J. Trop. Plant Pests Dis.
Vol. 24, No. 1, March 2024
Pages: 66–74

ISSN: 1411-7525
E-ISSN: 2461-0399
DOI : 10.23960/j.hptt.12466-74

RESEARCH PAPER

The endophytic potential, *Bacillus* spp. for controlling *Meloidogyne* sp. and increasing tomato growth and production

Winarto, Yulmira Yanti, Hasmiandy Hamid, & Yaherwandi

Manuscript received: 9 January 2023. Revision accepted: 18 November 2023. Available online: 20 February 2024.

ABSTRACT

Meloidogyne spp. is responsible of root swelling, one of the primary disease in tomato plants. Controlling this nematode is challenging due to its wide host range. The use of synthetic nematicides harms the environment; therefore, alternative controls, such as biological methods, are necessary. Among the biological agents, one group includes endophytic bacteria that reside in plant tissues and do not cause harm to plants. These bacteria enhance plant resistance to pests and pathogens while promoting plant growth. The study aimed to acquire endophytes, *Bacillus* spp. strains capable of controlling *Meloidogyne* sp. while stimulating the growth of tomato plants. The research employed a completely randomized design (CRD) comprising seven treatments and five replications. The treatments consisted of *B. cereus* strain SNE 2.2, TLE 2.3 and TLE 1.1, *B. pseudomycoides* strain EPL 1.1.3, *B. toyonensis* strain EPL 1.1.4, positive control (without the introduction of *Bacillus* spp. and inoculation with *Meloidogyne* sp.) and negative control (without *Bacillus* spp. and without *Meloidogyne* sp.). *Bacillus* spp. endophyites were introduced in two stages: into the seeds and into the roots of tomato seedlings for 15 min. The observed variables were the development of *Meloidogyne* sp., endophytic colonization of *Bacillus* spp., and plant growth. The results demonstrated that all *Bacillus* spp. were effective in controlling *Meloidogyne* sp. and enhancing the growth of tomato plants. The best isolate in controlling *Meloidogyne* sp. and increasing the growth of tomato plants was *B. cereus* strain SNE2.2.

Key words: *Bacillus* spp., endophyte, *Meloidogyne* sp., tomato

INTRODUCTION

The tomato plant is one of the essential horticultural commodities in Indonesia, serving as a source of minerals and vitamins (Kuklinsky-Sobral et al., 2004), possessing high economic value, and being cultivated commercially (Sikora et al., 2007). However, the attack of plant pest organisms leads to low productivity of tomatoes. *Meloidogyne* sp. is a crucial pest in tomato plants, causing root knot. This nematode exacerbates the severity of tomato wilt disease by *Ralstonia syzyggii* subsp. *Indonesiensis* (formerly *Ralstonia solanacearum*) (Pratiwi et al., 2020) and Fusarium wilt disease by *Fusarium oxysporum* (Wulandari et al., 2014). Nematodes are obligate parasites with multiple or polyphagous host plants. According to Sikora & Fernández (2005), damage by root-knot nematodes in several seasonal crops causes economic losses, such as 23–38% in tomatoes, 17–20% in eggplants, and 18–33% in melons. *Meloidogyne* sp. can release cellulose enzymes that hydrolyze cellulose; this breakdown of the cell wall's building blocks leads to cell wall damage and injury to the root cell tissue. Parasitism occurs when the nematodes move among cells to the area of cell elongation and begin feeding by injecting secretions from the esophageal glands into the root cells. This secretion induces physiological changes in the parasitized cells, resulting in galls (Siddiqui et al., 2014).

Nematode control using synthetic chemicals (nematicides) still plays an essential role because other control methods have not been able to provide satisfactory results. Nematode control methods using synthetic nematicides can cause negative impacts in the form of more resistant pathogens, killing beneficial natural enemies, disturbing ecosystem balance, and poisoning humans and pets (Yanti et al., 2019). For these reasons, alternative control is needed, including biological control by utilizing microorganisms such as the plant growth-promoting rhizobacteria (PGPR) group (Thokchom et al., 2017). Based on colonization, PGPR is grouped as follows: rhizobacteria, rhizoplane, and endophytes (Munif et al., 2000).

Endophytic bacteria can colonize plant tissue rapidly and reduce the chance of nematodes invading the same niche in the cortex. They secrete antibiotics or

Corresponding author:
Yulmira Yanti (yy.anthie79@gmail.com)

Plant Protection Departement, Faculty of Agriculture, Universitas Andalas, Padang, West Sumatra, 25163 Indonesia

stimulate the induction of plant resistance (Hallmann, 2001). The mechanism of endophytic bacteria in controlling nematodes can be in the form of direct antagonism by releasing secondary metabolites or an indirect mechanism through the induction of plant resistance in the form of induced systemic resistance (ISR) (Fitriani & Haryanti, 2016). According to Hallmann et al. (1997), ISR affects physiological processes in the roots, such as preventing the nematode feeding process, inhibiting the formation of feeding sites, and inhibiting the penetration and reproduction of nematodes.

Several researchers have also reported the utilization of *Bacillus* spp. to control *Meloidogyne* sp. Asyiah et al. (2015) reported that endophytic bacteria, *B. pumilus*, and *B. mycoides*, could suppress the *M. incognita* population and the number of galls on coffee plants by 33% and 39%, respectively. Sikora & Fernández (2005) used endophytic bacteria *Bacillus* sp. to control *M. incognita* on pepper plants. Furthermore, Rao et al. (2014) reported that *Bacillus* spp. could control *M. incognita* and decrease *F. oxysporum* the incidence in okra crops. In our previous research, we collected 5 strains of endophytic *Bacillus* spp. from tomato roots. The study aimed to obtain species of endophytic *Bacillus* spp. capable of controlling *Meloidogyne* sp. and increasing the growth of tomato plants.

MATERIALS AND METHODS

Research Site. This research was conducted at the Microbiology Laboratory, Plant Protection Department, and Experimental Garden, Faculty of Agriculture, Universitas Andalas, Padang, West Sumatra, Indonesia, from March to July 2021.

Research Methodology. The research employed a completely randomized design (CRD) comprising 7 treatments and 5 replications. The treatments consisted of *B. cereus* strain SNE 2.2, TLE 2.3 and TLE 1.1, *B. pseudomycoides* strain EPL 1.1.3, *B. toyonensis* strain EPL 1.1.4, positive control (without the introduction of *Bacillus* spp. and inoculation with *Meloidogyne* sp.), and negative control (without the introduction of *Bacillus* spp. and without being inoculated with *Meloidogyne* sp.).

Source of Endophytic *Bacillus* spp. and their Propagation. The source of *Bacillus* spp. is from West Sumatra, specifically from the districts of Agam, Solok, and Tanah Datar. *Bacillus* spp. endophytes propagated in liquid culture. A single colony of pure culture *Bacillus* spp. indigenous endophytes aged 2 × 24 hours was transferred into 25 mL of Nutrient Broth (NB) medium (0.5 g pepton, 0.5 g sodium chloride, 1000 mL distillate water) in a culture bottle (Scott bottle 0020 50 mL volume) and incubated on a rotary shaker for 24 hours at 150 rpm. Furthermore, 1 mL of pre-culture suspension was transferred to 49 mL of sterile coconut water (made by autoclaving for 20 min) in a culture bottle (Scott bottle 100 mL volume) for the main culture and incubated in the same way for 2 × 24 hours (Yanti et al., 2017). The endophytic bacterial suspension from the primary culture was determined for its population density based on a comparison with a McFarland scale of eight solutions (population density 10^8 cells/mL) (Ashoub & Amara, 2010).

A riphampicin mutant marker was used to track root colonization. Each *Bacillus* spp. was cultured on tryptic soy agar (TSA) (Himedia, USA) medium with Rifampicin levels of 0, 10, 20, 50, and 100 ppm, then incubated for 24–36 hours. The Rifampicin mutant marker was a mutant capable of growing at 100 ppm Rifampicin levels (Yanti et al., 2017). Colonies growing at 100 ppm Rifampicin were re-cultured on TSA medium and incubated for 24–36 hours, then the suspension was made with sterile distilled water (10^6 cells/mL).

Introduction of Endophytic *Bacillus* spp.. *Bacillus* spp. endophytes were introduced in 2 stages i.e seed treatment and root dipping treatment. For seed treatment, the tomato seeds (Warani variety) were surface sterilized with 2% NaOCl for 2 min, rinsed with distilled water, air-dried, and soaked in a suspension of *Bacillus* spp. endophytes with a population density of 10^8 cells/mL for 15 min. The seeds were then sown in 2 holes per pot tray. The seedlings were cultivated in a screen house for 3 weeks.

For root dipping, the roots of tomato seedlings were cleaned with running water and immersed in a suspension of *Bacillus* spp. endophytes with a population density of 10^8 cells/mL for 15 min (Yanti et al., 2017). Two tomato seedlings were then transplanted into a polybag with a diameter of 20 cm containing approximately 5 kg of sterile soil (65 % soil and manure 35 %). The tomato plants were cultivated at screen house.

Propagation of *Meloidogyne* spp.. The source of inoculum *Meloidogyne* sp. was obtained from tomato plants of the Warani cultivar, with roots showing symptoms of root knot nematodes in the field at Nagari Batu Palano, Agam Regency, West Sumatra. Groups of eggs from tomato roots were collected in a petri dish. The eggs were extracted from the infested

roots using 1% NaOCl. After extraction, the eggs were washed with tap water to remove the NaOCl (Hussey and Baker, 1973). A concentration of 1000 eggs/mL was inoculated onto 3-week-old tomato plants. The tomato plants were harvested 45 days after inoculation (DAI), and the egg groups were collected as a source of *Meloidogyne* sp. (Chawla et al., 2006).

Inoculation of *Meloidogyne* sp.. *Meloidogyne* sp. eggs are inoculated onto tomato plants (Warani variety) one week after planting (WAP) by sprinkling 10 mL of egg suspension (approximately 500 eggs) around the roots near the tomato stems (Harni & Samsudin, 2015).

Root Colonization of *Bacillus* spp.. Tomato seeds were introduced to mutant Rifampicin *Bacillus* spp. by soaking them for 5 min in bacterial suspension before planting them in sterile soil media. The roots of tomato seedlings were removed 9 days after introduction and 35 days after planting (DAP) (following *Meloidogyne* sp. inoculation). Tomato roots were sterilized by dipping them in 2% NaOCl and 2 min, followed by of distilled water, then macerated and diluted to 10^{-4}. Each suspension was homogenized, and 0.1 mL of the suspension was cultured on TSA media with 100 ppm Rifampicin content and incubated for 48 hours. The bacterial colonies growing on the media were counted based on morphological similarities with the mutants (Yanti et al., 2017).

Observation. The observed variable consist of: 1. The development of *Meloidogyne* sp. on tomato roots: a) Number of nematodes/300 g soil, b) Number of egg masses, c) Number of egg/egg mass; 2. The symptoms of *Meloidogyne* sp. on tomato: a) Number root-knots/plant, b) Number egg groups/plant, c) Amount of eggs in egg group, d) Number of nematodes in root/plant, e) Number of nematodes/300 g soil; 3. Colonization of root tissue of tomato plants introduced by *Bacillus* spp. mutant; 4. Growth and yield of tomato plants.

Data Analysis. All data obtained from pots experiments were analyzed using analysis of variance (ANOVA). The significant differences among treatments were determined according to the least significant differences (LSD) at $p< 0.05$ level of probability, using the CoStat software.

RESULTS AND DISCUSSION

The results showed that all isolates of *Bacillus* spp. introduced into seeds and roots of tomato plants significantly reduced the number of nematodes in the soil, egg mass, and egg/egg mass compared to control (Table 1). In terms of the number of nematodes, all treatments were not significantly different from each other but significantly different from the control. Four treatments were significantly different from the control in the number of nematodes, namely *B. cereus* strain SNE 2.2., *B. cereus* strain TLE 2.3, *B. pseudomycoides* strain EPL 1.1.4, and *B. cereus* strain TLE 1.1. In the total egg mass, all treatments were not significantly different from each other but significantly different from the control. *B. cereus* strain SNE 2.2 was the best treatment for reducing the number of nematodes, egg masses, and eggs/egg mass (6; 10; 274.67).

A comparison of diseased plants with nematodes and healthy plants can be seen in Figure 1, Figure 2, and Figure 3. Figure 1 shows the ratio of healthy roots

Table 1. *Meloidogyne* sp. development on tomato root that inoculated by endophytic *Bacillus* spp. 40 days after transplanting (DAT)

Treatments	Number of nematodes*		Number of egg mass		Number of egg/egg mass	
	Nematode*	Reduced (%)	Egg mass	Reduced (%)	Pieces	Reduced (%)
B. toyonensis strain EPL 1.1.3	18.00 ab	74.52	12.00 b	52.00	303.00	41.09
B. pseudomycoides strain EPL 1.1.4	16.33 b	76.88	10.33 b	58.66	336.00	34.67
B. cereus strain SNE 2.2	6.00 bc	91.51	10.00 b	60.00	274.67	67.46
B. cereus strain TLE 1.1	17.66 b	75.00	11.66 b	53.33	289.33	43.75
B. cereus strain TLE 2.3	15.66 b	77.84	11.00 b	56.00	514.33	55.67
Control	70.66 a	0.00	25.00 a	0.00	0.00	0.00

Mean values with different letters within each column denote significant ($p< 0.05$) differences between groups.
*Number of nematode/300 g soil.

to diseased tomato plants, indicating that the roots with endophytic *Bacillus* spp. treatments are healthier than those in the negative control. Figure 2 displays plants with endophytic *Bacillus* spp. treatments that are healthier than those in the negative control at 14 DPI. It can be observed that the plants treated with endophytic *Bacillus* spp. are healthier than those in the negative control. Figure 3 compares plants in each treatment 7 WAP, illustrating that *B. cereus* strain SNE2.2 is the most effective treatment for tomato plants attacked by *Meloidogyne* sp. compared to the positive control.

Tomato plants treated with *Bacillus* spp., both in the seeds and roots, demonstrated suppression in the development of swollen roots/plant, the number of egg groups/plant, the number of eggs in the egg clusters, the number of nematodes in the roots per plant, and the number of nematodes per soil compared to the negative controls (Table 2). Among the 5 isolates of *Bacillus* spp., each exhibited a distinct effect compared to the control. *B. cereus* strain SNE 2.2 proved to be the most effective isolate in controlling *Meloidogyne* sp. in tomato plants at 40 days after transplanting (DAT).

Root colonization by Rifampicin mutant of endophytic *Bacillus* spp. was assessed at 9 DAT and 35 DAT (after *Meloidogyne* sp. inoculation) (Table 3). It can be observed that on day 35 after being inoculated with *Meloidogyne* sp., all treatments resulted in a reduction the number of nematodes in root tissue. *B. toyonensis* strain EPL 1.1.3 exhibited the most effective colonization of root tissue after *Meloidogyne*

Figure 1. Comparison of tomato plant roots attacked by *Meloidogyne* sp. A. After being treated with *B. cereus* strain SNE 2.2; B. Negative control. (1) 45 DAI; (2) 60 DAI.

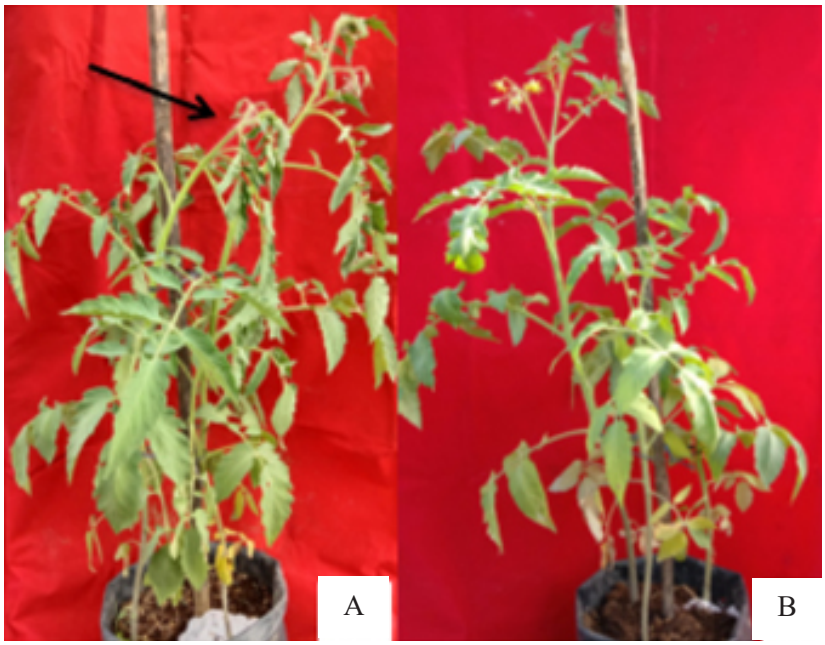

Figure 2. Comparison of diseased symptom (arrow) by nematodes infection on 14 DAI. A. Negative control; B. *B.cereus* strain SNE 2.2 treated and *Meloidogyne* sp. infected tomato roots (healthy).

sp. inoculation on tomato plants, with counts of 9.06 × 10^5 CFU/g at 9 DAI and 4.85 × 10^5 CFU/g at 35 DAI.

Tomato plants treated with *Bacillus* spp. on seeds and roots exhibited enhanced growth compared to the control group (Table 4). Among the treatments, tomatoes treated with *B. cereus* strain SNE 2.2 intruduced tomatoes showed higher than other treatments. Regarding the number of leaves, all treatments did not significantly differ from each other but were significantly different from the negative control. *B. cereus* strain SNE 2.2 resulted in the highest leaf count, with 15 leaves per plant. Furthermore, apart from promoting plant height and leaf number, the treatments also led to increased tomato plant weight. All treatments showed significant differences compared to the control group. Notably, *B. cereus* strain SNE

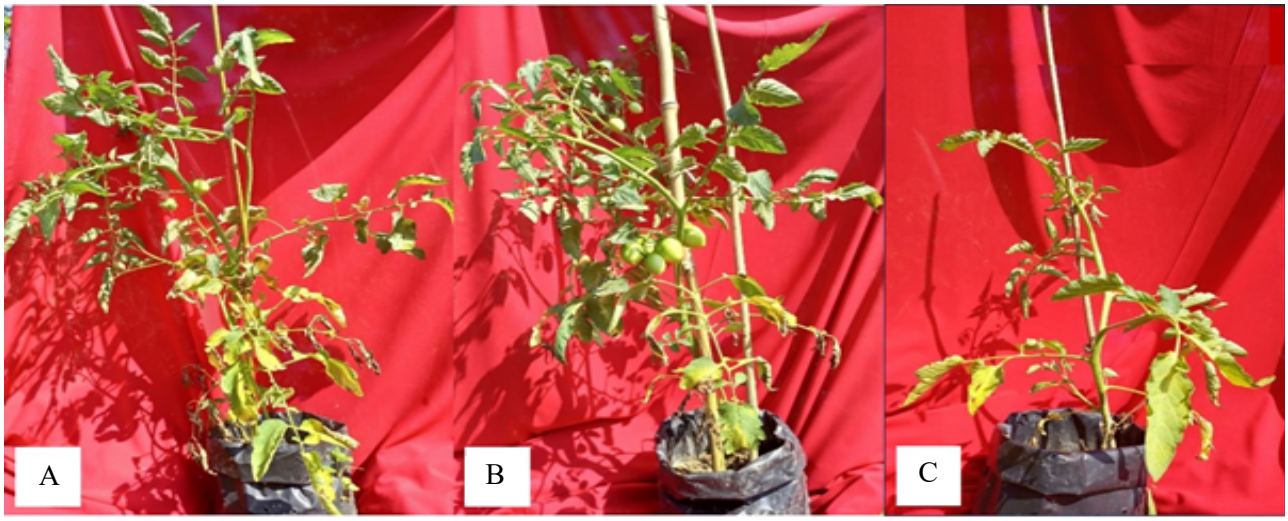

Figure 3. Comparison of appearance of tomato plants. A. *Bacillus cereus* strain SNE 2.2.; B. *Bacillus cereus* strain TLE 2.3; C. Positive control.

Table 2. Introduction of *Bacillus* spp. endophytes in controlling *Meloidogyne* sp. in tomato plants (40 DAT)

Treatments	Root-knot number/ plant	Egg group number/ plant	Amount of egg in egg group (grain)	nematodes number in root/plant	Nematodes number/300 g soil
Negative control	73.00 a	25.00 a	514.33 a	70.66 a	83.33 a
B. toyonensis strain EPL 1.1.3	37.33 b	12.00 b	289.33 bcde	18.00 ab	33.66 b
B. cereus strain TLE 1.1	30.66 b	11.66 b	274.67 cde	17.66 b	30.33 b
B. cereus strain TLE 2.3	26.33 b	11.00 b	253.00 de	16.33 b	28.66 b
B. pseudomycoides strain EPL 1.1.4	25.66 b	10.33 b	235.33 e	15.66 b	26.66 b
B. cereus strain SNE 2.2	7.66 b	10.00 b	0.0 f	6.00 bc	5.33 b
Positive control	0.0 c	0.0 c	0.0 f	0.0 c	0.0 c

*Mean values with different letters within each column denote significant ($p < 0.05$) differences between groups. DAT= day after transplanting.

Table 3. Colonization of root tissue of tomato plants introduced by *Bacillus* spp. mutant

Treatments	Root tissue colonization (CFU/g)	
	9 DAI*	35 DAI*
B. pseudomyciodes strain EPL 1.1.4	6.22 × 10^5	2.02 × 10^5
B. toyonensis strain EPL 1.1.3	9.06 × 10^5	4.85 × 10^5
B. cereus strain TLE 2.3	5.82 × 10^5	3.25 × 10^5
B. cereus strain SNE 2.2	5.06 × 10^5	2.35 × 10^5
B. cereus strain TLE 1.1	6.42 × 10^5	3.06 × 10^5

*Inoculated with *Meloidogyne* sp.

2.2 exhibited the most effective enhancement in fruit weight, with an average of 303.33 g/plant or 20.24 tons/ha, representing an increase of 213.81%.

The results demonstrated that *Bacillus* spp. endophytes were more effective in controlling *Meloidogyne* sp. compared to other biocontrol agents. *B. cereus* strain SNE2.2 exhibited the highest efficacy in controlling *Meloidogyne* sp. *Bacillus* spp. can produce secondary metabolites that aid in controlling *Meloidogyne* sp.. According to Ashoub & Amara (2010), the genus *Bacillus* can produce volatile compounds or metabolites, such as the chitinase enzyme, which can kill nematode larvae and eggs. Fang & Ramasamy (2015) noted that the chitinase enzyme produced by *Bacillus* spp. assists in the degradation of chitin present in the eggshell of nematodes, thereby interfering with the hatching of nematode eggs. *Bacillus* spp. can indirectly induce plant resistance by increasing salicylic acid, phytoalexins, peroxidases, PR proteins, and phenolic compounds (Miljaković et al., 2020). Induction of resistance affects physiological processes such as preventing the nematode feeding process, inhibiting the formation of feeding sites, and impeding nematode penetration and reproduction (Suryaningsih, 2008). The number of nematode egg groups influences the number of eggs that will hatch into nematode larvae. Sturz et al. (2000) found that endophytic bacteria isolated from tomato plants could suppress the formation of root cavities (*Meloidogyne* spp.) in tomato plants. Harni (2014) reported that endophytic bacterial filtrate acts as a toxin for nematode eggs, preventing them from hatching. The percentage of suppression of egg hatching of root lesion nematodes (*P. brachyurus*) on patchouli was 48.5–74.6% compared to controls two weeks after application.

The low nematode population in the roots can be observed from the staining of the roots of the infected tomato plants, which indicates the presence of nematodes in the roots. This reduction is attributed to the inhibition of nematode development in the roots. *Bacillus* spp. indigenous endophytes can induce plant resistance to suppress the development and reproduction of nematodes in plant tissues. Tian et al. (2022) reported that *Bacillus velezensis* strain Bv-25 exhibits significant potential as a biocontrol agent against *Meloidogyne incognita*, demonstrating both strong nematicidal activity and the ability to induce resistance in cucumber plants, leading to substantial benefits in field conditions.. Sikora et al. (2007) used endophytic bacteria *Bacillus* sp. to control *M. incognita* on pepper plants.

Bacillus spp. could colonize plant root tissue before and after *Meloidogyne* sp. inoculation on tomato plant roots. The tomato plants that were not treated with *Bacillus* spp. exhibited a higher level of suppression against *Meloidogyne* sp. compared to the treated plants. Kuklinsky-Sobral et al. (2004) stated that endophytic bacteria found on the host plant and can be isolated again have the potential to re-colonize the root tissue of the same host plant. Mekete et al. (2009) reported that antibiotics produced by *Pseudomonas* could suppress the *Pratylenchus loosi* population with a suppression rate of 63.10% to 95.24%. Additionally, Harni (2014) reported that the filtrate of endophytic bacteria, *B. subtilis, P. putida*, and *Achromobacter xylosoxidans* produced nematicide compounds that were effective in killing the nematode, *P. brachyurus*. The killing power of the filtrate is thought to be related to the compound 2,3 diacetylphorologlucinol and proteases, which inhibit egg hatching and kill nematodes (Kuklinsky-Sobral et al., 2004).

This study proved that the suppression of *Meloidogyne* sp. applied by *Bacillus* spp. endophytes as biocontrol agents increased plant height in the generative phase and plant yields. *B. cereus* strain SNE2.2 was the best treatment for increasing tomato

Table 4. Tomato plant growth that is introduced with endophytes, *Bacillus* spp. 40 days after plantation

Treatments	Plant height (cm)	Amount leaf (stem)	Weight fruit (g)	Production (t/ha)
B. cereus strain SNE 2.2	67.78	15.00 a	303.33 a	20.24
B. cereus strain TLE 2.3	57.86	14.60 a	296.67 a	19.77
B. pseudomycoides strain EPL 1.1.4	57.64	13.80 ab	273.33 ab	18.23
B. toyonesis strain EPL 1.1.3	53.64	12.20 abc	226.67 abc	15.09
B. cereus strain TLE 1.1	52.26	12.00 abc	223.33 abc	14.89
Negative control	49.16	10.80 bc	190.00 abcd	12.69
Positive control	41.86	9.40 c	96.66 d	6.41

*Mean values with different letters within each column denote significant ($p < 0.05$) differences between groups.

plant growth (Table 4). *Bacillus* spp. endophytes act as biological control agents for *Meloidogyne* sp., causing root swelling, and can also act as PGPR, producing growth hormone in the form of IAA for tomato plants. According to research by Habazar et al. (2021), *B. cereus* strain RBI2AB2.1 and B. subtilis strain RBIBPL 2.3 can control *Meloidogyne* spp. on tomatoes. Furthermore, Hrynkiewiezc & Baum (2012) described the bacteria *Bacillus* spp. as a growth-promoting bacterium (PGPR) that can increase plant growth in various ways, including increasing nutrition, producing phytohormones, and suppressing the development of pathogens. According to Khan et al. (2022), *Bacillus* spp. can produce phytohormonal compounds such as auxins, cytokinins, ethylene, gibberellins, and abscisic acid, stimulating plant growth and ultimately impacting crop yields. The findings indicated that various Bacillus species exhibited efficacy in managing Meloidogyne sp. and promoting the growth of tomato plants. Among the isolates studied, B. cereus strain SNE2.2 stood out as the most effective in both controlling Meloidogyne sp. and fostering the growth of tomato plants.

CONCLUSION

All *Bacillus* spp. endophytes could control *Meloidogyne* sp. and increase the growth of tomato plants. *B. cereus* strain SNE 2.2 was the best results in controlling *Meloidogyne* sp. and increasing tomato plant growth.

ACKNOWLEDGMENTS

The author would like to thank Andalas University for funding this research under Decree Number 561/UN16.R/XII/KPT/2021 and Agreement/ Contract Number T/2/UN.16.17/PP.Pangan-PTU KRP2GB-Unand/ LPPM/2021.

FUNDING

Andalas University funded this research under Decree Number 561/UN16.R/XII/KPT/2021 and Agreement/Contract Number T/2/UN.16.17/ PP.Pangan-PTU KRP2GB-Unand/ LPPM/2021.

AUTHORS' CONTRIBUTIONS

W and YY considered and planned the experiment. W carried out the propagation of *Meloidogyne* spp. and inoculation. Y carried out the propagation and the application of *Bacillus* spp. HH was collecting observation data. Y performing analysis data. HH prepared the manuscript. The authors provided responses and comments on the research flow, data analysis, interpretation, and the shape of the manuscript. All the authors have read and approved the final manuscript.

COMPETING INTEREST

Authors are required to declare any competing interests, such as financial or non-financial interests, professional or personal relationships that are directly or indirectly connected to the work submitted for publication. If there is no competing interest regarding your publication, you are also required to declare.

REFERENCES

Ashoub AH & Amara MT. 2010. Biocontrol activity of some bacterial genera against root-knot nematode, *Meloidogyne incognita. J. Am. Sci.* 6(10): 321–328.

Chawla G, Singh KP, & Srivastava AN. 2006. Study on propagation of root knot nematode, *Meloidogyne incognita* (Kofoid & White) Chitwood, through tuberose (*Polianthes tuberosa* Linn.) bulbs. *Journal of Ornamental Horticulture*. 9(1): 73–74.

Fang Y & Ramasamy RP. 2015. Current and prospective methods for plant disease detection. *Biosensors* (Basel). 5(3): 537-561. https://doi.org/10.3390/bios5030537

Fitriani HP & Haryanti S. 2016. Pengaruh penggunaan pupuk nanosilika terhadap pertumbuhan tanaman tomat (*Solanum lycopersicum)* var. bulat [Effect of nanosilica fertilizer on plant growth of tomato (*Solanum lycopersicum*) var. bulat]. *Buletin Anatomi dan Fisiologi*. 24(1): 34–41.

Habazar T, Winarto, Obel, Yanti Y, Dani MR, & Monica D. 2021. Biocontrol of *Meloidogyne* sp. on tomato plants by selected *Bacillus* spp. *IOP Conf. Ser.: Earth Environ. Sci*. 757: 012019. https://doi.org/10.1088/1755-1315/757/1/012019

Hallmann J, Quadt-Hallmann A, Mahaffee WF, & Kloepper JW. 1997. Bacterial endophytes in agricultural crops. *Can. J. Microbiol.* 43(10): 895–914. https://doi.org/10.1139/m97-131

Hallmann J. 2001. Plant interactions with endophytic

bacteria. In: Jeger MJ & Spence NJ (Eds.). *Biotic Interactions in Plant Pathogen Associations.* pp. 87–119. CABI Publishing, Wallingford. https://doi.org/10.1079/9780851995120.0087

Harni R & Samsudin. 2015. Pengaruh formula bionematisida bakteri endofit *Bacillus* sp. terhadap infeksi nematoda *Meloidogyne* sp. pada tanaman kopi [Effect of endophytic bionematicide *Bacillus* sp. on the infection of *Meloidogyne* sp. of coffee plants]. *J. TIDP*. 2(3): 143–150.

Harni R. 2014. Prospek penggunaan bakteri endofit untuk pengendalian nematoda *Pratylenchus brachyurus* pada tanaman nilam [Prospects for using endophytic bacteria to control *Pratylenchus brachyurus* nematode in patchouli plants]. *Perspektif: Review Penelitian Tanaman Industri*. 13(1): 1–12.

Hrynkiewiezc K, & Baum C. 2012. The potential of rhizosphere microorgamisms to promote the plant growth in disturbed soils. In: Malik A., & Grohmann (Eds). *Environmental protection strategies for sustainable development.* USA

Hussey RS & Barker KR. 1973. A comparison of methods of collecting inocula of *Meloidogyne* spp., including a new technique. *Plant Disease Reporter*. 57(12): 1025–1028.

Khan AR, Mustafa A, Hyder S, Valipour M, Rizvi ZF, Gondal AS, Yousuf Z, Iqbal R, & Daraz U. 2022. *Bacillus* spp. as bioagents: uses and application for sustainable agriculture. *Biology*. 11(12): 1763. https://doi.org/10.3390/biology11121763

Kuklinsky-Sobral J, Araújo WL, Mendes R, Geraldi IO, Pizzirani-Kleiner AA, & Azevedo JL. 2004. Isolation and characterization of soybean-associated bacteria and their potential for plant growth promotion. *Environ. Microbiol*. 6(12): 1244–1251. https://doi.org/10.1111/j.1462-2920.2004.00658.x

Mekete T, Hallmann J, Kiewnick S, & Sikora R. 2009. Endophytic bacteria from Ethiopian coffee plants and their potential to antagonise *Meloidogyne incognita. Nematology*. 11(1): 117-127. https://doi.org/10.1163/156854108X398462

Miljaković D, Marinković J, & Balešević-Tubić S. 2020. The significance of *Bacillus* spp. in disease suppression and growth promotion of field and vegetable crops. *Microorganisms.* 8(7): 1037. https://doi.org/10.3390/microorganisms8071037

Munif A, Hallmann J, & Sikora RA. 2000. Evaluation of the biocontrol activity of endophytic bacteria from tomato against *Meloidogyne incognita. Proceedings of the Mededelingen Faculteit Landbouwkundige En Toegepaste Biologische Wetenschappen*, Vol. 65. pp. 471–480. Universiteit Gent. Belgium.

Pratiwi NWK, Amrulloh R, Auly FE, & Kurniawati F. 2020. Deteksi dan identifikasi nematoda puru akar (*Meloidogyne* spp.) pada tanaman bit menggunakan metode DNA barcoding [Detection and identification root-knot nematode (*Meloidogyne* spp.) in sugar beet using DNA barcoding method]. *Jurnal Fitopatologi Indonesia*. 16(1): 1–8. https://doi.org/10.14692/jfi.16.1.1-8

Rao MS, Dwivedi MK, Kumar RM, Chaya MK, Rathnamma K, Rajinikanth R, Grace GN, Priti K, Shree NV, Kamalnath M, Prabu P, Krishna CG, Rini P, & Shivananda TN. 2014. Evaluation of bio-efficacy of *Bacillus subtilis* (NBAIMCC-B-01211) against disease complex caused by *Meloidogyne incognita* and *Fusarium oxysporum* f.sp. *vasinfectum* in okra. *Pest Manage. Hortic. Ecsyst*. 20(2): 217–221.

Siddiqui ZA, Shehzad M, & Alam S. 2014. Interactions of *Ralstonia solanacearum* and *Pectobacterium carotovorum* with *Meloidogyne incognita* on potato. *Arch. Phytopathol. Plant Prot*. 47(4): 449–455. https://doi.org/10.1080/03235408.2013.811810

Sikora RA & Fernández E. 2005. Nematode parasites of vegetables. In: Luc M, Sikora RA, & Bridge J (eds). *Plant Parasitic Nematodes in Subtropical and Tropical Agriculture*. 2nd Edition. pp 319-392. CABI Publishing, Wallingford. https://doi.org/10.1079/9780851997278.0319

Sikora RA, Schäfer K, & Dababat AA. 2007. Modes of action associated with microbially induced in planta suppression of plant-parasitic nematodes. *Australas. Plant Pathol*. 36(2): 124–134. https://doi.org/10.1071/AP07008

Sturz AV, Christie BR, & Nowak J. 2000. Bacterial endophytes: Potential role in developing sustainable systems of crop production. *Crit. Rev. Plant Sci.* 19(1): 1–30. https://doi.org/10.1016/S0735-2689(01)80001-0

Suryaningsih E. 2008. Pengendalian penyakit sayuran yang ditanam dengan sistem budidaya pada pertanian periurban [Disease control method for several vegetables planted in mosaic farming system in periurban agriculture]. *J. Hort.* 18(2): 200–211.

Tian XL, Zhao XM, Zhao SY, Zhao JL, & Mao ZC. 2022. The biocontrol functions of *Bacillus velezensis* strain Bv-25 against *Meloidogyne incognita*. *Front. Microbiol*. 13: 843041. https://doi.org/10.3389/fmicb.2022.843041

Thokchom E, Thakuria D, Kalita MC, Sharma CK, & Talukdar NC. 2017. Root colonization by host-specific rhizobacteria alters indigenous root endophyte and rhizosphere soil bacterial communities and promotes the growth of mandarin orange. *Eur. J. Soil Biol*. 79: 48–56. https://doi.org/10.1016/j.ejsobi.2017.02.003

Wulandari D, Sulistyowati L, & Muhibuddin A. 2014. Keaneakaragaman jamur endofit pada tanaman tomat (*Lycopersicum esculentum* Mill.) dan kemampuan antagonisnya terhadap *Phytophthora infestans* [Diversity of endophytic fungi in tomato (*Lycopersicum esculentum* Mill.) plants and their antagonistic ability against *Phytophthora infestans*]. *Jurnal HPT (Hama Penyakit Tumbuhan).* 2(1): 110–118.

Yanti Y, Habazar T, & Resti Z. 2017. Formulasi padat rhizobakteria indigenus *Bacillus thuringiensis* TS2 dan waktu penyimpanan untuk mengendalikan penyakit pustul bakteri *Xanthomonas axonopodis* pv. *glycines* [Solid formulation of indigenous rhizobacteria *Bacillus thuringiensis* TS2 and storage time to control bacterial pustule disease *Xanthomonas axonopodis* pv. *glycines*]. *J. Trop. Plant Pests Dis*. 17(1): 9–18. https://doi.org/10.23960/j.hptt.1179-18

Yanti Y, Hamid H, & Reflin R. 2019. Indigenous rhizobacteria screening from tomato to control *Ralstonia syzigii* subsp. *indonesiensis* and promote plant growth rate and yield. *J. Trop. Plant Pests Dis.* 18(2): 177–185. https://doi.org/10.23960/j.hptt.218177-185