 and September 2016.

The jujube trees in the study area were planted in the 1970s (Figure 1b). In 2016, approximately 46-year-old trees with a height of approximately 4 m and a diameter of approximately 15 cm were distributed in the study areas. In addition, a rain shelter with transparent vinyl was installed above the tree crown to prevent the fall of fruits because of rain (Figure 1b). Jujube field experiment was arranged using a rectangular plot design measuring 8 m wide × 24 m long with a distribution of 10 trees (Figure 1b). The following two treatment groups were used in the field experiment, each with three replicates: (1) control without *L. antibioticus* HS124 inoculation and (2) *L. antibioticus* HS124 inoculation.

The *L. antibioticus* HS124 cultures were prepared in CY medium. The liquid form of microbial product containing 10^{10} CFU/mL of *L. antibioticus* HS124 (GCM+, Purne, Jangseong, Korea) was used for large scale cultivation of *L. antibioticus* HS124. Typically, 150 mL of *L. antibioticus* HS124 liquid product was inoculated into 500 L of CY medium and cultured at 30 °C for seven days using a fermenter. The *L. antibioticus* HS124 cultures were diluted with tap water (1:2 v/v) and poured onto the soil adjacent to the tree roots at approximately 2-week intervals 14 times from April to September 2016.

2.6. Mineral Concentration, Fruit Characteristics, and Yield

To determine the fruit characteristics (length and diameter) and fruit yield after treatments, six jujube trees in each plot were selected and the fruits of branches with a similar length and height and facing south compared to a standard were harvested in October 2016. The weight of fresh fruit from a standard branch in each sample tree was recorded to determine the yield. In addition, fruit length and diameter were calculated with respect to 10 fruit in each sample tree.

Undamaged jujube fruits were selected and washed thoroughly with distilled water to remove dirt and air-dried. The fruit was finely chopped into small pieces using a sharp knife and the seeds were separated from the pulp. The prepared sample was pretreated using a dry decomposition method [42]. For dry decomposition, 0.7 g of the sample was ashed at 550 °C, then 10 mL of a diluted solution (HCl:distilled water = 1:1) was added to the ash, followed by decomposition at 25 °C for six hours. Thereafter, a funnel was inserted into a 50-mL volumetric flask, the ash solution was filtered through filter paper, massed with distilled water, and used as a test solution. The mineral compositions were determined using a procedure described by the AOAC (Association of Official Analytical Chemists) [42]. Each mineral (potassium (K), calcium (Ca), magnesium (Mg), and phosphorus (P)) was detected with an inductively coupled plasma optical emission spectrometer (PerkinElmer, Waltham, MA, USA).

2.7. Statistical Analysis

All statistical calculations were performed using the Statistical Package for the Social Sciences (SPSS) software, version 23 (Armonk, New York, NY, USA). The results are reported as the mean ± standard deviation. The data were evaluated by *t*-tests with significance considered at $p < 0.05$.

3. Results

3.1. Inhibitory Effect of *L. antibioticus* HS124 on Growth of Fungal Pathogens

3.1.1. Chitinase Activity

The growth of *L. antibioticus* HS124 was low until five days post-inoculation (Figure 2) and exhibited a rapid increase six days post-inoculation. The maximum growth (5.67×10^7 CFU/mL) was

found seven days post-inoculation. Subsequently, the growth moderately decreased until the end of the inoculation period (Figure 2).

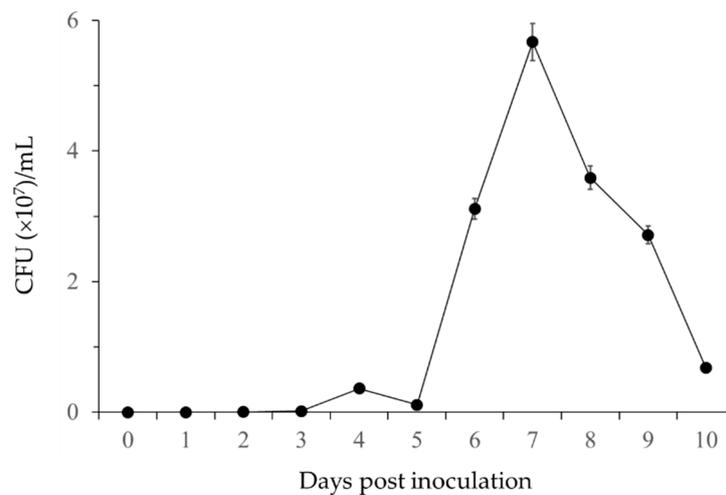


Figure 2. Cell growth curve of *L. antibioticus* HS124 in CY medium at 30 °C for 10 days. Error bars represent the standard deviation of three replications. CFU: colony forming unit.

The chitinase activity of *L. antibioticus* HS124 increased after two days and eventually reached a maximum value of 81.1 unit/mL over a period of four days (Figure 3). Thereafter, chitinase activity remained steady until eight days post-inoculation (Figure 3).

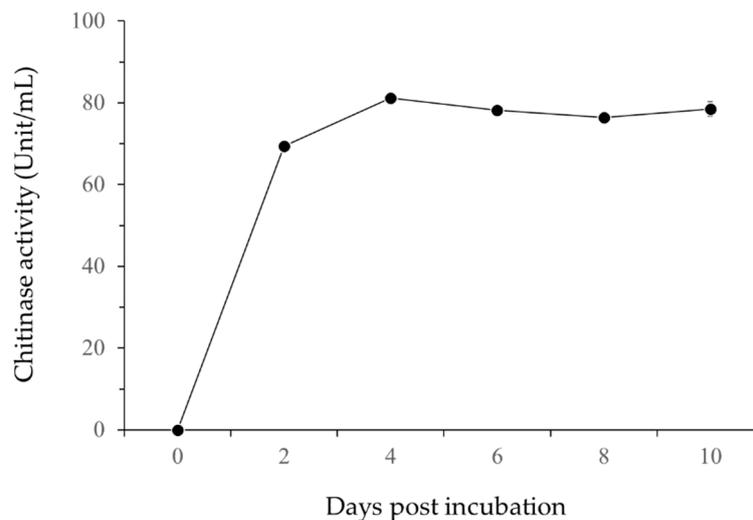


Figure 3. Chitinase activity of *L. antibioticus* HS124 cultured in CPD broth at 30 °C for 10 days. Quantitative measurement of chitinase enzymes produced by strain HS124 was done using a UV-spectrophotometer at 420 nm.

3.1.2. Growth Inhibition of Phytopathogenic Fungi by *L. antibioticus* HS124

The antifungal activities of *L. antibioticus* HS124 against different phytopathogenic fungi, including *B. cinerea*, *B. dothidea*, and *C. gloeosporioides*, were assayed in CY agar medium using the dual culture test (Figure 4). *L. antibioticus* HS124 showed the highest inhibition (27.25%) against *B. dothidea* and the lowest inhibition (20.56%) against *C. gloeosporioides* (Figure 4). Moreover, it showed 22.03% mycelial growth inhibition against *B. cinerea* (Figure 4).

The hyphal morphologies by *L. antibioticus* HS124 were abnormal, showing degradation, deformation, and lysis, compared to controls not inoculated with *L. antibioticus* HS124. The controls showed normal hyphal structures (Figure 5).

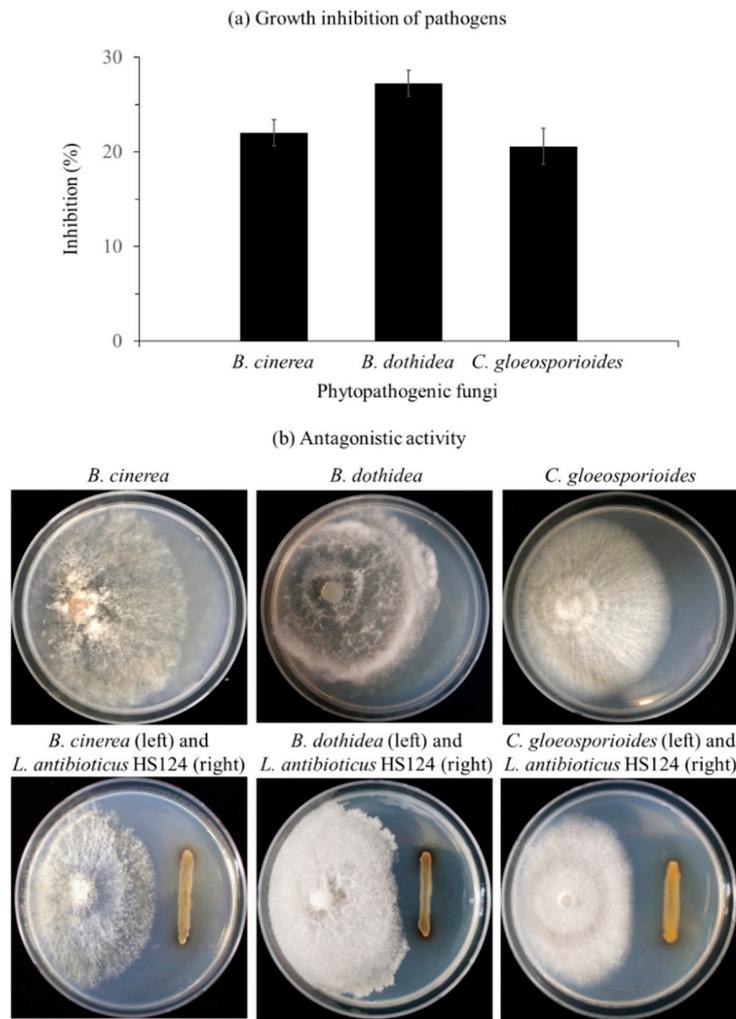


Figure 4. Inhibitory effect of *L. antibioticus* HS124 on mycelial growth of *B. cinerea*, *B. dothidea*, and *C. gloeosporioides* (a), and antagonistic activity of *L. antibioticus* HS124 against fungal pathogens (b) by the dual culture method.

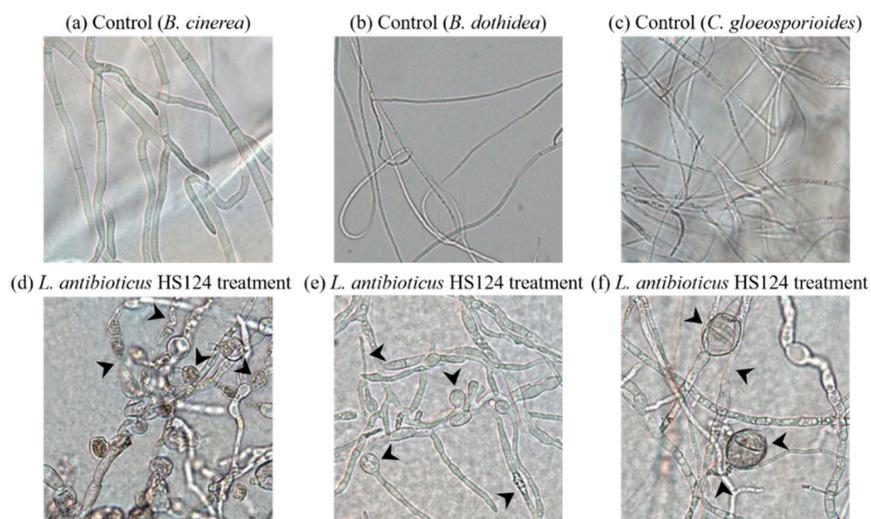


Figure 5. Deformed hyphal morphologies of *B. cinerea* (d); *B. dothidea* (e); and *C. gloeosporioides* (f) affected by *L. antibioticus* HS124 compared to *B. cinerea* control (a); *B. dothidea* control (b); and *C. gloeosporioides* control (c) under a light microscope. Arrows indicate hyphal alterations with swelling and bulbous structures caused by *L. antibioticus* HS124.

3.2. Effect of *L. antibioticus* HS124 on Fruit Yield

3.2.1. IAA Production

L. antibioticus HS124 produced auxin (Figure 6). The IAA concentration steadily increased for six days, eventually reaching a maximum value of 9.3 mg/L. Thereafter, the IAA concentration decreased rapidly (Figure 6).

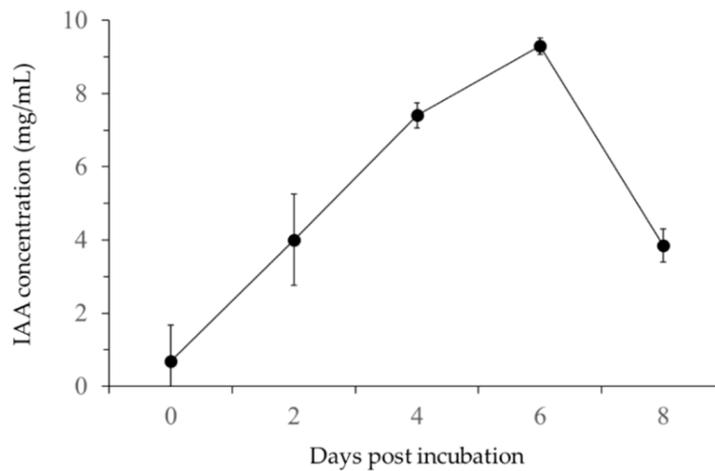


Figure 6. IAA (indole-3-acetic acid) production by *L. antibioticus* HS124.

3.2.2. Mineral Concentration, Characteristics, and Yield of Jujube Fruits

Potassium (K) was the most abundant mineral (2.12 g/kg) in the fruit, followed by P (0.21 g/kg) (Table 1). The fruit contained relatively low concentrations of Ca and Mg. A significant increase in the concentrations of K, Ca, Mg, and P was observed in the jujube fruits inoculated with *L. antibioticus* HS124 when compared to that of the control group (Table 1). The K and Mg concentrations in fruits with bacterial inoculation were 1.8- and 2.0-fold more than that of control group, respectively (Table 1). Moreover, the P and Ca concentrations in fruits that were inoculated with bacteria were 1.4- and 1.6-fold more than that of control group, respectively (Table 1).

Table 1. Mineral concentration, characteristics, and yields of jujube fruits in control and treatment groups with *L. antibioticus* HS124 inoculation in cultivated orchards.

Treatment	Fruit Mineral (g/kg)				Fruit Characteristics (mm)		Fruit Yield (g)
	K	Ca	Mg	P	Length	Diameter	
Control	1.18 ± 0.18 *	0.05 ± 0.01 *	0.04 ± 0.01 *	0.15 ± 0.02 *	29.66 ± 1.67 *	23.77 ± 1.24 *	2165.78 ± 221.19 *
Bacterial inoculation	2.12 ± 0.17 *	0.08 ± 0.01 *	0.08 ± 0.01 *	0.21 ± 0.03 *	38.37 ± 1.41 *	28.99 ± 1.53 *	6284.67 ± 1207.23 *

* A significant difference between treatments was observed by *t*-test at $p < 0.05$.

The length of the jujube fruits ranged from 35.39 to 40.85 mm with *L. antibioticus* HS124 inoculation and from 26.93 to 33.70 mm in the control without *L. antibioticus* HS124 inoculation (Table 1). The length of the fruits inoculated with bacteria was significantly higher than that of the control (Table 1). The average diameter of the jujube fruits ranged from 23.77 mm in the control to 28.99 mm with bacterial inoculation (Table 1). Regarding fruit characteristics of jujube cultivated in the orchard, the length and diameter of fruits inoculated with bacteria were significantly higher than those of the control fruits (Table 1).

The average yield of jujube fruit ranged from 2165.78 g/branch in the control to 6284.67 g/branch in trees inoculated with *L. antibioticus* HS124 (Table 1). A remarkable improvement in fruit yield was observed in treatment with *L. antibioticus* HS124 as compared to that of control (Table 1).

4. Discussion

Cell wall-degrading enzymes such as chitinase produced by antagonistic bacteria are known to play key roles in the suppression of phytopathogenic fungi [25–30]. In the present study, results of quantitative chitinase assay indicated that strain HS124 produced high levels of chitinase in colloidal containing medium (Figure 3). Therefore, the action of chitinase could be involved in antagonism against these fungal pathogens, including *B. cinerea*, *B. dothidea*, and *C. gloeosporioides* (Figures 4 and 5). In the presence of *L. antibioticus* HS124, the hyphae of phytopathogenic fungi showed abnormal morphologies, including swelling and bulb formation (Figure 5). Our results are in agreement with previous report of Won et al. [27,28], showing that chitinase enzymes produced by *Bacillus licheniformis* MH48 were involved in the control of diseases caused by fungi such as *Fusarium oxysporum*, *B. cinerea*, *Glomerella cingulate*, *Pestalotia diospyri*, and *Pestalotiopsis karstenii*. In a similar experiment, Brzezinska and Jankiewicz [43] showed that a chitinase secreted by *Aspergillus niger* LOCK62 had antifungal activities against various other fungi such as *Fusarium solani*, *Fusarium culmorum*, and *B. cinerea*. According to Velusamy and Kim [44], a chitinase secreted by *Enterobacter* sp. could decompose mycelia of *C. gloeosporioides* and *B. cinerea* and inhibit their growth. The antagonistic activity of *L. antibioticus* HS124 against phytopathogenic fungi resulted in a significant increase in the production of jujube fruits in cultivated orchards (a 2.9-fold increase) compared to the control (Table 1). Control jujube fruit and trees were easily infected by fungal diseases (Figure 1c), resulting in wilting and dropping of fruit during production. Fungal pathogen-infected fruits were discarded during harvesting (Figure 1b), thus decreasing the fruit production (Table 1). In particular, our field observations frequently found anthracnose disease which led to a reduction in the production of jujube fruit because of the increased flower wilting and fruit dropping. In addition, *L. antibioticus* HS124 produced auxin (Figure 6). Auxin acts at the time of cell division after fertilization of flowers [31,33,34], thus increasing the size of the fruit and improving the yield of trees inoculated with *L. antibioticus* HS124 (Table 1). PGPR also provides nutrients to the soil by fixing nitrogen from the atmosphere and solubilizing phosphorus in the soil [26,45]. Plants with increased absorption of nitrogen and phosphorus can promote photosynthesis and protein biosynthesis, thus increasing the size and yield of the fruit [46–49]. In cultivated orchard experiments, the size and yield of jujube fruits from trees inoculated with *L. antibioticus* HS124 were significantly increased (Table 1).

Well development in root system of trees are the most essential attribute for enhancing minerals and nutrient uptake [25,27,28,49]. Auxin produced by *L. antibioticus* HS124 inoculation might promote root development and stimulate the formation of absorbent root hairs and lateral roots [24,25,27,28,31,32]. This finding indicates that jujube trees inoculated with *L. antibioticus* HS124 could absorb more mineral elements such as K, Ca, Mg, and P in fruits (Table 1). Because PGPR including *Lysobacter* sp. can secrete organic acids and mineralize mineral elements such as K, Ca, Mg, Fe, and P from soil and enrich them in the soil [26,45,47,49], roots developed by auxin can promote the absorption of minerals and nutrient elements [24,25,27,32,48,50]. In addition, the photosynthesis of plants in the nitrogen-enhanced soils caused by nitrogen fixation from PGPR is enhanced, resulting in increased growth and mineral absorption in plants [26,45,48–52]. According to Ipek et al. [45] and Pirlak et al. [51] apples and strawberries with PGPR such as *Pseudomonas* sp., *Bacillus* sp., *Alcaligenes* sp., *Staphylococcus* sp., and *Agrobacterium* sp. can promote the photosynthesis caused by nitrogen fixation and stimulate the growth and contents of mineral elements. Results of the present study indicate that *L. antibioticus* HS124 is not only an effective biocontrol agent for phytopathogenic fungi, but also a beneficial agent for increasing the fruit mineral content and yield in jujube farms.

5. Conclusions

L. antibioticus HS124 could suppress fungal diseases caused by *B. cinerea*, *B. dothidea*, and *C. gloeosporioides* by producing lytic enzymes, including chitinase (Figures 3–5), thereby decreasing the fruit wilting and dropping during the production and harvest periods of cultivated jujube orchards (Figure 1c). The improvement in size and yield of jujube fruits in the present study could be associated

with the secretion of auxin like IAA by *L. antibioticus* HS124 (Table 1). Therefore, inoculating *L. antibioticus* HS124 to jujube trees should be considered as a potential way to control fungal diseases and increase the production of jujube fruits in a sustainable and ecological cultivation system of orchards.

Author Contributions: Conceptualization, funding acquisition and project administration, Y.-S.A.; investigation and experiments, J.-H.K., S.-J.W., and J.-H.M.; data analysis, J.-H.K. and S.-J.W.; resources, C.-W.K.; writing—original draft preparation, J.-H.K.; writing—review and editing, Y.-S.A.

Funding: This study was supported by the R&D program for Forest Science & Technology Projects (No. 2018122B10-1820-AB01) funded by the Korea Forest Service (Korea Forestry Promotion Institute). Additionally, this research was supported by a grant (No. 2018R1D1A1B07050052) of the National Research Foundation (NRF) of Korea under the Basic Science Research Program.

Acknowledgments: The authors would like to thank Kil-Yong Kim at the Chonnam National University for the technical assistance in the field and laboratory. The senior author also thanks Yun-Serk Park, CEO of Purne for their skillful assistance in analyzing the bacteria.

Conflicts of Interest: The authors declare no conflicts of interest.

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