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Molecular characterization of *Gilbertella persicaria*, the causal agent of postharvest soft rot in Barbados cherry (*Malpighia glabra* L.), and evaluation of chitosan and selected plant extracts under *in vitro* and *in vivo* conditions

Nguyen Quoc Thai[1*], Phan Thi Minh Thu[1], Nguyen Thi Truc Mai[1], Le Phuoc Thanh[2], and Le Thanh Toan[2]

[1]*Faculty of Agriculture-Fisheries, Mekong University, Viet Nam*

[2]*College of Agriculture, Can Tho University, Viet Nam*

**Corresponding author (nguyenquocthai@mku.edu.vn)*

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ABSTRACT

Postharvest soft rot is one of the most destructive diseases affecting Barbados cherry (Malpighia glabra L.), causing substantial losses in fruit quality and marketability in Dong Thap Province, Viet Nam. This study identified Gilbertella persicaria as the causal agent of postharvest soft rot through morphological characterization and ITS rDNA sequence analysis, and evaluated the inhibitory efficacy of chitosan and selected plant extracts under both in vitro and in vivo conditions. In vitro assays demonstrated that chitosan at concentrations of 0.6–1.0% completely inhibited mycelial growth and markedly suppressed spore production, whereas garlic extract (6–8%) exhibited strong but short-lived antifungal activity. Under in vivo conditions, chitosan at 0.8–1.0% significantly reduced disease incidence (32.5–37.5%) and lesion area (8.9–16.6%) compared with untreated controls. These findings demonstrate the potential of chitosan as an eco-friendly and effective alternative for managing postharvest soft rot in Barbados cherry.

1. INTRODUCTION

Barbados cherry (*Malpighia glabra* L.) is a highly nutritious tropical fruit, particularly rich in vitamin C and antioxidants. Owing to its high economic value and increasing market demand, Barbados cherry has become an important crop in many tropical and subtropical regions. In Viet Nam, Barbados cherry cultivation is concentrated in several provinces in the Mekong Delta, with Dong Thap being one of the significant producing areas, providing an important source of income for local growers. Barbados cherry fruits are highly prone to postharvest spoilage because of their thin epidermis, delicate flesh, and elevated respiration rate, which collectively render them vulnerable to fungal infection.

Among postharvest diseases affecting Barbados cherry, soft rot caused by *Gilbertella persicaria* (Mucorales order) is considered one of the most serious. The pathogen is characterized by rapid mycelial growth, cotton-like colonies, non-septate hyphae, and spherical sporangia and characteristic sporangiospores, often causing complete rot of the fruit within just a few days of storage (Zhang et al., 2020; Souza et al., 2023).

The management of postharvest fungal diseases still depends largely on synthetic fungicides. However, growing concerns about chemical residues, pathogen resistance, and food safety have spurred the search for environmentally friendly alternatives (Bautista-Baños et al., 2006; Romanazzi et al., 2018). In recent years, natural compounds such as

plant extracts and biopolymers have received considerable attention for postharvest disease management. Garlic extracts and essential oils derived from lemongrass and *Ageratum conyzoides* have demonstrated antifungal activity against several postharvest pathogens, mainly due to their bioactive compounds (Ankri & Mirelman, 1999; Burt, 2004; Tzortzakis & Economakis, 2007; Chahal et al., 2021; Cuong et al., 2021). In addition, chitosan has been widely reported as an effective natural antifungal agent because of its ability to inhibit fungal growth, form a protective coating, and induce host defense responses (Bautista-Baños et al., 2006; Romanazzi et al., 2018).

Despite growing interest in natural antifungal compounds, research on managing soft rot caused by *Gilbertella persicaria* in Barbados cherry remains scarce. Therefore, the objectives of this study were to determine the occurrence of postharvest soft rot in Barbados cherry, identify its causal pathogen using morphological and molecular approaches, and evaluate the inhibitory efficacy of selected plant extracts and chitosan under both *in vitro* and *in vivo* conditions.

2. MATERIALS AND METHODS

2.1. Survey of the current status of soft rot disease and isolation of the causative agent

A survey of the current status of postharvest soft rot disease in Barbados cherry was conducted in two representative Barbados cherry-growing areas in Dong Thap Province, namely Binh Nghi and Binh An communes. Fruit samples were collected at harvest time and stored in the laboratory. Direct observation and monitoring were conducted to record postharvest disease symptoms and classify the main types of damage based on the fruits' external appearance.

Barbados cherry fruits exhibiting typical soft rot symptoms were selected for pathogen isolation. Samples were cut from the boundary of diseased tissues, surface sterilized with 1% NaOCl solution for 1 minute, rinsed 2-3 times with sterile distilled water, and dried on sterile filter paper. Tissue samples were aseptically transferred onto potato dextrose agar (PDA) Petri dishes and incubated at 28 ± 2°C. Emerging colonies were repeatedly subcultured to obtain pure isolates. Pathogenicity of the purified isolates was confirmed by inoculating healthy Barbados cherry fruits and re-isolating the pathogen in accordance with Koch's postulates.

Morphological characteristics of the fungal colonies, including colony color, growth pattern, hyphal structure, and spore morphology, were examined under a light microscope. Molecular identification was performed through amplification and sequencing of the rDNA region using the ITS1 (5'-TCCGTAGGTGAACCTGCGG-3') and ITS4 (5'-TCCTCCGCTTATTGATATGC-3') primer pair. The obtained sequences were compared with the GenBank database using BLAST to identify pathogenic fungal species. Sequence alignment was performed using ClustalW, and phylogenetic relationships were inferred using the Neighbor-Joining method implemented in MEGA 12 (White et al., 1990; Zhang et al., 2020).

2.2. Evaluating the efficacy of chitosan and plant extracts against *G. persicaria* under *in vitro* conditions

The experiment was designed as a completely randomized design with 21 treatments, including plant extracts at various concentrations, chitosan, and a control treatment of distilled water. Specifically, the treatments were established as follows: extracts of Billy goat weed at concentrations of 2, 4, 6, 8, and 10%; lemongrass at concentrations of 1, 2, 4, 6, and 8%; garlic at concentrations of 1, 2, 4, 6, and 8%; chitosan at concentrations of 0.2, 0.4, 0.6, 0.8, and 1.0%; and a control (sterilized distilled water). Each treatment was replicated 5 times.

The fresh plant materials, including garlic (*Allium sativum)*, lemongrass (*Cymbopogon citratus*), and billy goat weed (*Ageratum conyzoides*), were harvested, washed with distilled water, and drained. The materials were dried at room temperature, ground into a powder, and then extracted with distilled water in a water bath at 60–62°C for 15 minutes. The extract was filtered through sterile filter paper and diluted to achieve the experimental concentrations (Toan & Nam, 2021). Chitosan was dissolved in a 1% acetic acid solution, stirred until completely dissolved, and the pH was adjusted to approximately 5.0 before use (Duy et al., 2014).

Plant extracts or chitosan solutions were added directly to PDA medium that had been melted and cooled to approximately 55-60°C, then poured into Petri dishes (10 mL/dish). After the medium solidified, a mycelial plug of *G. persicaria* (5 mm-diameter) was placed in the center of the Petri dish. The Petri dishes were incubated at room temperature (Elamawi & El-Shafey, 2013).

Colony diameters were measured along two perpendicular axes at 1, 2, and 3 days after inoculation (DAI). Inhibitory efficacy (IE, %) was calculated by using the formula as (1).

$$IE\ (\%) = [(CD_{control} - CD_{treatment}) / CD_{control}] \times 100\% \quad (1)$$

In this formula, $CD_{control}$ is the colony diameter of the control and $CD_{treatment}$ is the colony diameter of the treated treatment (Atlas, 2010).

At 3 DAI, three mycelial plugs (5 mm in diameter) were excised from the colony margin, added to 10 mL of sterile distilled water, and vortexed to create a spore suspension. The suspension was filtered through cheesecloth, and the spore density was counted with a Neubauer counting chamber (Toan & Nam, 2021).

The highly effective treatments identified in the *in vitro* assay were selected for subsequent *in vivo* evaluation.

2.3. Evaluating the efficacy of controlling Barbados cherry fruit rot caused by *G. persicaria* under *in vivo* conditions

The experiment was conducted under controlled laboratory conditions using a completely randomized design with six treatments: garlic extract (6% and 8%), chitosan (0.6%, 0.8%, and 1.0%), and a sterile distilled water control. Each treatment consisted of four replicates, with ten fruits per replicate.

Barbados cherry fruits of physiological ripeness, uniform in size, and free from disease symptoms were directly purchased from the orchard and brought to the laboratory within 24 hours of harvesting. The fruits were washed with water to remove dirt, air-dried, then surface-sterilized with a 1% NaOCl solution for 1 minute, rinsed with sterile distilled water, and air-dried.

The *G. persicaria* isolate was cultured on PDA medium. Spore suspensions were prepared at a density of 10^6 spores/mL. Barbados cherry fruits were artificially inoculated by dipping the entire fruits into the spore suspension for 1 minute, then drying under sterile conditions for 4 hours to allow the spores to attach and begin their germination. After the infection period, the fruits were treated by dipping them into the solution of each treatment (garlic or chitosan) for 2 minutes. For the control treatment, the fruits were dipped in sterile distilled water. After treatment, the fruits were allowed to dry naturally, then 10 fruits from each replicate were placed in a plastic box lined with absorbent paper moistened with sterile distilled water. The percentage of fruit rot (%) and lesion area ratio/fruit (%) were recorded at 1, 3, 5, and 7 days after treatment (DAT). The control efficacy against fruit rot incidence and lesion area ratio per fruit at 5 and 7 DAT was calculated using Abbott's formula (Abbott, 1925) as (2).

$$E = (C-T) \times 100/C \quad (2)$$

where C: percentage of fruit rot (%) or lesion area ratio/fruit (%) in the control; T: percentage of fruit rot (%) or lesion area ratio/fruit (%) in the treated treatment.

2.4. Data analysis

Data were analyzed using SPSS version 20.0. Analysis of variance (ANOVA) was performed, and treatment means were separated using Duncan's multiple range test at the 5% significance level. The sequencing results of two isolates, F16 and F19, were analyzed using FinchTV software (version 1.4.0) to examine distinct nucleotide peaks and background noise. Subsequently, the obtained PCR product sequences were compared against the NCBI database (https://blast.ncbi.nlm.nih.gov) using the BLAST tool to assess species-level identity and query coverage. Representative sequences, together with reference sequences retrieved from GenBank, were used to construct a phylogenetic tree (Kumar et al., 2024).

3. RESULTS AND DISCUSSION

3.1. Occurrence of postharvest soft rot disease on Barbados cherry in Dong Thap Province and identification of the causal pathogen

Survey results from two concentrated Barbados cherry-growing areas in Dong Thap Province showed that soft rot disease incidence was very high (95–100%), with soft rot as the dominant disease at an average rate of 81.5%, much higher than fruit rot (16.0%) (Table 1). These findings indicate that soft rot is the predominant postharvest disease affecting Barbados cherry in the surveyed areas. Soft rot disease on Barbados cherry fruit initially appeared as slightly sunken, light-brown lesions on the fruit surface. The lesions rapidly expanded and coalesced into large necrotic areas, resulting in softening and maceration of the fruit tissues. Abundant cottony, hyaline fungal mycelia developed on the fruit surface, accompanied by black sporangia formed externally on the mycelial mass. Ultimately, the entire fruit became soft and water-soaked, exuded

fluids, and underwent complete decay. In contrast, the initial symptoms of fruit rot disease also appeared as slightly sunken, light-brown lesions on the fruit surface, but with a dark center in the middle of the lesion. The disease progressed rapidly, and scattered patches of white mycelia gradually developed on the lesion surface. Infected fruits gradually lost moisture, while the fruit peel became wrinkled and changed color from yellow to dark brown or black. As the disease progressed, diseased fruits were covered with dense white fungal mycelia, and the lesions penetrated deeply into the fruit flesh tissues (Figure 1). The fungal isolate obtained from soft rot lesions was inoculated onto healthy Barbados cherry fruits to verify Koch's postulates. The inoculated fruits developed symptoms similar to those observed on naturally infected fruits collected from the orchards, including the formation of characteristic sporangiospores (Figure 2).

Table 1. Postharvest disease composition on Barbados cherries in two communes of Dong Thap Province, 2025

Survey area	Disease incidence (%)	Disease percentage of each disease (%)	
		Soft rot	Fruit rot
Binh Nghi	95.0	85.0	10.0
Binh An	100.0	78.0	22.0
Average (%)	97.5	81.5	16.0

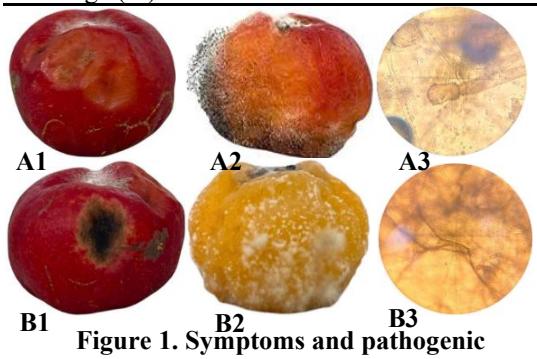

Figure 1. Symptoms and pathogenic characteristics of soft rot and fruit rot diseases on Barbados cherry fruits

Soft rot symptoms on fruits at 3 days (A1) and 5 days (A2) after harvest (DAH), and characteristic sporangiophore (A3); fruit rot symptoms on fruits at 3 (B1) and 5 (B2) DAH, and fungal mycelia without conidia (B3).

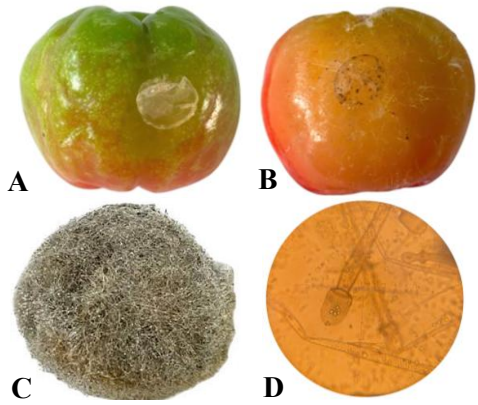

Figure 2. Symptoms and fungal characteristics of soft rot disease on Barbados cherry fruits

Symptoms at 1 DAI (A), 2 DAI (B), and 4 DAI (C), with characteristic sporangiophore formation on infected fruits (D).

Among the ten fungal isolates obtained, isolates F16 and F19 exhibited pathogenicity on Barbados cherry fruits and were selected for morphological observation. The colonies of both isolates grew rapidly (64.5±2 mm in diameter after 24 h at 28.3°C) on PDA, producing cottony white colonies that gradually became grayish with age. The fungus produced coenocytic (aseptate) hyphae and erect sporangiophores that were mostly unbranched, hyaline to light brown, and occasionally curved near the apex. Sporangia, 52.3–110.0 (84.4) x 55.7–121 (85.7) µm (n = 30), were initially white to yellowish-brown, becoming dark brown to black at maturity, with persistent walls and a distinct longitudinal suture. Columellae were hyaline and variable in shape, including globose, ellipsoid, cylindrical, obovoid, or pyriform forms. Sporangiospores, 6.9–8.3 (7.3) x 7.5–11.5 (9.6) µm (n = 30) were unicellular, hyaline, globose to ellipsoid or irregular in shape, and commonly possessed characteristic filiform appendages at both ends (Figure 3). Based on these morphological features and comparisons with previous descriptions reported by Zhang et al. (2020) and Souza et al. (2023), the pathogen causing soft rot was identified as belonging to the genus *Gilbertella*.

BLAST analysis of the ITS rDNA sequences (amplified using ITS1/ITS4 primers) revealed that both the F16 isolate (from fruits harvested in Binh Nghi, Dong Thap) and the F19 isolate (from fruit

samples in Binh An, Dong Thap) showed 100% sequence identity and 100% query coverage with reference sequences of *Gilbertella persicaria* available in GenBank. Furthermore, the phylogenetic analysis showed that the F16 and F19 isolates clustered closely with NCBI reference isolates of *Gilbertella persicaria*, remaining distinct from those of *G. hainanensis* (Figure 4).

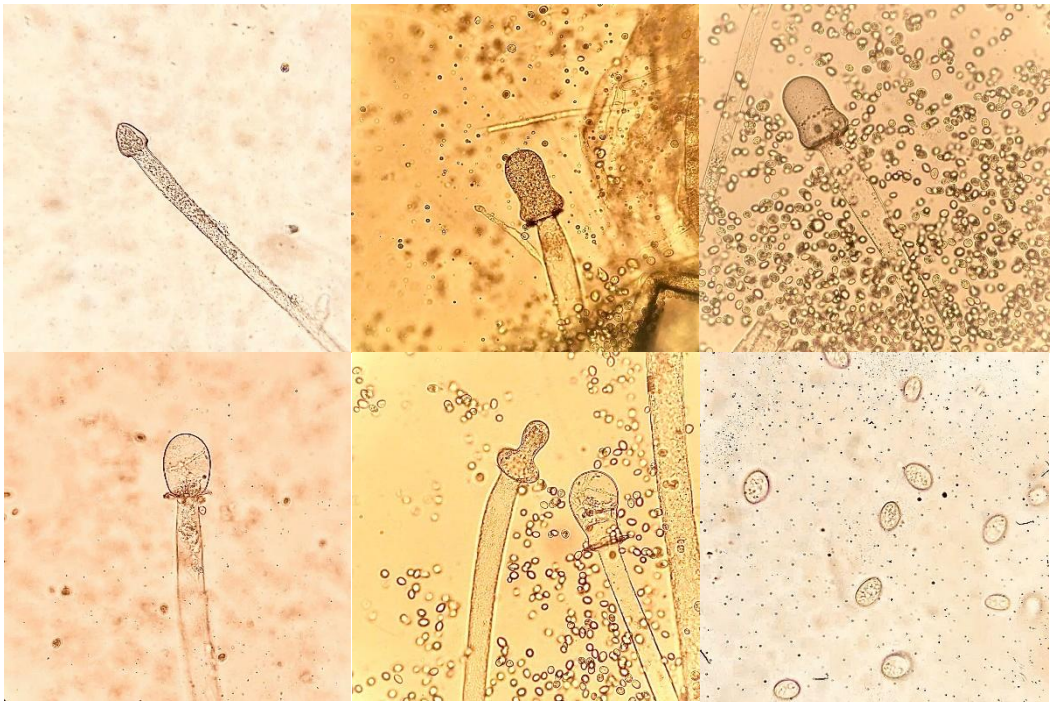

Figure 3. Morphological characteristics of *Gilbertella persicaria* isolated from diseased Barbados cherry fruits and cultured on potato dextrose agar (PDA) for 2 days

Colony morphology on PDA (A). Sporangia (B-C). Sporangiophores with columellae (D-H). Sporangiospores (I). Scale bars: B-H = 50 µm; I = 10 µm.

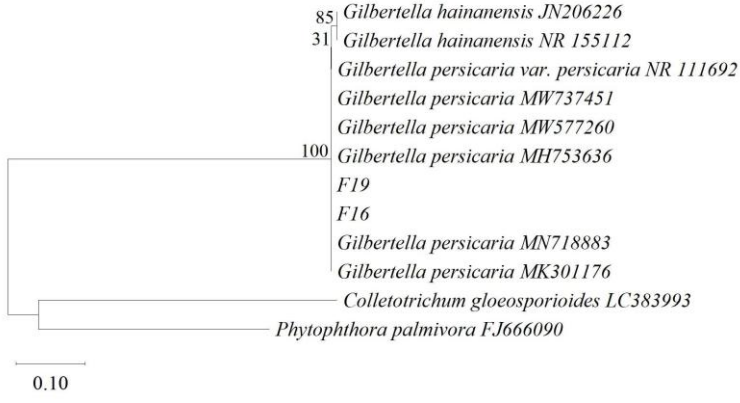

Figure 4. Phylogenetic tree of fungal isolates F16 (Binh Nghi commune, Dong Thap Province) and F19 (Binh An commune, Dong Thap Province) causing soft rot disease in Barbados cherries.

The combined evidence from pathogenicity tests, morphological observations, and ITS rDNA sequence analysis conclusively identified *Gilbertella persicaria* as the causal agent of postharvest soft rot in Barbados cherry. The confirmed isolates were subsequently used to evaluate the inhibitory efficacy of chitosan and selected plant extracts under laboratory and fruit storage conditions.

3.2. *In vitro* inhibitory effects of chitosan and plant extracts on mycelial growth and sporulation of *G. persicaria*

Experimental results showed that the inhibitory effect against *G. persicaria* depended significantly on the type of treatment agent and the concentration used (Table 2). Among all tested treatments, chitosan exhibited the strongest and most consistent inhibitory activity against *G. persicaria* (Figure 5). Complete inhibition of mycelial growth was observed at concentrations ≥ 0.6%, accompanied by a substantial reduction in spore production. The superior efficacy of chitosan may be attributed to its ability to disrupt fungal cell membrane integrity, alter cell permeability, and interfere with essential physiological processes involved in fungal growth and reproduction. In addition, chitosan may act as a physical barrier that limits pathogen development and enhances host resistance mechanisms, thereby providing dual protective effects against postharvest diseases (Bautista-Baños et al., 2006; Hernández-Lauzardo et al., 2011; Romanazzi et al., 2018).

Table 2. The inhibitory effect of chitosan and plant extracts on the growth of fungal mycelium and spore production of *G. persicaria*

Treatments	Colony diameters (cm)			Inhibitory efficacy (%)			Spore density (10^5 spore mL^{-1})
	1 DAI	2 DAI	3 DAI	1 DAI	2 DAI	3 DAI	
Billy goat weed 2%	6.3^{ab}	8.0^{a}	8.5^{a}	0.0^{g}	0.0^{j}	0.0^{e}	83.0^{a}
Billy goat weed 4%	6.0^{c}	7.2^{cd}	8.5^{a}	0.0^{g}	0.0^{j}	0.0^{e}	84.0^{a}
Billy goat weed 6%	6.4^{a}	8.1^{a}	8.5^{a}	0.0^{g}	0.0^{j}	0.0^{e}	91.5^{a}
Billy goat weed 8%	6.2^{bc}	7.7^{ab}	8.5^{a}	0.0^{g}	0.0^{j}	0.0^{e}	94.5^{a}
Billy goat weed 10%	6.1^{c}	6.8^{d-f}	8.5^{a}	0.0^{g}	2.6^{ij}	0.0^{e}	96.0^{a}
Lemongrass 1%	5.0^{e}	6.7^{ef}	8.5^{a}	6.9^{e}	2.6^{ij}	0.0^{e}	29.5^{de}
Lemongrass 2%	5.2^{de}	6.7^{ef}	8.5^{a}	2.8^{f}	2.6^{ij}	0.0^{e}	25.5^{d-f}
Lemongrass 4%	5.1^{e}	7.1^{c-e}	8.5^{a}	5.7^{e}	0.0^{j}	0.0^{e}	24.0^{d-f}
Lemongrass 6%	5.1^{e}	7.5^{bc}	8.5^{a}	5.7^{e}	0.0^{j}	0.0^{e}	23.0^{d-f}
Lemongrass 8%	5.1^{e}	6.6^{f}	8.5^{a}	5.6^{e}	5.0^{i}	0.0^{e}	19.0^{ef}
Garlic 1%	3.8^{f}	6.0^{g}	8.5^{a}	29.1^{d}	11.8^{h}	0.0^{e}	43.5^{bc}
Garlic 2%	1.7^{g}	4.6^{h}	8.5^{a}	68.3^{c}	32.4^{g}	0.0^{e}	24.0^{d-f}
Garlic 4%	1.4^{h}	4.2^{i}	8.5^{a}	74.3^{b}	38.8^{f}	0.0^{e}	24.5^{d-f}
Garlic 6%	0.0^{i}	1.2^{k}	5.5^{b}	100.0^{a}	82.4^{c}	35.1^{d}	12.5^{f}
Garlic 8%	0.0^{i}	0.4^{l}	5.7^{b}	100.0^{a}	93.5^{b}	32.9^{d}	13.0^{f}
Chitosan 0.2%	0.0^{i}	2.8^{j}	4.9^{c}	100.0^{a}	58.8^{e}	42.1^{c}	12.5^{f}
Chitosan 0.4%	0.0^{i}	1.5^{k}	2.9^{d}	100.0^{a}	77.9^{d}	65.9^{b}	22.0^{d-f}
Chitosan 0.6%	0.0^{i}	0.0^{l}	0.6^{e}	100.0^{a}	100.0^{a}	92.7^{a}	32.0^{c-e}
Chitosan 0.8%	0.0^{i}	0.0^{l}	0.5^{e}	100.0^{a}	100.0^{a}	94.6^{a}	33.5^{cd}
Chitosan 1%	0.0^{i}	0.0^{l}	0.5^{e}	100.0^{a}	100.0^{a}	94.6^{a}	21.5^{d-f}
Control	5.4^{d}	6.8^{d-f}	8.5^{a}	0.0^{g}	0.0j	0.0^{f}	49.5^{b}
Significance	**	**	**	**	**	**	**
CV (%)	4.74	7.16	3.05	4.08	6.89	10.90	23.54

*Note: Values in the same column followed by the same letter are not significantly different ($p > 0.05$); ** indicates significant differences at $p < 0.01$.*

Garlic extract at concentrations of 6–8% also exhibited notable antifungal activity during the early stages of incubation. However, its inhibitory effect declined over time, suggesting limited persistence of the bioactive compounds responsible for pathogen suppression. This observation is consistent with previous reports indicating that allicin and related organosulfur compounds exhibit strong antimicrobial activity but are highly unstable and prone to degradation during storage (Ankri & Mirelman, 1999).

Conversely, Billy goat weed extract appeared to stimulate fungal growth and sporulation. Lemongrass extract did not significantly suppress mycelial growth but reduced sporulation of *G. persicaria*. The limited efficacy observed may be attributed to the use of aqueous extracts, which exhibit lower antifungal activity than essential oils or active fractions. This result is consistent with studies showing that the antifungal efficacy of lemongrass is primarily observed in essential oils with concentrated monoterpene content, whereas

the efficacy of water extracts is often limited (Burt, 2004; Tzortzakis & Economakis, 2007).

Overall, *in vitro* results confirmed that chitosan ≥ 0.6% was the most potent and stable inhibitor of *G. persicaria*, whereas garlic extract at 6-8% showed significant but primarily short-term efficacy. These treatments were therefore selected for the postharvest treatment experiment on Barbados cherry fruits.

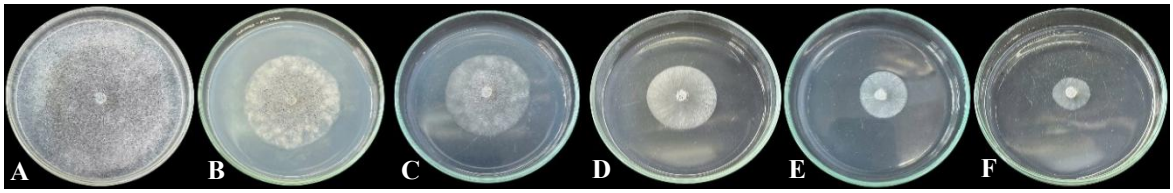

Figure 5. The inhibitory effect of chitosan and plant extracts on the growth of *G. persicaria* mycelia at 3 DAI

Control (A), Garlic 6% (B), Garlic 8% (C), Chitosan 0.6% (D), Chitosan 0.8% (E), and Chitosan 1% (F)

3.3. The efficacy of chitosan and garlic extract in controlling Barbados cherry fruit rot caused by *G. persicaria* under *in vivo* conditions

Under storage conditions (28.3°C and 92% humidity), all chitosan and garlic treatments effectively delayed disease development during the initial stages after inoculation. However, disease incidence and lesion expansion gradually increased over time, reflecting the progressive nature of soft rot caused by *G. persicaria*. Despite this increase, the treated fruits consistently exhibited lower disease levels than the untreated ones, confirming the protective effects of both chitosan and garlic extract.

Table 3. Percentage of fruit rot and lesion area ratio/fruit of Barbados cherry caused by the fungus *G. persicaria* at different time points after treatment (T=28.3°C, H=92%)

Treatments	Percentage of fruit rot (%)				Lesion area ratio/fruit (%)			
	1 DAT	3 DAT	5 DAT	7 DAT	1 DAT	3 DAT	5 DAT	7 DAT
Garlic 6 %	0.0	5.0^{b}	50.0^{b}	82.5^{bc}	0.0	2.3^{b}	27.3^{b}	74.5^{ab}
Garlic 8 %	0.0	0.0^{b}	45.0^{b}	90.0^{ab}	0.0	0.0^{b}	26.8^{b}	81.8^{ab}
Chitosan 0.6 %	0.0	0.0^{b}	52.5^{b}	82.5^{bc}	0.0	0.0^{b}	25.1^{b}	65.6^{b}
Chitosan 0.8%	0.0	0.0^{b}	37.5^{b}	82.5^{bc}	0.0	0.0^{b}	16.6^{bc}	61.8^{b}
Chitosan 1%	0.0	0.0^{b}	32.5^{b}	70.0^{c}	0.0	0.0^{b}	8.9^{c}	63.0^{b}
Control	2.5	30.0^{a}	75.0^{a}	100.0^{a}	1.0	28.0^{a}	60.3^{a}	92.8^{a}
Significance	ns	**	*	**	ns	**	**	ns
CV (%)	19.05	22.63	20.81	10.34	5.32	19.53	22.98	17.05

*Note: data were converted to arcsin√x before statistical processing; values in the same column followed by the same letter are not significantly different (p > 0.05), ** indicates significant differences at p < 0.01; ns: not significant.*

Chitosan at 0.8–1.0% exhibited higher and more consistent control efficacy, as indicated by consistently lower disease incidence and lesion area throughout the observation period (Table 3). The superior efficacy of chitosan may be attributed to its ability to form a semipermeable film on the fruit surface, which restricts spore penetration and germination while simultaneously reducing water loss and delaying the deterioration of host tissue quality. In addition, chitosan has been reported to elicit defense responses in plant tissues, thereby enhancing fruit resistance during storage (Bautista-Baños et al., 2006; Hernández-Lauzardo et al., 2011; Romanazzi et al., 2018).

Garlic extract at 6–8% also reduced disease severity compared with the untreated control; however, its efficacy was less consistent than that of chitosan, particularly under extended storage conditions. This is consistent with the biological properties of allicin, the principal antifungal compound in garlic, which is prone to degradation and loss of activity over time (Ankri & Mirelman, 1999).

The disease-suppression data further confirmed chitosan's superior performance during fruit storage. At 1.0%, chitosan provided the greatest level of protection, reducing disease incidence by 56.7% and lesion area by 85.3% at 5 DAT. Although efficacy

declined slightly at later assessment times, chitosan consistently outperformed garlic extract and maintained significantly greater disease suppression throughout the storage period (Table 4).

Table 4. The efficacy of chitosan and garlic extract in controlling Barbados cherry fruit rot caused by the fungus *G. persicaria* at different time points after treatment (T=28.3 °C, H=92%)

Treatments	Efficacy in reducing the percentage of fruit rot (%)		Efficacy in reducing lesion area ratio/fruit (%)	
	5 DAT	7 DAT	5 DAT	7 DAT
Garlic 6%	33.4^{bc}	17.5^{b}	54.8^{c}	19.7^{b}
Garlic 8%	40.0^{b}	10.0^{c}	55.6^{c}	11.9^{c}
Chitosan 0.6%	30.0^{c}	17.5^{b}	58.4^{c}	29.3^{a}
Chitosan 0.8%	50.0^{a}	17.5^{b}	72.4^{b}	33.5^{a}
Chitosan 1%	56.7^{a}	30.0^{a}	85.3^{a}	32.1^{a}
Control	0.0^{d}	0.0^{d}	0.0^{d}	0.0^{d}
Significance	**	**	**	**
CV (%)	9.89	8.14	7.85	8.28

*Note: data were converted to arcsin√x before statistical processing; values in the same column followed by the same letter are not significantly different ($p > 0.05$); ** indicates significant differences at $p < 0.01$.*

The *in vivo* experimental results are consistent with *in vitro* results and recent reports on the effective control of postharvest rot diseases caused by *G. persicaria* on various tropical fruits (Zhang et al., 2020; Souza et al., 2023). Chitosan at 0.8–1.0% proved to be the most effective treatment for managing postharvest soft rot of Barbados cherry, whereas garlic extract at 6-8% may serve as a supplementary measure under short-term storage conditions.

4. CONCLUSION

This study demonstrated that postharvest soft rot is one of the major postharvest diseases affecting Barbados cherry in Dong Thap Province, Viet Nam, and identified *Gilbertella persicaria* as its causal agent based on morphological characteristics, pathogenicity tests, and ITS rDNA sequence analysis. Among the tested treatments, chitosan showed the strongest and most consistent antifungal activity against *G. persicaria* under both *in vitro* and *in vivo* conditions. Chitosan at concentrations of 0.8–1.0% effectively suppressed disease development, reduced lesion expansion, and provided superior protection compared with garlic and other plant extracts. These findings highlight the potential of chitosan as a safe, environmentally friendly, and effective alternative for managing postharvest soft rot in Barbados cherry. Further studies should focus on optimizing application strategies and evaluating treatment performance under commercial storage conditions.

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CONFLICT OF INTEREST

The authors declare no conflict of interest.

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