HortScience 60(1):140–145. 2025. https://doi.org/10.21273/HORTSCI18267-24

Comparative Performance of Biorational and Chemical Products for the Management of Bacterial Blight on Lilac

Cansu Oksel, Christina Jennings, Prabha Liyanapathirenage, Madhav Parajuli, Kumuditha D. Hikkaduwa Epa Liyanage, Terri Simmons, and Fulya Baysal-Gurel

Department of Agricultural Sciences and Engineering, Tennessee State University, Otis L. Floyd Nursery Research Center, McMinnville, TN 37110, USA

Keywords. disease management, lilac, *Pseudomonas syringae* pv. *syringae*, woody ornamental

Abstract. **Bacterial blight caused by *Pseudomonas syringae* pv. *syringae* is a common disease of lilac (*Syringa reticulata* L.). This bacterium spreads plant to plant via insect vectors, wind, rain, and contaminated tools; hence, managing this disease is challenging. This study reports the efficacy of biological and chemical products for managing bacterial blight on lilac 'Ivory Silk'. All plants except the noninoculated, nontreated control plants were spray inoculated with *P. syringae* pv. *syringae* strain FBG0464 (10^8 cfu·mL^{-1}). Pydiflumetofen 6.9% + difenoconazole 11.5% (Postiva), thyme oil [Proud 3 (5.56%) and Tril-21 (15.5%) (preventive application)] and *Bacillus amyloliquefaciens* F272 (Stargus) were applied preventively 3 days before bacterial inoculation. Copper octanoate (Camelot O) (curative application) treatment was applied on the same day as the bacterial inoculation. Didecyl dimethyl ammonium chloride (KleenGrow) and thyme oil [Tril-21 (15.5%) (curative application)] were applied 7 days after inoculation. The plants were assessed weekly for bacterial blight, defoliation, and phytotoxicity. The experiments were conducted in 2021, 2022, and 2023. The final mean disease severity on inoculated, nontreated plants was 22.0%, 63.8%, and 64.0% in 2021, 2022, and 2023, respectively. All treatments significantly reduced bacterial blight compared with the inoculated, nontreated control. In 2021, plants treated with pydiflumetofen 6.9% + difenoconazole 11.5%, *B. amyloliquefaciens* F272 and thyme oil [Proud 3 (5.56%) and Tril-21 (15.5% as curative application)] had significantly lower bacterial blight severity compared with the rest of the treatments. In 2022, there were no significant differences in disease severity among the treatments. In 2023, plants treated with copper octanoate had the lowest disease severity. There was no phytotoxicity or defoliation in any of the treated plants. These findings can aid in developing suitable solutions for controlling bacterial blight caused by *P. syringae* pv. *syringae* on lilac.**

Lilac (*Syringa* L.), a member of the Oleaceae family, is a deciduous flowering shrub commonly grown in East Asia, Europe, and North America (Hongxia et al. 2004; Toth et al. 2015; Varga et al. 2019). Lilac has decorative yellowish-white, lilac, purple, and pink flowers blooming in April and May and large green leaves (Gasecka et al. 2023; Kosiada 2016; Yiğit et al. 2022). In 2019, lilac contributed $20.5 million in total sales in the United States (National Agriculture Statistics Service 2020). Bacterial blight (*P. syringae* pv. *syringae*) is a common disease of lilac, which has been reported to cause significant economic losses on lilac (Scheck et al. 1996).

The bacterial blight pathogen *P. syringae* pv. *syringae* is the most harmful plant pathogenic bacteria, which can cause disease in more than 180 plant species (Islam et al. 2020; Lamichhane et al. 2015; Little et al. 1998). The pathogen spreads from infected tissues to healthy plants primarily through wind, rain splash, and insects (Tattar 1989). It enters the plant via natural openings and wounds, usually on leaves or shoots, and causes brown spots on stems (Tattar 1989) and necrotic spots surrounded by yellow halos on leaves (Keshtkar et al. 2016). Leaf and shoot infections may cause dieback and death blossoms (Tattar 1989).

P. syringae pv. *syringae* has epiphytic and endophytic phases (Hirano and Upper 2000). The epiphytic phase includes growth and survival on seemingly healthy blossoms and leaves throughout the growing season. During the summer, the pathogen will cause leaf spot lesions and survive epiphytically on outwardly healthy leaf surfaces (Hirano and Upper 2000). The epiphytic phase provides easy dispersal of the pathogen and an immediate source of inoculum for disease (Hirano et al. 1994; Lamichhane et al. 2014). For example, the pathogen is already present in intercellular spaces of leaves as the epiphytic phases, and the population size grows in response to rain that further increases the probability of brown spot disease occurrence. For endophytic phases, bacteria can colonize leaf scars and move systemically to new sites to form new colonies. It can lead to dead-bud, blossom blight, and canker symptoms (Kennelly et al. 2007; Sundin et al. 1988). As such, the initial epiphytic populations of *P. syringae* pv. *syringae* on plant surfaces can serve as good predictors of their subsequent endophytic populations within plant tissues and potential disease outbreaks under favorable environmental conditions (Rouse et al. 1985; Xin et al. 2018).

Cooler temperatures and frequent rain favor the development of *P. syringae* pv. *syringae* infection (Hirano and Upper 2000). Severe spring epidemics have often been reported after cool and wet weather periods or after frost (Kennelly et al. 2007; Latorre et al. 2002; Nejad et al. 2004). Warmer and/or cooler temperatures enable the epiphytic pathogen population to switch to an endophytic phase population, which will systematically colonize the dormant buds where the pathogen overwinters. In the following spring, dormant and visually healthy but already infected buds can serve as the source of inoculum for blossom colonization (Crosse 1956; Lamichhane et al. 2014). In addition, winter frost damage has resulted in weakened plants, which has been shown to consistently correlate with an increased incidence of infection by *P. syringae* pv. *syringae* (Kennelly et al. 2007).

Management of the bacterial blight caused by *P. syringae* pv. *syringae* includes some challenges due to the lack of effective chemical or biological control measures, the epiphytic and endophytic life cycle of the pathogen, and copper resistance strains. Common strategies for controlling bacterial blight caused by *P. syringae* pv. *syringae* predominantly relies on copper compounds (Kennelly et al. 2007; Scheck and Pscheidt 1998) or other heavy metals, fungicides, and disinfectants (Chase 1986). Copper compounds are routinely applied to reduce *P. syringae* pv. *syringae* populations, thereby preventing infections of tree crops, including woody plants (Aiello et al. 2015; Cazorla et al. 2002; Scheck and Pscheidt 1998). Previously, copper compounds have been reported to reduce the epiphytic population of *P. syringae* pv. *syringae* and have shown moderate disease control in several hosts (Gilardi et al. 2010; Wimalajeewa et al. 1991). However, many strains of

Received for publication 15 Oct 2024. Accepted for publication 5 Nov 2024.
Published online 30 Dec 2024.
This project was funded by the IR-4 Project (US Department of Agriculture-National Institute of Food and Agriculture award number 2021-34383-34848). We thank Huma Gro, Kemin Industries Inc., Marrone Bio Innovations, Pace 49 Inc., SePRO, and Syngenta for donating the conventional fungicides and biorational products used in this study.
Mention of trade names of commercial products in the publication is solely for the purpose of providing specific information and does not imply recommendation or endorsement by Tennessee State University.
F.B.-G. is the corresponding author. E-mail: fbaysalg@tnstate.edu.

P. syringae pv. *syringae* isolated from woody ornamentals and fruit trees exhibited copper resistance (Aiello et al. 2015; Cazorla et al. 2002; Renick et al. 2008; Scheck et al. 1996; Spotts and Cervantes 1994). The presence of leaf-surface biofilms has been found to potentially shield bacteria in the epiphytic stage from the effects of copper exposure, consequently making the treatments ineffective (Lamichhane et al. 2015; Morris and Monier 2003). This led us to find effective and sustainable alternative management strategy to control *P. syringae* pv. *syringae*. The biological control method is known to reduce the toxicity and maintain eco-friendly environmental conditions (Islam et al. 2020). For instance, *Bacillus* spp., *Pseudomonas* spp., and *Streptomyces* spp. as biological control agents are reported to be effective against various plant pathogens including *P. syringae* pv. *syringae* (Doolotkeldieva and Bobusheva 2020; Gilardi et al. 2010; Völksch and Weingart 1998). In recent years, multiple studies have demonstrated the effectiveness of natural extracts from various plants in managing strains in the *P. syringae* species (Balestra et al. 2009; Simonetti et al. 2019). However, there is a lack of specific studies on controlling *P. syringae* pv. *syringae* on lilac. The absence of comparative treatments for lilac makes it difficult to predict the efficacy of chemical and biological control agents in managing *P. syringae* pv. *syringae*. Implementing effective treatments could aid in controlling the pathogen and allow for the evaluation of the effectiveness of copper compounds, fungicides, and biological products known for their ability to broaden the spectrum against *P. syringae* pv. *syringae*. The objective of this study was to evaluate the efficacy of biological and chemical products for controlling *P. syringae* pv. *syringae* on lilacs.

Materials and Methods

Inoculum preparation. Isolate FBG0464 of *P. syringae* pv. *syringae* (GenBank accession: PP229537) isolated from lilac was obtained from a culture maintained at the Baysal-Gurel laboratory at the Tennessee State University Otis L. Floyd Nursery Research Center in McMinnville, TN, USA. The isolate was grown on King's B agar (King et al. 1954) for 2 to 3 d at 28 °C. Single colonies were transferred into nutrient broth and grown overnight at 28 °C on an orbital shaker (Thermo Scientific MaxQ 2000 CO2, Waltman, MA, USA) at 150 rpm. Cultures were then centrifuged (Thermo Scientific Sorvall Legend X1R) and optical density was adjusted to 10^8 cfu·mL^{-1} (OD600:0.2).

Treatments. The treatments included copper octanoate (Camelot O), didecyl dimethyl ammonium chloride (KleenGrow), pydiflumetofen + difenoconazole (Postiva), thyme oil [Proud 3 (5.56%) and Tril-21 (15.5%)] and *B. amyloliquefaciens* F727 (Stargus) (Table 1).

Experimental design. The experiments were performed in a shade house (under 56% shade) at the Tennessee State University Otis L. Floyd Nursery Research Center in McMinnville, TN, USA. Three-year-old container-grown lilac 'Ivory Silk' (*Syringa reticulata*) plants were used in the experiments. Plants were fertilized with 10 g of 18N–6P–8K Nutricote controlled-release granular fertilizer (Florikan E.S.A. LLC, Sarasota, FL, USA) and 7.5 g of Miracle-Gro® water soluble all-purpose plant food (Scoot's Miracle-Gro Products, Inc., Marysville, OH, USA) was mixed in 1 L of water and applied to lilac plants (200 mL per plant) on 14 May 2021, 10 May 2022, and 10 May 2023. The experiments were conducted from 4 Jun to 8 Jul 2021 as trial 1, 14 Jun to 15 Jul 2022 as trial 2, and 9 Jun to 24 Jul 2023 as trial 3. The average maximum temperatures were 33.7 and 32.7 °C for trial 1, 31.8 and 32.3 °C for trial 2, 28.3 and 30.5 °C for trial 3; average minimum temperature was 11.9 and 14.1 °C for trial 1, 18.3 and 20.6 °C for trial 2, 16.1 and 19.4 °C for trial 3; and total rainfall amounts were 57.5 mm and 9.9 mm for trial 1, 27.8 mm and 8.1 mm for trial 2, 72.1 mm and 3.2 mm for trial 3. Plants were arranged in a completely randomized design with five single-plant replications.

Plants were preventatively treated with pydiflumetofen + difenoconazole (Postiva), *B. amyloliquefaciens* F727 (Stargus), thyme oil [Proud 3 (5.56%) and Tril-21(15.5%) (preventative application)] 3 d before *P. syringae* pv. *syringae* inoculation. Plants were treated with copper octanoate (Camelot O) on the same day as inoculation of *P. syringae* pv. *syringae*. Treatments and *P. syringae* pv. *syringae* inoculation suspension were applied as a foliar spray with a backpack CO_2 pressurized sprayer equipped with a TeeJet XR8002VS at 30 psi and plants were sprayed to runoff. After pathogen inoculation, plants were covered for 24 h with a transparent plastic bag to maintain a humidity-saturated environment. After 7 d of *P. syringe* pv. *syringae* inoculation, plants were treated with didecyl dimethyl ammonium chloride (KleenGrow) and thyme oil [Tril-21 (15.5%)] as a curative application. Treatments were applied at 3-, 7-, 10-, and 14-d intervals (Table 1).

Plants were irrigated using emitters (Orbit 55032 $^1/_2$-inch BRS Sprinkler Head; Orbit® Inc., North Salt Lake, UT, USA) for 15 min twice a day during the experiments. Average maximum temperatures were 33.7, 31.8, and 28.3 °C; average minimum temperatures were 11.9, 18.3, and 16.1 °C; and the total rainfall was 57.1, 27.85 and 72.1 mL, for the duration of trial 1, trial 2, and trial 3, respectively.

Recording plant growth and bacterial blight disease. The initial plant height and width were recorded on 5 May 2021, 9 Jun 2022, and 9 Jun 2023. Final height and width were recorded on 8 Jul 2021, 15 Jul 2022, and 24 Jul 2023. Plant height measurements were taken from the base of the stem at the substrate level to the top of the terminal bud on the main stem. The plant width was measured as the average of the widest part from leaf tip to leaf tip and the width perpendicular to the widest part. The increase in plant height or width was calculated by subtracting the initial height/width from the final height/width. Bacterial blight was evaluated by assessing the percentage of infected leaves with small, round spots that started as light brown and became necrotic (Monchiero et al. 2015). The scale used ranged from 0% to 100% leaves affected by *P. syringae* pv. *syringae*. Defoliation was determined by counting the number of primary leaves missing from each node on the main stem, divided by the total number of nodes per main stem × 100 (Guan and Nutter 2002). Plants were evaluated weekly for disease severity and defoliation until 8 Jul 2021, 15 Jul 2022, and 24 Jul 2023.

Statistical analysis. The area under the disease progress curve (AUDPC) was calculated

Table 1. Details of biological and chemical products used for this study.

Treatment (active ingredient)	Trade name	Application rate	Application date[i]	Application type	Manufacturer
Bacillus amyloliquefaciens F727 (96.4%)	Stargus	10 mL·L^{-1}	1, 4, 6	Preventative	Marrone Bio Innovations, Davis, CA, USA
Copper octanoate (10%)	Camelot O	10 mL·L^{-1}	2, 4, 6	Curative	SePRO Corporation, Carmel, IN, USA
Didecyl dimethyl ammonium chloride (7.5%)	KleenGrow	4 mL·L^{-1}	4, 6	Curative	PACE 49 Inc, Delta, B.C., Canada
Pydiflumetofen (6.9%) + difenoconazoledifenoconazole (11.5%)	Postiva	2.18 mL·L^{-1}	2, 4, 6	Curative	Syngenta Crop Protection LLC, Greensboro, NC, USA
Thyme oil (5.56%)	Proud 3	10 mL·L^{-1}	1, 3, 4, 5, 6	Preventative	Huma Gro, Gilbert, AZ, USA
Thyme oil (15.5%)	Tril-21 (PathoCURB)	5 mL·L^{-1}	1, 3, 5	Preventative	Kemin Industries Inc, Des Moines, IA, USA
Thyme oil (15.5%)	Tril-21 (PathoCURB)	5 mL·L^{-1}	4, 6	Curative	Kemin Industries Inc, Des Moines, IA, USA

[i]Application date: 1 = 3 d before *Pseudomonas syringae* pv. *syringae* inoculation, 2 = same day with *Pseudomonas syringae* pv. *syringae* inoculation, 3 = 3 d after *Pseudomonas syringae* pv. *syringae* inoculation, 4 = 7 d after *Pseudomonas syringae* pv. *syringae* inoculation, 5 = 10 d after *Pseudomonas syringae* pv. *syringae* inoculation, 6 = 14 d after *Pseudomonas syringae* pv. *syringae* inoculation.

Table 2. Biological and chemical products' effects on final bacterial blight (*Pseudomonas syringae* pv. *syringae*) severity (0% to 100% affected), and AUDPC of lilac (*Syringa reticulata*) grown in a shade house, McMinnville, TN, USA.

Treatments	Application date[i]	Final severity (%)			AUDPC[ii]		
		2021	2022	2023	2021	2022	2023
Bacillus amyloliquefaciens F727	1, 4, 6	4.5 ± 0.9 d[iii]	16.3 ± 3.8 bc	12.0 ± 0.5 bc	113.75 ± 18.94 bcd	224.88 ± 34.95 b	203.00 ± 15.99 b
Copper octanoate	2, 4, 6	10.5 ± 1.2 b	13.8 ± 4.3 bc	9.0 ± 1.3 d	145.25 ± 27.21 bc	211.75 ± 54.90 b	229.25 ± 43.70 b
Didecyl dimethyl ammonium chloride	4, 6	8.0 ± 2.0 c	12.5 ± 3.2 bc	10.0 ± 1.1 cd	165.90 ± 33.16 b	147.88 ± 26.21 bc	164.50 ± 28.50 b
Pydiflumetofen + difenoconazole	2, 4, 6	2.5 ± 0.0 d	24.4 ± 11.9 bc	10.0 ± 1.1 cd	61.95 ± 7.12 de	285.69 ± 120.48 b	162.75 ± 21.36 b
Thyme oil (5.56%)	1, 3, 4, 5, 6	4.0 ± 1.0 d	21.3 ± 5.5 b	13.0 ± 0.9 b	91.00 ± 10.53 cd	231.00 ± 60.40 b	222.25 ± 19.68 b
Thyme oil (15.5%)	1, 3, 5	7.5 ± 1.1c	12.5 ± 1.4 bc	11.5 ± 0.6 bc	113.75 ± 15.01 bcd	166.25 ± 8.75 bc	187.25 ± 9.82 b
Thyme oil (15.5%)	4, 6	4.0 ± 0.6 d	11.3 ± 2.4 bc	11.0 ± 0.6 bcd	88.90 ± 16.67 cd	154.88 ± 40.63 bc	211.75 ± 18.02 b
Inoculated, nontreated control		22.0 ± 1.2 a	63.8 ± 6.6 a	64.0 ± 1.9 a	357.00 ± 33.93 a	859.25 ± 93.06 a	1249.50 ± 72.91 a
Noninoculated, nontreated control		1.0 ± 0.0 e	1.9 ± 1.2 c	0.5 ± 0.5 e	17.50 ± 4.43 e	27.56 ± 17.03 c	12.25 ± 12.25 c
P value		<0.0001	<0.0001	<0.0001	<0.0001	<0.0001	<0.0001
F		35.46	8.21	193.18	20.65	15.01	127.99

[i] Application date: 1 = 3 d before *Pseudomonas syringae* pv. *syringae* inoculation, 2 = same day with *P. syringae* pv. *syringae* inoculation, 3 = 3 d after *P. syringae* pv. *syringae* inoculation, 4 = 7 d after *P. syringae* pv. *syringae* inoculation, 5 = 10 d after *P. syringae* pv. *syringae* inoculation, 6 = 14 d after *P. syringae* pv. *syringae* inoculation.

[ii] AUDPC: $\Sigma([(x_i + x_{i-1})/2](t_i - t_{i-1}))$; where xi is the bacterial blight rating at each evaluation time and $(t_i - t_{i-1})$ is the number of days between evaluations.

[iii] Means followed by a different lowercase letter within a column are significantly different ($P \leq 0.05$). One-way analysis of variance was used to evaluate treatment effects on area under the disease progress curve (AUDPC). Means were compared using Fisher's least significant difference test with an $\alpha = 0.05$. Final disease severity was analyzed according to general linear mixed model with a logit link and beta distribution (PROC GLIMMIX).

according to the formula: $\Sigma([(x_i + x_{i-1})/2](t_i - t_{i-1}))$; where xi is the bacterial blight rating at each evaluation time and $(t_i - t_{i-1})$ is the number of days between evaluations. The effects of biological and chemical product treatments on plant height and width increase and AUDPC were analyzed using one-way analysis of variance with PROC GLM procedure (SAS Inc., Cary, NC, USA). Means were separated using Fisher's least significant difference test ($\alpha = 0.05$). The final disease severity and defoliation were analyzed using a general linear mixed model with a logit link and beta distribution (using PROC GLIMMIX). Means were separated using LS means.

Results

In trial 1, all treatments significantly reduced final disease severity and AUDPC compared with the inoculated, nontreated control plants had 22.0% of final disease severity (Table 2). Lilac treated with pydiflumetofen + difenoconazole, *B. amyloliquefaciens* F727, and thyme oil [5.56% and 15.5% (curative application)] had significantly lower final disease severity compared with plants treated with didecyl dimethyl ammonium chloride, copper octanoate, and thyme oil (15.5%, preventative application). Pydiflumetofen + difenoconazole-treated plants had the lowest AUDPC. There were no significant differences in terms of plant height increment among the chemical and biological products, inoculated, nontreated control and noninoculated, nontreated control plants (Table 3). However, the plants treated with pydiflumetofen + difenoconazole showed a significantly lower width increase compared with the thyme oil (15.5%, preventive and curative application) and inoculated, nontreated control.

In trial 2, inoculated, nontreated plants had the highest final disease severity of 63.8% (Table 2). All treatments significantly reduced disease severity and AUDPC compared with the inoculated, nontreated control plants. There were no differences between treatments other than inoculated, nontreated control. There were no significant differences in terms of plant height and width increment among the treatments and inoculated, nontreated control plants (Table 3).

In trial 3, the final disease severity was 64% on inoculated, nontreated control plants (Table 2). All treatments significantly reduced disease severity and AUDPC compared with the inoculated, nontreated control. Plants treated with copper octanoate, didecyl dimethyl ammonium chloride, and pydiflumetofen + difenoconazole had the lowest disease severity compared with other treatments. Treatments had no difference in plant height and width increment relative to inoculated, nontreated control plants (Table 3).

In trials 1 and 3, defoliation was not observed in any treated, inoculated, nontreated,

Table 3. Biological and chemical products' effects on plant growth of lilac (*Syringae reticulata*) grown in a shade house, McMinnville, TN, USA.

Treatments	Application date[i]	Height increase (cm)[ii]			Width increase (cm)[iii]		
		2021	2022	2023	2021	2022	2023
Bacillus amyloliquefaciens F727	1, 4, 6	28.40 ± 16.94 a[iv]	8.50 ± 5.63 a	10.40 ± 0.40 a	10.60 ± 5.80 ab	2.63 ± 0.38 a	12.00 ± 0.79 a
Copper octanoate	2, 4, 6	44.20 ± 18.42 a	8.75 ± 3.12 a	12.60 ± 1.12 a	3.50 ± 1.41 ab	7.00 ± 2.90 a	12.20 ± 0.37 a
Didecyl dimethyl ammonium chloride	4, 6	37.60 ± 18.11 a	9.75 ± 3.04 a	12.40 ± 0.93 a	9.40 ± 4.03 ab	2.75 ± 1.05 a	12.20 ± 0.46 a
Pydiflumetofen + difenoconazole	2, 4, 6	17.80 ± 11.19 a	5.25 ± 2.21 a	11.20 ± 0.73 a	1.80 ± 0.73 b	4.13 ± 1.78 a	12.30 ± 0.25 a
Thyme oil (5.56%)	1, 3, 4, 5, 6	26.40 ± 10.70 a	7.75 ± 3.47 a	11.40 ± 0.60 a	9.30 ± 2.86 ab	4.75 ± 1.59 a	12.10 ± 0.29 a
Thyme oil (15.5%)	1, 3, 5	28.20 ± 19.56 a	8.50 ± 2.50 a	11.60 ± 1.03 a	18.40 ± 7.42 a	5.25 ± 1.31 a	12.10 ± 0.29 a
Thyme oil (15.5%)	4, 6	54.40 ± 17.11 a	7.00 ± 3.85 a	12.00 ± 1.45 a	16.40 ± 8.74 ab	4.63 ± 0.55 a	12.10 ± 0.24 a
Inoculated, nontreated control		48.60 ± 15.16 a	8.50 ± 4.19 a	12.20 ± 1.36 a	17.30 ± 6.89 a	5.50 ± 1.67 a	12.60 ± 0.73 a
Noninoculated, nontreated control		33.00 ± 10.68 a	11.50 ± 3.10 a	11.80 ± 0.86 a	9.40 ± 4.77 ab	6.25 ± 1.80 a	12.10 ± 0.29 a
P value		0.7981	0.9814	0.8730	0.3458	0.5930	0.9960
F		0.57	0.29	0.46	1.17	0.82	0.15

[i] Application date: 1 = 3 d before *Pseudomonas syringae* pv. *syringae* inoculation, 2 = same day with *P. syringae* pv. *syringae* inoculation, 3 = 3 d after *P. syringae* pv. *syringae* inoculation, 4 = 7 d after *P. syringae* pv. *syringae* inoculation, 5 = 10 d after *P. syringae* pv. *syringae* inoculation, 6 = 14 d after *P. syringae* pv. *syringae* inoculation.

[ii] Height increase = final height – initial height.

[iii] Width increase= [(final widest width – initial widest width) + (final perpendicular width – initial perpendicular width)] ÷ 2.

[iv] Means followed by a different lowercase letter within a column are significantly different ($P \leq 0.05$). One-way analysis of variance was used to evaluate treatment effects on height increase, and width increase. Means were compared using Fisher's least significant difference test with an $\alpha = 0.05$.

or noninoculated, nontreated lilac plants. However, in trial 2, inoculated, nontreated control plants showed 3.8% defoliation. No phytotoxicity was observed on any of the treated or nontreated control plants.

Discussion

The *P. syringae* pathovar is stated to be one of the 10 most important phytobacteria, having an economic importance due to the wide host range and the great spread capacity on different hosts (Kennelly et al. 2007; Mansfield et al. 2012). Many methods have been reported to manage *P. syringae*, including cultural management such as proper pruning, using resistant plants, biological control, and chemical management based on copper compounds (Cordova et al. 2023). However, these methods have not always been successful. Cultural practices are related to environmental conditions such as temperature and humidity, which can influence the disease's development. In this study, three trials in shade house conditions were conducted to evaluate the efficacy of chemical and biochemical products on bacterial blight of lilac to develop recommendations for controlling *P. syringae* pv. *syringae*.

In the current study, all the biological and chemical products significantly reduced bacterial blight in lilac. The plants treated with copper octanoate, a chemical product used in this study, exhibited significantly lower severity of reduced bacterial blight and disease progress compared with the inoculated, nontreated control plants. This finding is notable despite the reported copper resistance of *P. syringae* pv. *syringae* in woody plants (Scheck and Pscheidt 1998) and did not control bacterial blight on lilac (Pscheidt and Bassinette 2013). Previously, consistent with our findings, the copper octanoate has been reported to be effective to reduce Xanthomonas blight on schefflera (*Schefflera arboricola*) caused by *Xanthomonas campestris* pv. *hederae* and bacterial leaf blight on cornelian cherry (*Cornus mas*) and chrysanthemum (*Chrysanthemum morifolium*) caused by *P. syringae* (Chase 1986; Mmbaga and Nnodu 2006). The bactericidal mechanism of copper-based compounds, Cu (II), induces oxidative stress when reduced to Cu (I). This stress can damage the bacterial cell membrane through lipid peroxidation, leading to membrane permeability and eventual cell death (Husseini and Akkopru 2020; Teitzel et al. 2006).

The chemical product didecyl dimethyl ammonium chloride was an effective treatment in reducing disease severity and AUDPC. This is comparable to other studies that demonstrate the ability of didecyl dimethyl ammonium chloride to suppress bacterial leaf spot on tomato and pepper caused by *Xanthomonas campestris* pv. *vesicatoria* and bacterial gall on loropetalum caused by *Pseudomonas amygdali* pv. *loropetalli* (Copes et al. 2019). Didecyl dimethyl ammonium chloride has a positive charge, which allows it to form an electrostatic bond with the negatively charged cell wall of microorganisms. This bond disrupts the cell wall permeability, leading to the disturbance of nutrient flow into the cell, waste discharge from the cell, and protein denaturation (Juergensen et al. 2000). Consequently, didecyl dimethyl ammonium chloride causes the leakage of intracellular molecules and the subsequent death of bacterial cells (Yoshimatsu and Hiyama 2007).

Pydiflumetofen + difenoconazole is known to quickly move from the leaf surface into the wax layer, becoming rain fast and creating a layer of protection within 24 h. It begins by slowly penetrating and spreading within the plant tissue, providing further disease control (Jennings et al. 2024). Pydiflumetofen + difenoconazole has been reported to be effective on controlling the disease severity of Pseudomonas leaf spot of magnolia (*Magnolia soulangeana*) and Xanthomonas leaf spot of geranium (*Pelargonium* 'Americana Bright Red') (Baysal-Gurel et al. 2020; Krasnow and Norman 2022). Similar efficacy was provided by pydiflumetofen + difenoconazole against *P. syringae* pv. *syringae* on lilac in the current study.

Thyme oil has been reported to have anti-biofilm and anti-phytotoxin effects on *P. syringae* strains (Oliva et al. 2015). However, thyme oil did not significantly affect *P. syringae* of kiwifruit (*Actinidia deliciosa*) (Monchiero et al. 2015). In the current study, lilac treated with thyme oil [5.56% and 15.5% (preventative and curative application)] had significantly less disease compared with the inoculated, nontreated control plants.

In the current study, *B. amyloliquefaciens* strain 727 was found to be effective in reducing disease severity and AUDPC compared with the inoculated, nontreated control. *Bacillus* spp. exhibit significant antimicrobial activity against different phytopathogens including *P. syringae* pv. *syringae* (Broniarek-Niemiec et al. 2023; Islam et al. 2020; Völksch and Weingart 1998). They use a mode of action that is associated to the production of lipopeptides that permeabilize the host membrane (Cawoy et al. 2011; Fira et al. 2018; Laverty et al. 2011) and are known to reduce pathogen development (Etesami et al. 2023). Moreover, applications of *B. amyloliquefaciens* induce host resistance that can suppress subsequent infection by pathogens (Ahmed et al. 2022; Chowdhury et al. 2013; Lahlali et al. 2022; Ongena et al. 2005; Preecha et al. 2010) as well as enhance plant growth (Gowtham et al. 2018; Kloepper et al. 2004; Pérez-García et al. 2011).

In conclusion, biological and chemical products in the current study have been shown to have an important role in reducing bacterial blight on lilac under shade house conditions. Among the trials, copper octanoate exhibited less effectiveness in reducing disease severity compared with other treatments in trial 1. It is important to note that temperature can significantly influence the efficacy of copper treatments. Higher temperatures can lead to rapid inactivation of copper (Sharan et al. 2010), and trial 1 recorded the highest temperatures among all trials. This may explain the lower efficacy of copper octanoate in reducing disease severity during this trial. Furthermore, the current study could provide alternative treatments to copper applications, in the case of the copper resistance isolate of *P. syringae* pv. *syringae*. Treatment with thyme oil (15.5%, preventative application) was efficient in all trials; application of didecyl dimethyl ammonium chloride and pydiflumetofen + difenoconazole in trial 1 and trial 3 and *B. amyloliquefaciens* strain 727 in trial 1 have also proved to be a useful tool to control bacterial blight. Therefore, this study can be used to consider new treatments and formulate management plans for the control of *P. syringae* pv. *syringae* on lilac.

References Cited

Ahmed W, Zhou G, Yang J, Munir S, Ahmed A, Liu Q, Zhao Z, Ji G. 2022. *Bacillus amyloliquefaciens* WS-10 as a potential plant growth-promoter and biocontrol agent for bacterial wilt disease of flue-cured tobacco. Egypt J Biol Pest Control. 32(1):25. https://doi.org/10.1186/s41938-022-00527-5.

Aiello D, Ferrante P, Vitale A, Polizzi G, Scortichini M, Cirvilleri G. 2015. Characterization of *Pseudomonas syringae* pv. *syringae* isolated from mango in Sicily and occurrence of copper-resistant strains. J Plant Pathol. 97:273–282.

Balestra GM, Heydari A, Ceccarelli D, Ovidi E, Quattrucci A. 2009. Antibacterial effect of *Allium sativum* and *Ficus carica* extracts on tomato bacterial pathogens. Crop Prot. 28(10): 807–811. https://doi.org/10.1016/j.cropro.2009.06.004.

Baysal-Gurel F, Oksel C, Simmons T, Jennings C. 2020. Evaluation of bactericides for control of Pseudomonas leaf spot on Magnolia. Plant Disease Management Report. 15:OT016.

Broniarek-Niemiec A, Børve J, Puławska J. 2023. Control of bacterial canker in stone fruit trees by chemical and biological products. Agronomy. 13(4):1166. https://doi.org/10.3390/agronomy13041166.

Cawoy H, Bettiol W, Fickers P, Ongena M. 2011. Bacillus-based biological control of plant diseases, p 273–302. In: Stoytcheva M (ed). Pesticides in the modern world: Pesticides use and management. IntechOpen, London, UK.

Cazorla FM, Arrebola E, Sesma A, Pérez-Garcia A, Codina JC, Murillo J, de Vicente A. 2002. Copper resistance in *Pseudomonas syringae* strains isolated from mango is mainly encoded by plasmids. Phytopathology. 92(8):909–916. https://doi.org/10.1094/PHYTO.2002.92.8.909.

Chase AR. 1986. Effect of experimental bactericides on three bacterial disease of foliage plants. J Environ Hortic. 4(2):37–41. https://doi.org/10.24266/0738-2898-4.2.37.

Chowdhury SP, Dietel K, Randle M, Schmid M, Junge H, Borris R, Hartmann A, Grosch R. 2013. Effects of *Bacillus amyloliquefaciens* FZB42 on lettuce growth and health under pathogen pressure and its impact on the rhizosphere bacterial community. PLoS One. 8(7):e68818. https://doi.org/10.1371/journal.pone.0068818.

Copes WE, Mavrodi OV, Mavrodi DV. 2019. Control of *Pseudomonas amygdali* pv. *loropetalli* on metal, wood, and *Loropetalum chinense* stem surface. Plant Health Prog. 20(4):270–277. https://doi.org/10.1094/PHP-09-19-0068-RS.

Cordova P, Rivera-González JP, Rojas-Martínez V, Fiore N, Bastías R, Zamorano A, Vera F, Barrueto J, Díaz B, Ilabaca-Díaz C, Bertaccini A, Higuera G. 2023. Phytopathogenic *Pseudomonas syringae* as a threat to agriculture: Perspectives of

a promising biological control using bacteriophages and microorganisms. Horticulturae. 9(6):712. https://doi.org/10.3390/horticulturae9060712.
Crosse JE. 1956. Bacterial canker of stone fruits: II. Leaf scar infection of cherry. J Hort Sci. 31(3):212–224. https://doi.org/10.1080/00221589.1956.11513871.
Doolotkeldieva T, Bobusheva S. 2020. Characterization of *Pseudomonas syringae* pv. *syringae* from diseased stone fruits in Kyrgyzstan and testing of biological agents against pathogen. Int J Phytopathol. 9(2):71–91. https://doi.org/10.33687/phytopath.009.02.3270.
Etesami H, Jeong BR, Glick BR. 2023. Biocontrol of plant diseases by *Bacillus* spp. Physiol Mol Plant Pathol. 126:102048. https://doi.org/10.1016/j.pmpp.2023.102048.
Fira DJ, Dimkić I, Beric T, Lozo J, Stanković S. 2018. Biological control of plant pathogens by *Bacillus* species. J Biotechnol. 285:44–55. https://doi.org/10.1016/j.jbiotec.2018.07.044.
Gasecka M, Krzymińska-Bródka A, Magdziak Z, Czuchaj P, Bykowska J. 2023. Phenolic compounds and organic acid composition of *Syringae vulgaris* L. flowers and infusions. Molecules. 28(13):5159. https://doi.org/10.3390/molecules28135159.
Gilardi G, Gullino ML, Garibaldi A. 2010. Evaluation of spray programs for the management of leaf spot incited by *Pseudomonas syringae* pv. *syringae* on tomato cv. Cuore di bue. Crop Prot. 29(4):330–335. https://doi.org/10.1016/j.cropro.2009.11.010.
Gowtham HG, Murali M, Brijesh-Singh S, Lakshmeesha TR, Narasimha-Murthy K, Amruthesh KN, Niranjana SR. 2018. Plant growth promoting rhizobacteria-*Bacillus amyloliquefaciens* improves plant growth and induces resistance in chili against anthracnose disease. Biol Control. 126:209–217. https://doi.org/10.1016/j.biocontrol.2018.05.022.
Guan J, Nutter FW. 2002. Relationships between percentage defoliation, dry weight, percentage reflectance, leaf-to-stem ration, and green leaf area index in the alfalfa leaf spot pathosystem. Crop Sci. 42(4):1264–1273. https://doi.org/10.2135/cropsci2002.1264.
Hirano SS, Clayton MK, Upper CD. 1994. Estimation of and temporal changes in means and variances of populations of *Pseudomonas syringae* on snap bean leaflets. Phytopathology. 84(9):934–940. https://doi.org/10.1094/Phyto-84-934.
Hirano SS, Upper CD. 2000. Bacteria in the leaf ecosystem with emphasis on *Pseudomonas syringae*—A pathogen, ice nucleus, and epiphyte. Microbiol Mol Biol Rev. 64(3):624–653. https://doi.org/10.1128/MMBR.64.3.624-653.2000.
Hongxia C, Gaoming J, Shuying Z. 2004. The distribution, origin and evolution of Syringa. Zhiwu Yanjiu. 24:141–145.
Husseini A, Akkopru A. 2020. The possible mechanisms of copper resistance in the pathogen *Pseudomonas syringae* pathovars in stone fruit trees. Phytoparasitica. 48(5):705–718. https://doi.org/10.1007/s12600-020-00828-1.
Islam M, Sultana R, Hasan M, Alam S, Sikdar B, Kamaruzzaman M, Islam A. 2020. Characterization and biocontrol measures of *Pseudomonas syringae* pv. *syringae* associated with citrus blast disease. Vegetos. 33(3):555–569. https://doi.org/10.1007/s42535-020-00138-1.
Jennings C, Simmons T, Parajuli M, Oksel C, Liyanapathiranage P, Hikkaduwa Epa Liyanage K, Baysal-Gurel F. 2024. Chemical control of powdery mildew of bigleaf hydrangea. HortScience. 59(2):173–178. https://doi.org/10.21273/HORTSCI17500-23.
Juergensen L, Busnarda J, Caux PY, Kent RA. 2000. Fate, behavior, and aquatic toxicity of the fungicide DDAC in the Canadian environment. Environ Toxicol. 15(3):174–200. https://doi.org/10.1002/1522-7278(2000)15:3<174::AID-TOX4>3.0.CO;2-P.
Kennelly MM, Cazorla FM, de Vicente A, Ramos C, Sundin GW. 2007. *Pseudomonas syringae* diseases of fruit trees: Progress toward understanding and control. Plant Dis. 91(1):4–17. https://doi.org/10.1094/PD-91-0004.
Keshtkar AR, Khodakaramian G, Rouhrazi K. 2016. Isolation and characterization of *Pseudomonas syringae* pv. *syringae* which induce leaf spot on walnut. Eur J Plant Pathol. 146(4):837–846. https://doi.org/10.1007/s10658-016-0962-2.
King ED, Ward MK, Raney DE. 1954. Two simple media for the demonstration of pyocyanin and fluorescin. J Lab Clin Med. 44(2):301–307.
Kloepper JW, Ryu CM, Zhang S. 2004. Induced systemic resistance and promotion of plant growth by *Bacillus* spp. Phytopathology. 94(11):1259–1266. https://doi.org/10.1094/PHYTO.2004.94.11.1259.
Kosiada T. 2016. Ascochyta blight (*Ascochyta syringae*) of lilac (*Syringae vulgaris* L.). Acta Sci Pol Hortorum Cultus. 15(4):27–34.
Krasnow C, Norman D. 2022. Efficacy of Postiva™ for management of bacterial diseases of ornamental crops. Appl Microbiol. 2(2):302–308. https://doi.org/10.3390/applmicrobiol2020022.
Lahlali R, Ezrari S, Radouane N, Kenfaoui J, Esmaeel Q, Hamss HE, Belabess Z, Barka EA. 2022. Biological control of plant pathogens: A global perspective. Microorganisms. 10(3):596. https://doi.org/10.3390/microorganisms10030596.
Lamichhane JR, Varvaro L, Parisi L, Audergon JM, Morris CE. 2014. Disease and frost damage of woody plants caused by *Pseudomonas syringae*: Seeing the forest for the trees, p 235–295. In: Spark DL (ed). Advances in agronomy. Academic Press, Cambridge. https://doi.org/10.1016/B978-0-12-800132-5.00004-3.
Lamichhane JR, Messéan A, Morris CE. 2015. Insights into epidemiology and control of diseases of annual plants caused by the *Pseudomonas syringae* species complex. J Gen Plant Pathol. 81(5):331–350. https://doi.org/10.1007/s10327-015-0605-z.
Latorre BA, Lillo C, Rioja ME. 2002. Effects of temperature, free moisture duration and inoculum concentration on infection of sweet cherry by *Pseudomonas syringae* pv. *syringae*. Phytoparasitica. 30(4):410–419. https://doi.org/10.1007/BF02979689.
Laverty G, Gorman SP, Gilmore BF. 2011. The potential of antimicrobial peptides as biocides. Int J Mol Sci. 12(10):6566–6596. https://doi.org/10.3390/ijms12106566.
Little EL, Bostock RM, Kirkpatrick BC. 1998. Genetic characterization of *Pseudomonas syringae* pv. *syringae* strains from stone fruits in California. Appl Environ Microbiol. 64(10):3818–3823. https://doi.org/10.1128/AEM.64.10.3818-3823.1998.
Mansfield J, Genin S, Magori S, Citovsky V, Sriariyanum M, Ronald P, Dow M, Verdier V, Beer SV, Machado MA, Toth I, Salmond G, Foster GD. 2012. Top 10 plant pathogenic bacteria in molecular plant pathology. Mol Plant Pathol. 13(6):614–629. https://doi.org/10.1111/j.1364-3703.2012.00804.x.
Monchiero M, Gullino ML, Pugliese M, Spadaro D, Garibaldi A. 2015. Efficacy of different chemical and biological products in the control of *Pseudomonas syringae* pv. *actinidiae* on kiwifruit. Australasian Plant Pathol. 44(1):13–23. https://doi.org/10.1007/s13313-014-0328-1.
Morris CE, Monier JM. 2003. The ecological significance of biofilm formation by plant-associated bacteria. Annu Rev Phytopathol. 41:429–453. https://doi.org/10.1146/annurev.phyto.41.022103.134521.
Mmbaga MT, Nnodu EC. 2006. Biology and control of bacterial leaf blight of *Cornus mas*. HortScience. 41(3):721–724. https://doi.org/10.21273/HORTSCI.41.3.721.
National Agriculture Statistics Service. 2020. 2019 census of horticultural specialties. 2017 Census of Agriculture. https://www.nass.usda.gov/Publications/AgCensus/2017/Online_Resources/Census_of_Horticulture_Specialties/HORTIC.pdf. [accessed 15 Mar 2024].
Nejad P, Ramstedt M, Granhall U. 2004. Pathogenic ice-nucleation active bacteria in willows short rotation forestry. For Pathol. 34(6):369–381. https://doi.org/10.1111/j.1439-0329.2004.00378.x.
Oliva MM, Carezzano ME, Giuliano M, Daghero J, Zygadlo J, Bogino P, Giordano W, Demo M. 2015. Antimicrobial activity of essential oils of *Thymus vulgaris* and *Origanum vulgare* on phytopathogenic strains isolated from soybean. Plant Biol (Stuttg). 17(3):758–765. https://doi.org/10.1111/plb.12282.
Ongena M, Duby F, Jourdan E, Beaudry T, Jadin V, Dommes J, Thonart P. 2005. *Bacillus subtilis* M4 decreases plant susceptibility towards fungal pathogens by increasing host resistance associated with differential gene expression. Appl Microbiol Biotechnol. 67(5):692–698. https://doi.org/10.1007/s00253-004-1741-0.
Pérez-García A, Romero D, de Vicente A. 2011. Plant protection and growth stimulation by microorganisms: Biotechnological applications of bacilli in agriculture. Curr Opin Biotechnol. 22(2):187–193. https://doi.org/10.1016/j.copbio.2010.12.003.
Preecha C, Sadowsky MJ, Prathuangwong S. 2010. Lipopeptide surfactin produced by *Bacillus amyloliquefaciens* KPS46 is required for biocontrol efficacy against *Xanthomonas axonopodis* pv. *glycines*. Kasetsart J Nat Sci. 44:84–99.
Pscheidt JW, Bassinette JP. 2013. Bactericides for management of bacterial blight of lilac. Plant Dis Manag Rep. 7:OT011.
Renick LJ, Cogal AG, Sundin GW. 2008. Phenotypic and genetic analysis of epiphytic *Pseudomonas syringae* populations from sweet cherry in Michigan. Plant Dis. 92(3):372–378. https://doi.org/10.1094/PDIS-92-3-0372.
Rouse DI, Nordheim EV, Hirano SS, Upper CD. 1985. A model relating the probability of foliar disease incidence to the population frequencies of bacterial plant pathogens. Phytopathology. 75(5):505–509. https://doi.org/10.1094/Phyto-75-505.
Scheck HJ, Pscheidt JW, Moore LW. 1996. Copper and streptomycin resistance in strains of *Pseudomonas syringae* from Pacific Northwest nurseries. Plant Dis. 80(9):1034–1039. https://doi.org/10.1094/PD-80-1034.
Scheck HJ, Pscheidt JW. 1998. Effects of copper bactericides on copper-resistant and -sensitive strains of *Pseudomonas syringae* pv. *syringae*. Plant Dis. 82(4):397–406. https://doi.org/10.1094/PDIS.1998.82.4.397.
Sharan R, Chhibber S, Attri S, Reed RH. 2010. Inactivation and injury of *Escherichia coli* in a copper water storage vessel: Effects of temperature and pH. Antonie Van Leeuwenhoek. 97(1):91–97. https://doi.org/10.1007/s10482-009-9395-7.

Simonetti G, Pucci N, Brasili E, Valletta A, Sammarco I, Carnevale E, Pasqua G, Loreti S. 2019. In vitro antimicrobial activity of plant extracts against *Pseudomonas syringae* pv. *actinidiae* causal agent of bacterial canker in kiwifruit. Plant Biosystems - An International Journal Dealing with All Aspects of Plant Biology. 154(1):100–106. https://doi.org/10.1080/11263504.2019.1699194.

Spotts RA, Cervantes LA. 1994. *Pseudomonas* canker of pear trees in Oregon, cultivar resistance, and effect of trunk guards on canker incidence and bacterial survival on bark. Plant Dis. 78(9):907–910. https://doi.org/10.1094/PD-78-0907.

Sundin GW, Olson BD, Jones AL. 1988. Overwintering and population dynamics of *Pseudomonas syringae* pv. *syringae* and *Pseudomonas syringae* pv. *morsprunorum* on sweet and sour cherry trees. Can J Plant Pathol. 10(4): 281–288. https://doi.org/10.1080/07060668809501701.

Tattar TA. 1989. Infectious diseases, p 18–32. In: Tattar TA (ed). Diseases of shade trees. Academic Press, San Diego, CA, USA.

Teitzel GM, Geddie A, de Long SK, Kirisits MJ, Whiteley M, Parsek MR. 2006. Survival and growth in the presence of elevated copper: Transcriptional profiling of copper-stressed *Pseudomonas aeruginosa*. J Bacteriol. 188(20): 7242–7256. https://doi.org/10.1128/JB.00837-06.

Toth G, Barabas C, Toth A, Kery A, Beni S, Boldizsar I, Varga E, Noszal B. 2015. Characterization of antioxidant phenolics in *Syringae vulgaris* L. flowers and fruits by HPLC-DAD-ED-MS. Biomed Chromatogr. 30(6):923–932. https://doi.org/10.1002/bmc.3630.

Varga E, Barabas C, Toth A, Boldizsar I, Noszal B, Toth G. 2019. Phenolic composition, antioxidant and antinociceptive activities of *Syringae vulgaris* L. bark and leaf extracts. Nat Prod Res. 33(11):1664–1669. https://doi.org/10.1080/14786419.2018.1425855.

Völksch B, Weingart H. 1998. Toxin production by pathovars of *Pseudomonas syringae* and their antagonistic activities against epiphytic microorganisms. J Basic Microbiol. 38(2): 135–145. https://doi.org/10.1002/(SICI)1521-4028(199805)38:2<135::AID-JOBM135>3.0.CO;2-Y.

Wimalajeewa DSL, Cahill R, Hepworth G, Schneider HG, Washbourne JW. 1991. Chemical control of bacterial canker (*Pseudomonas syringae* pv. *syringae*) of apricot and cherry in Victoria. Aust J Exp Agric. 31(5):705–708. https://doi.org/10.1071/EA9910705.

Xin XF, Kvitko B, He SY. 2018. *Pseudomonas syringae*: What it takes to be a pathogen. Nat Rev Microbiol. 16(5):316–328. https://doi.org/10.1038/nrmicro.2018.17.

Yiğit R, Çoklar H, Akbulut M. 2022. Some physicochemical and phytochemical properties of *Syringa vulgaris* L. flower tea: Influence of flower drying technique, brewing method and brewing time. Food Measure. 16(5):4185–4197. https://doi.org/10.1007/s11694-022-01511-1.

Yoshimatsu T, Hiyama K-I. 2007. Mechanism of the action of didecyl dimethyl ammonium chloride (DDAC) against *Escherichia coli* and morphological changes of the cells. Biocontrol Sci. 12(3):93–99. https://doi.org/10.4265/bio.12.93.