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IN VITRO AND *IN VIVO* EVALUATION OF BACTERIAL ANTAGONISTS AGAINST TOMATO BACTERIAL SPOT DISEASE

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Abstract: This study evaluates the biocontrol potential of bacterial antagonists against *Xanthomonas vesicatoria* (Xv), the causative agent of bacterial spot disease in tomato, through both *in vitro* and *in vivo* assessments. Thirty bacterial isolates exhibiting epiphytic and endophytic traits were screened for antimicrobial activity against Xv. *In vitro* assays identified isolates 35E and 6E as the most effective, producing inhibition zones of 25.99±0.41 mm and 24.70±0.61 mm, respectively. *In vivo* trials demonstrated that isolate 35E consistently reduced disease severity from 40–41% in the positive control to approximately 19%, achieving stable suppression under *in vivo* conditions. Molecular characterization revealed that isolate 35E showed 96.56% 16S rDNA sequence similarity to *Bacillus cereus*, supporting its known biocontrol and plant growth-promoting properties. The results highlight the promise of *Bacillus cereus* CA18 as an effective biocontrol agent, offering a sustainable alternative to chemical pesticides for managing bacterial spot disease in tomato.

Keywords: Bacterial antagonists, Biocontrol, *Xanthomonas vesicatoria*, *Bacillus cereus*

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1. Introduction

Tomato (*Solanum lycopersicum* L.) is a warm-season, herbaceous plant in the Solanaceae family, closely related to potato, pepper and eggplant (Kimura and Sinha, 2008; Liu et al., 2022). Tomato is cultivated in open fields, greenhouses and other protected systems, under a broad range of climates from temperate to subtropical regions (Schwarz et al., 2014; Gatahi, 2020; Akotowanou et al., 2022). It is rich in vitamins (especially vitamin C), minerals, carotenoids such as lycopene, and other antioxidants that contribute to human health and diet quality (Quinet et al., 2019; Maurya et al., 2025). Production is geographically concentrated: a small number of countries (including China, the USA and Italy) account for most processing tomato output, and climate-change projections suggest that rising temperatures could reduce processing tomato production in major producing regions by around 6% by 2050 (Cammarano et al., 2022). Türkiye is consistently among the world's top tomato producers and has been listed within the 15 leading tomato-producing countries since at least the early 2000s (Peralta et al., 2006). Tomato varieties account for about 40% of Türkiye's total vegetable yield, underlining their central role in the national horticultural sector (Kayikcioglu et al., 2020).

Tomatoes suffer from diseases caused by fungi, bacteria and viruses leading to large yield losses. The primary pathological threats to tomato cultivation are diverse, encompassing fungal pathogens such as *Fusarium* spp. (wilt), *Phytophthora* spp. (late blight), *Botrytis cinerea* (gray mold), and *Alternaria* spp.: (early blight) (Panthee and Chen, 2010). Furthermore, viral agents like the tomato yellow leaf curl and spotted wilt viruses, alongside bacterial infections most notably *Xanthomonas* spp. (bacterial spot) constitute the major diseases affecting this crop (Havryliuk et al., 2024). To discuss *Xanthomonas* spp. in more detail, several Xanthomonas species (notably *X. euvesicatoria*, *X. vesicatoria*, *X. perforans*, *X. gardneri*) cause bacterial spot on tomato and pepper, a worldwide threat with necrotic lesions on leaves, stems, and fruit, defoliation, spotting, and fruit drop (Larrahondo-Rodríguez et al., 2022). Xanthomonas spreads efficiently via contaminated seed and seedlings, plant material, wind driven rain, and aerosols, and is favored by warm, humid conditions (Ketehouli et al., 2024). Regarding the control of this disease, copper based chemicals remain the main control tool, but field resistance is widespread, limiting efficacy. Breeding for resistance is complicated by high pathogen genetic diversity and rapid evolution of new races that overcome (Larrahondo-Rodríguez et al., 2022). Biological control using beneficial bacteria is a key alternative to chemical pesticides for managing plant diseases, including Xanthomonas-caused bacterial spot in tomato. The utilization of bacterial biocontrol agents in modern

agriculture relies on a complex interplay of several key mechanisms, primarily antibiosis via antimicrobial metabolites, competition for limited nutrients and space, quorum quenching, and the activation of induced systemic resistance (ISR) within the host plant. To ensure efficacy, various application methods are employed, including seed biopriming, soil inoculation, and foliar sprays, often as a core component of integrated pest management programs (Giovanardi et al., 2025). The practical application of bacterial biocontrol agents against Xanthomonas species in tomato cultivation is supported by diverse experimental evidence. For instance, seed biopriming and foliar spraying with *Pseudomonas putida* KD 91/1 have been shown to reduce *X. euvesicatoria* infections by up to 45% while simultaneously enhancing plant biomass (Eryigit et al., 2025). Similarly, specific strains of *Bacillus velezensis* strains 71 and *Paenibacillus peoriae* To99 exhibit potent disease control through the secretion of antimicrobial metabolites such as oxydifficidin and polymyxin A, effectively suppressing bacterial spot in both growth chamber and greenhouse environments (Olishevska et al., 2023). Recent research also highlights the efficacy of endophytic and phyllosphere derived bacteria. Endophytes like *Bacillus safensis* and *Pseudomonas lactis*, isolated from neem, have proven effective as both preventive and curative foliar treatments against *X. campestris* pv. *vesicatoria* (Fernandes et al., 2026). In addition, the metabolites of *Burkholderia gladioli* C101 significantly lower *X. perforans* severity in a dose-dependent manner (Shantharaj et al., 2021). Finally, microbiome profiling has identified Gluconobacter japonicus T12B, a phyllosphere resident, as a robust antagonist that utilizes organic acid production to combat *Xanthomonas* sp. in both *in vitro* and *in vivo* conditions (Teo et al., 2025).

Despite growing interest in biological control of bacterial spot disease, studies specifically evaluating epiphytic and endophytic bacterial isolates from existing culture collections against tomato bacterial spot disease remain limited. Furthermore, most published work focuses on a narrow range of well-known biocontrol agents, leaving the potential of less characterized isolates largely unexplored. This study aimed to evaluate the effectiveness of 30 bacterial antagonists selected from an established culture collection (Özaktan and Yay, 2022) as biocontrol agents against bacterial spot disease in tomatoes through *in vitro* and *in vivo* tests. To the best of our knowledge, this is one of the few studies to systematically screen epiphytic and endophytic isolates from an existing collection specifically against *Xanthomonas vesicatoria*, contributing to the identification of putative biocontrol candidates as sustainable alternatives to copper based chemical pesticides.

2. Materials and Methods

During the 2022–2023 study period, we selected 30 bacterial isolates exhibiting both epiphytic and endophytic traits from existing stocks at the Ege University Faculty of Agriculture. These specific strains belong to the collection originally cataloged in the study by Özaktan and Yay (2022) (Table 1).

Table 1. Information on the bacterial isolates used in this study and their characteristics

Isolate Code	Isolate No	Description
CA18	35E	Endophyte, Non-fluorescent, Gram (+)
2/9	55	Epiphyte, Non-fluorescent, Gram (+)
CB13	25E	Endophyte, Non-fluorescent, Gram (+)
CB18/2	37E	Endophyte, Non-fluorescent, Gram (-)
CB2/3	61E	Endophyte, Fluorescent, Gram (-)
MS180	117	Epiphyte, Fluorescent, Gram (-)
MS70	103	Epiphyte, Fluorescent, Gram (-)
CC37/2	83E	Endophyte, Non-fluorescent, Gram (-)
C9/1	25	Epiphyte, Non-fluorescent, Gram (-)
TR21/1	146	Epiphyte, Fluorescent, Gram (-)
KD13	6E	Endophyte, Non-Fluorescent, Gram (-)
CB36/1	80E	Endophyte, Non-fluorescent, Gram (-)
CC44	112E	Endophyte, Fluorescent, Gram (-)
CA41/1	99E	Endophyte, Non-fluorescent, Gram (+)
CB2/2	5E	Endophyte, Non-fluorescent, Gram (-)
30	41	Epiphyte, Fluorescent, Gram (-)
A506	102	Epiphyte, Fluorescent, Gram (-)
CB10	21E	Endophyte, Non-fluorescent, Gram (-)
66/3	93	Epiphyte, Non-fluorescent, Gram (+)
Eh 325	90	Epiphyte, Fluorescent, Gram (-)
CC30	68E	Endophyte, Fluorescent, Gram (-)

Table 1. Information on the bacterial isolates used in this study and their characteristics (continue)

Isolate Code	Isolate No	Description
CA33/2	73E	Endophyte, Fluorescent, Gram (-)
14/1y	30	Epiphyte, Fluorescent, Gram (-)
18/1 k	101	Epiphyte, Fluorescent, Gram (-)
CA28/1	56E	Endophyte, Non-fluorescent, Gram (-)
CC35/1	76E	Endophyte, Fluorescent, Gram (-)
MS109	149	Epiphyte, Fluorescent, Gram (-)
30/1 M	116	Epiphyte, Fluorescent, Gram (-)
CB9/3	19E	Endophyte, Non-fluorescent, Gram (-)
CB4	8E	Endophyte, Non-fluorescent, Gram (-)

2.1. *In vitro* Suppression of Xv by Bacterial Antagonists

The ability of the bacterial isolates to suppress Xv through antibiotic or lytic metabolite production was examined using a modified agar-based assay (Wang et al., 2022). Briefly, Petri dishes (9 cm) with KB medium were coated with 100 µl of a 24-hour Xv culture (standardized to 0.1 OD at 600 nm). After a 30-minute drying period, 24–48 h cultures of the antagonist strains were spotted onto the pathogen-laden medium. The plates were incubated for 48 h, after which the inhibition zones surrounding the colonies were measured (mm). The biocontrol potential was evaluated based on the mean diameters derived from three independent experimental trials.

2.2. *In vivo* Suppression of Xv by Bacterial Antagonists

In this study, the *in vivo* efficacy of two selected bacterial isolates and their combination was evaluated against the Xv pathogen. The experiment was established as a completely randomized design with four replicates, each consisting of one plant per treatment. The seed inoculation process involved soaking 1% sodium hypochlorite (NaOCl) sterilized tomato seeds in a bacterial carboxymethyl cellulose (CMC) (0.1%) mixture for 30 minutes, followed by a brief drying period at 24 °C. Once the seedlings reached the three-leaf developmental stage in sterile peat, they were transplanted (Lwin and Ranamukhaarachchi, 2006). For the challenge inoculation, the Xv provided from EUZF culture collection was adjusted to a concentration of 10^8 cfu/ml (OD_{600} = 0.35) and sprayed onto the foliage. Treatments were randomly assigned to experimental units prior to inoculation. Post-inoculation conditions were strictly regulated (25 °C, 16h light, 70% RH). Disease progression was monitored for three weeks, at which point severity was graded on a scale of 0 (healthy) to 4 (extensive necrosis/death) (Al-Dahmani et al., 2003). These categorical scores were transformed into percentage data via the Townsend–Heuberger formula. The reliability of the results was ensured by conducting the in vivo assays at least twice.

2.3. Molecular Classification of Superior Bacterial Strain Through 16S Sequencing Analysis

To extract DNA, bacterial culture grown on KB medium (24 °C) for 24–48 h was processed using the boiling lysis technique. Following the protocol described by Omar et al., (2014), bacterial pellets were obtained by centrifuging OD_{600} suspensions at 15,000×g. This was then heated at 100 °C for 10 min in 40 µL of ultrapure water and stored at −20 °C after a quick cooling and centrifugation step. For the subsequent Polymerase Chain Reaction (PCR) stage, the 16S rDNA region (approximately 1428 bp) was targeted using universal primers 27F and 1492R (Hodkinson and Lutzoni, 2009). The reaction was carried out in a mixture containing 1 U Taq DNA polymerase, 100 ng DNA, and 1.5 mM MgCl among other essential reagents. Thermal cycling involved 35 repetitions at an annealing temperature of 50 °C. Finally, the amplified 16S rRNA product was confirmed on 1.5% agarose gels, purified with a QIAquick kit, and sequenced by Medsantek, followed by BLASTn analysis for GenBank identification.

2.4. Statistical Analysis

Statistical processing was conducted in the R environment, specifically employing the 'agricolae' library (De Mendiburu and Simon, 2015). For the *in vitro* assay, a one-way analysis of variance (ANOVA) was performed on inhibition zone diameters obtained from triplicate *in vitro* tests. For the *in vivo* assay, each of the two independent trials was analyzed separately using one-way ANOVA, with four replicates per treatment in each trial. Where significant differences were found, mean values were separated using Duncan's multiple range test ($P<0.05$). All results are presented with their respective standard deviations (Genç and Soysal, 2018).

3. Results

3.1. *In vitro* Efficacy of Antagonistic Bacterial Isolates for the Biological Control of Xv

Analysis of variance revealed statistically significant differences ($P<0.05$) among the tested isolates in terms of their inhibition zone capacities at a 95% confidence level.

The highest antimicrobial activity was observed in isolates 35E (25.99±0.41 mm), 6E (24.70±0.61 mm), and 30 (24.60±0.47 mm), which demonstrated significantly superior performance compared to all other isolates ($P<0.05$). These were followed by isolates 19E

(18.81±2.01 mm) and 116 (18.69±2.02 mm), which showed the second-highest inhibitory activity. Isolates with moderate activity included 149 (18.51±0.97 mm), 146 (18.43±1.12 mm), 73E (16.65±0.69 mm), 101 (16.37±1.26 mm), and 5E (15.73±0.87 mm). A gradual decline in activity was observed across the remaining isolates: 103 (15.04±1.62 mm), 112E (14.87±0.96 mm), 76E (14.03±1.46 mm), 37E (13.59±0.29 mm), 90 (13.49±1.45 mm), 102 (12.93±0.53 mm), 41 (11.50±0.95 mm), 117 (11.38±0.42 mm), 68E (9.22±1.31 mm), and 25 (8.06±2.22 mm). The weakest inhibition values were recorded for isolates 99E (3.80±3.37 mm) and 83E (3.19±5.52 mm). Finally, isolates 21E, 25E, 55, 56E, 61E, 80E, 8E, and 93 showed no antimicrobial activity against the target pathogen under the experimental conditions (0.00±0.00 mm) (Figure 1).

3.2. *In vivo* Efficacy of Superior Antagonistic Bacterial Isolates for the Biological Control of Xv

In vivo biocontrol experiments were conducted in two independent trial sets to evaluate the suppressive effects of the isolates and their combinations on disease severity. Analysis of variance (ANOVA) indicated statistically significant differences among the treatments at a 95% confidence level (P<0.05), and the means were separated using Duncan's Multiple Range Test.

In the first *in vivo* trial, the highest disease severity was recorded in the positive control (41.33±12.17). Compared to the control, all treatment groups led to a reduction in disease severity. The 6E+35E combination (18.52±7.13) and isolate 35E (19.50±6.16) exhibited the strongest suppressive effect, significantly differing from the positive control (P<0.05). Isolate 6E (27.75±16.73) also reduced disease severity, though it did not differ significantly from either the control or the other treatment groups.

In the second *in vivo* trial, the positive control again showed the highest disease severity (40.50±15.45). The efficacy of individual isolate applications was more pronounced in this trial. Isolate 35E (19.00±5.47) and isolate 6E (22.66±8.23) significantly reduced disease severity compared to the positive control (P<0.05), representing the most effective treatments. Unlike the first trial, the 6E+35E combination (29.16±11.87) showed comparatively higher disease severity and did not differ significantly from either the control or the individual isolate treatments (Figure 2).

Overall, isolate 35E demonstrated the most stable biocontrol efficacy by consistently and significantly reducing disease severity across both independent trials, whereas isolate 6E and the combination treatment exhibited variable performance between experimental sets.

3.3. Results of Molecular Classification of Superior Bacterial Strain Through 16S Sequencing Analysis

The molecular characterization of the most effective bacterial strain with their corresponding NCBI BLAST analysis data is detailed in Table 2. Based on these sequence alignments, isolate 35E was most closely related to be *Bacillus cereus* CA18 (Table 2).

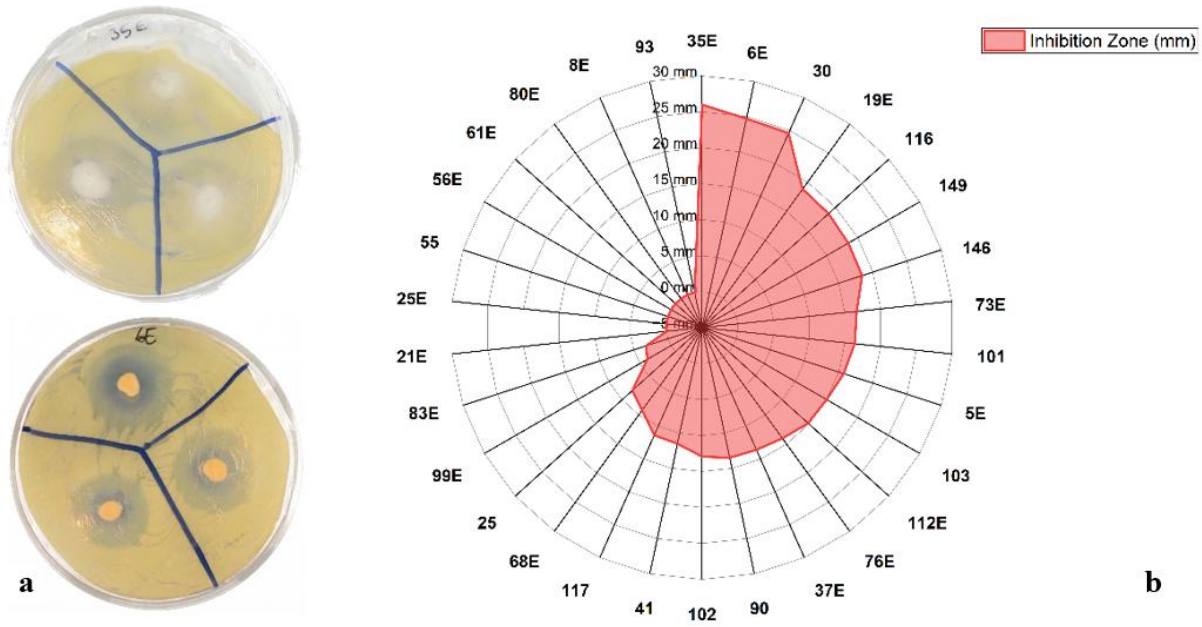

Figure 1. *In vitro* antagonistic activity of bacterial isolates against Xv a) Representative photographs showing inhibition zones formed around bacterial colonies on KB medium . b) Mean inhibition zone diameters (mm) of all tested isolates.

Table 2. Molecular identification of the most effective bacterial isolate based on 16S rDNA sequence similarity analysis using BLASTn against the NCBI GenBank database

Isolate Code	No	Results of Similarity in the NCBI database	Similarty Rate(%)	NCBI Accession Number
CA18	35E	*Bacillus cereus* strain N12	96.56	ON685200

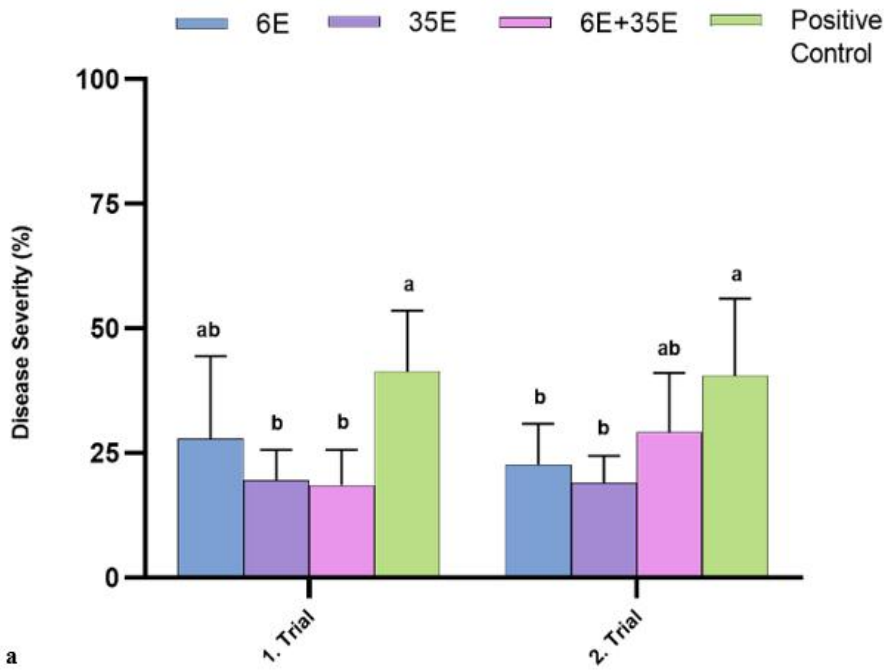

Figure 2. *In vivo* biocontrol efficacy of bacterial isolates against Xv on tomato plants. a) Mean disease severity (%) recorded across treatments in two independent trials. Values represent means±standard deviation. Different letters indicate statistically significant differences among treatments (Duncan's multiple range test, P<0.05). b) Representative photographs of disease symptoms observed on tomato plants treated with isolate 35E and positive control after three weeks post-inoculation.

4. Discussion

The present study evaluated the biocontrol potential of bacterial antagonists against *Xanthomonas vesicatoria* (Xv), the causative agent of bacterial spot disease in tomatoes, through *in vitro* and *in vivo* assessments. The results demonstrated that specific bacterial isolates, particularly 35E (*Bacillus cereus* CA18) and 6E, exhibited significant antagonistic activity against Xv, supporting their potential as effective biocontrol agents.

In vitro assays revealed considerable variability in antimicrobial efficacy among the tested isolates. Isolates 35E, 6E, and 30 showed the highest inhibition zones of 25.99±0.41 mm, 24.70±0.61 mm, and 24.60±0.47 mm, respectively, indicating strong production of antimicrobial substances or lytic metabolites capable of suppressing Xv growth. In contrast, eight isolates (21E, 25E, 55, 56E, 61E, 80E, 8E, and 93) showed no inhibitory activity (0.00±0.00 mm), while the weakest active

isolates, 99E and 83E, produced inhibition zones of only 3.80±3.37 mm and 3.19±5.52 mm, respectively. This finding aligns with previous research highlighting Bacillus species as prolific producers of antimicrobial compounds, such as lipopeptides and bacteriocins, which contribute to their biocontrol activity (Olishevska et al., 2023; Giovanardi et al., 2025). The complete lack of activity in several isolates underscores the importance of strain selection for effective biocontrol.

In vivo trials further confirmed the suppressive effects of 35E and 6E on bacterial spot severity. In the first trial, isolate 35E reduced disease severity from 41.33±12.17% in the positive control to 19.50±6.16%, while the 6E+35E combination achieved a similar reduction to 18.52±7.13%. In the second trial, isolate 35E again demonstrated strong suppression, reducing disease severity from 40.50±15.45% in the positive control to 19.00±5.47%, and isolate 6E reduced it to 22.66±8.23%. These results are consistent with findings reported by Eryigit et al. (2025), who demonstrated up to 45% reduction in *X. euvesicatoria* infections using *Pseudomonas putida* KD 91/1, further supporting the efficacy of bacterial biocontrol agents against Xanthomonas species.

Isolate 35E consistently demonstrated the most stable and significant disease reduction across both trials, reducing disease severity to approximately 19% compared to 40–41% in the positive control. This stability may be attributed to the well documented capacity of Bacillus species to form endospores, which confer resilience under variable environmental conditions, as well as their ability to colonize plant surfaces effectively and produce a broad spectrum of antimicrobial compounds, including lipopeptides and bacteriocins (Olishevska et al., 2023; Giovanardi et al., 2025). The variable performance of the 6E+35E combination treatment between the two trials is noteworthy. In the first trial, the combination achieved disease severity similar to isolate 35E alone (18.52±7.13%), whereas in the second trial it performed less effectively (29.16±11.87%). This inconsistency may reflect competitive interactions between the two isolates when applied together, potentially limiting the colonization or metabolite production of one or both strains. Similar antagonistic interactions between co-applied biocontrol strains have been reported in the literature, highlighting the complexity of designing effective microbial consortia (Giovanardi et al., 2025). Regarding the relationship between *in vitro* and *in vivo* tests, it is important to note that strong *in vitro* inhibition does not always translate directly into effective *in vivo* disease suppression. While isolates 35E, 6E, and 30 all showed similarly high inhibition zones in vitro, only 35E demonstrated consistently stable suppression *in vivo*. This discrepancy underscores the importance of conducting *in vivo* validation alongside *in vitro* screening, as plant microbe interactions, rhizosphere competition, and environmental conditions can significantly influence biocontrol efficacy under more complex biological settings. Several limitations of the present study should be acknowledged. Future studies should address these limitations through field trials. Such inconsistencies highlight the need for further optimization of application strategies, including formulation and timing, to maximize field performance.

Molecular identification most closely related to isolate 35E as *Bacillus cereus* CA18, a species well-documented for its biocontrol properties and plant growth-promoting traits. Isolate 35E, previously characterized as CA18 by Ozaktan et al. (2015), is a gram-positive bacterium known for its PGPR capabilities, such as IAA synthesis and siderophore-mediated inhibition of fungal pathogens such as *F. oxysporum* f.sp. *cucumerinum*. This identification supports the practical relevance of this isolate, as *Bacillus sp.* strains have been reported to suppress various phytopathogens through multiple mechanisms, including antibiosis, competition, and induced systemic resistance (ISR) activation (Giovanardi et al., 2025).

The integration of bacterial biocontrol agents into tomato disease management offers a promising alternative to traditional copper-based chemicals, which face challenges owing to pathogen resistance and environmental concerns (Larrahondo-Rodríguez et al., 2022). The observed effectiveness of the tested isolates under both *in vitro* and *in vivo* conditions supports their potential inclusion in integrated pest management (IPM) programs aimed at sustainable disease control.

Future research should focus on elucidating the specific antimicrobial compounds produced by these isolates, their modes of action, and their interactions with tomato microbiomes. Additionally, field trials are necessary to validate these findings under diverse environmental conditions and assess their long-term impact on disease suppression and crop yield. The exploration of synergistic effects among bacterial strains and optimization of delivery methods could further enhance the efficacy of biocontrol.

5. Conclusion

In conclusion, this study identified isolate 35E (*Bacillus cereus* CA18) as a promising bacterial antagonist against *Xanthomonas vesicatoria* under controlled experimental conditions. These findings provide a preliminary foundation for developing biocontrol strategies to manage bacterial spot disease in tomato cultivation. However, broader claims regarding its practical application in field settings should be approached with caution, as the current results are limited to greenhouse conditions. Field trials and further molecular and biochemical characterization are needed to fully validate the potential of isolate 35E as a sustainable alternative to copper based chemical pesticides in integrated pest management programs.

Author Contributions

The percentages of the author' contributions are presented below. The author reviewed and approved the final version of the manuscript.

	U.Ş.
C	100
D	100
S	100
DCP	100
DAI	100
L	100
W	100
CR	100
SR	100
PM	100
FA	100

C=concept, D= design, S= supervision, DCP= data collection and/or processing, DAI= data analysis and/or interpretation, L= literature search, W= writing, CR= critical review, SR= submission and revision, PM= project management, FA= funding acquisition.

Conflict of Interest

The author declared that there is no conflict of interest.

Ethical Consideration

Ethics committee approval was not required for this study because of there was no study on animals or humans.

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References

Akotowanou, O. C. A., Adjou, E. S., Olubi, A. B., Kougblenou, S. D., Ahoussi, E. D., & Sohounhloué, D. C. (2022). The tomato (Solanum lycopersicum L.) in community development: An overview focuscent achievements and future prospective. *International Journal of Frontiers in Biology and Pharmacy Research, 3*(2), 008–016. https://doi.org/10.53294/ijfbpr.2022.3.2.0061

Al-Dahmani, J. H., Abbasi, P. A., Miller, S. A., & Hoitink, H. A. (2003). Suppression of bacterial spot of tomato with foliar sprays of compost extracts under greenhouse and field conditions. *Plant Disease, 87*(8), 913–919. https://doi.org/10.1094/PDIS.2003.87.8.913

Cammarano, D., Jamshidi, S., Hoogenboom, G., Ruane, A. C., Niyogi, D., & Ronga, D. (2022). Processing tomato production is expected to decrease by 2050 due to the projected increase in temperature. *Nature Food, 3*(6), 437–444.

De Mendiburu, F., & Simon, R. (2015). Agricolae—Ten years of an open source statistical tool for experiments in breeding, agriculture and biology. *PeerJ PrePrints*. https://doi.org/10.7287/peerj.preprints.1404v1

Eryigit, G., Ozaktan, H., & Sanver, U. (2025). Pseudomonas putida has potential for biological control of bacterial spot of tomato, caused by Xanthomonas euvesicatoria. *Phytopathologia Mediterranea*, 64(2).

Fernandes, S., Bhoj, A., Kulkarni, P., Vaidya, P., Ranade, Y., & Sharma, P. (2026). Biocontrol activity of endophytic isolates Bacillus safensis and Pseudomonas lactis from Azadirachta indica against the pathogen Xanthomonas campestris pv. Vesicatoria in tomato plants. *Biotechnology and Applied Biochemistry, 73*(1), 302–314. https://doi.org/10.1002/bab.70023

Gatahi, D. M. (2020). Challenges and opportunities in tomato production chain and sustainable standards. *International Journal of Horticultural Science & Technology, 7*(3), 235-262. https://doi.org/10.22059/ijhst.2020.300818.361

Genç, S., & Soysal, M. İ. (2018). Parametric and nonparametric post hoc tests. Black Sea Journal of Engineering and Science, 1(1), 18-27.

Giovanardi, D., Biondi, E., Biondo, N., Quiroga, N., Modica, F., Puopolo, G., & Pérez Fuentealba, S. (2025). Sustainable and innovative biological control strategies against Pseudomonas syringae pv. Tomato, Pseudomonas savastanoi pv. Phaseolicola and Xanthomonas spp. Affecting vegetable crops: A review. *Frontiers in Plant Science, 16*, 1536152. https://doi.org/10.3389/fpls.2025.1536152

Havryliuk, L., Beznosko, I., Humennyi, D., Gentosh, D., & Bashta, O. (2024). Review of the main diseases of Solanum lycopersicum and methods of chemical control of pathogens. *Ukrainian Black Sea Region Agrarian Science, 28*(4), 32–40. https://doi.org/10.56407/bs.agrarian/4.2024.32

Hodkinson, B. P., & Lutzoni, F. (2009). A microbiotic survey of lichen-associated bacteria reveals a new lineage from the Rhizobiales. *Symbiosis, 49*(3), 163–180. https://doi.org/10.1007/s13199-009-0049-3

Kayikcioglu, H. H., Duman, İ., Asciogul, T. K., Bozokalfa, M. K., & Elmacı, Ö. L. (2020). Effects of tomato-based rotations with diversified pre-planting on soil health in the Mediterranean soils of Western Turkey. *Agriculture, Ecosystems & Environment, 299*, 106986. https://doi.org/10.1016/j.agee.2020.106986

Ketehouli, T., Pasche, J., Buttrós, V. H., Goss, E. M., & Martins, S. J. (2024). The underground world of plant disease: Rhizosphere dysbiosis reduces above-ground plant resistance to bacterial leaf spot and alters plant transcriptome. *Environmental Microbiology, 26*(7), e16676. https://doi.org/10.1111/1462-2920.16676

Kimura, S., & Sinha, N. (2008). Tomato (Solanum lycopersicum): A model fruit-bearing crop. *Cold Spring Harbor Protocols, 2008*(11), pdb-emo105.

Larrahondo-Rodríguez, E., Liao, Y.-Y., & Huerta, A. I. (2022). Diagnostic guide for bacterial spot of tomato and pepper. *Plant Health Progress, 23*(3), 355–361. https://doi.org/10.1094/PHP-11-21-0140-DG

Liu, W., Liu, K., Chen, D., Zhang, Z., Li, B., El-Mogy, M. M., Tian, S., & Chen, T. (2022). Solanum lycopersicum, a model plant for the studies in developmental biology, stress biology and food science. *Foods, 11*(16), 2402. https://doi.org/10.3390/foods11162402

Lwin, M., & Ranamukhaarachchi, S. (2006). Development of biological control of Ralstonia solanacearum through antagonistic microbial populations. *International Journal of Agriculture and Biology, 8*(5), 657–660.

Maurya, H. K., Lata, R., Sundar, S., Mitra, D., Singh, H., & Yadav, G. (2025). Scientific advances in tomato (Solanum lycopersicum L.) cultivation, agronomic innovation and genetic improvement: A comprehensive review. *Journal of Advances in Biology & Biotechnology, 28*(10), 781–803.

Olishevska, S., Nickzad, A., Restieri, C., Dagher, F., Luo, Y., Zheng, J., & Déziel, E. (2023). Bacillus velezensis and Paenibacillus

peoriae strains effective as biocontrol agents against Xanthomonas bacterial spot. *Applied Microbiology, 3*(3), 1101–1119. https://doi.org/10.3390/applmicrobiol3030076

Omar, B. A., Atif, H. A., & Mogahid, M. E. (2014). Comparison of three DNA extraction methods for polymerase chain reaction (PCR) analysis of bacterial genomic DNA. *African Journal of Microbiology Research, 8*(6), 598–602. https://doi.org/10.5897/AJMR2013.6459

Özaktan, H., & Yay, A. Ö. (2022). Domateste kök bakterilerinin bakteriyel benek hastalığı Etmeni pseudomonas syringae pv. Tomato ve kuraklık stresine karşı Etkisinin araştırılması. In *Ege 4. Uluslararası Uygulamalı Bilimleri Kongresi* (pp. 147–154).

Ozaktan, H., Çakır, B., Gül, A., Yolageldi, L., Akköprü, A., Fakhraei, D., & Akbaba, M. (2015). Isolation and evaluation of endophytic bacteria against Fusarium oxysporum f. Sp. Cucumerinum infecting cucumber plants*. Austin Journal of Plant Biology, 1*(1), 1003.

Panthee, D. R., & Chen, F. (2010). Genomics of fungal disease resistance in tomato. *Current Genomics, 11*(1), 30–39.

Peralta, I. E., Spooner, D. M., Razdan, M. K., & Mattoo, A. K. (2006). History, origin and early cultivation of tomato (*Solanaceae*). In M. K. Razdan & A. K. Mattoo (Eds.), *Genetic improvement of solanaceous crops* (Vol. 2, pp. 1–24). CRC Press. https://doi.org/10.1201/b10744-2

Quinet, M., Angosto, T., Yuste-Lisbona, F. J., Blanchard-Gros, R., Bigot, S., Martinez, J.-P., & Lutts, S. (2019). Tomato fruit development and metabolism. *Frontiers in Plant Science, 10*, 1554. https://doi.org/10.3389/fpls.2019.01554

Schwarz, D., Thompson, A. J., & Kläring, H.-P. (2014). Guidelines to use tomato in experiments with a controlled environment. *Frontiers in Plant Science, 5*, 625. https://doi.org/10.3389/fpls.2014.00625

Shantharaj, D., Williams, M., Potnis, N., & Liles, M. (2021). Burkholderia gladioli C101 metabolites protect tomato plants against Xanthomonas perforans infection. *Journal of Plant Diseases and Protection, 128*(2), 379–390. https://doi.org/10.1007/s41348-020-00416-9

Teo, K. W. E., Liew, Y. J. M., Mazumdar, P., Mohd Suhaimi, N. S., Chan, K.-G., & Chua, K.-O. (2025). Phyllosphere microbiome shifts reveal Gluconobacter japonicus T12B as a biocontrol agent against Xanthomonas in tomato through genomic and functional characterization. *Journal of Applied Microbiology, 136*(11), lxaf273. https://doi.org/10.1093/jambio/lxaf273

Wang, B., Li, L., Lin, Y., Shen, D., Shao, X., Zhong, C., & Qian, G. (2022). Targeted isolation of biocontrol agents from plants through phytopathogen co-culture and pathogen enrichment. *Phytopathology Research, 4*(1), 19. https://doi.org/10.1186/s42483-022-00124-2