

Management of Strawberry Grey Mold Disease Using Biocontrol Agents and Plant Extracts

P. Sakthi Priya^{1*}, Srushtideep Angidi², Uday Kumar Thera³, S. V. Nandeesh¹, Thangaswamy Rajesh¹

¹Department of Plant Pathology, College of Post Graduate Studies in Agricultural Sciences, Central Agricultural University, Imphal, India

²Department of Plant Pathology, North Dakota State University, Fargo, ND, USA

³Department of Plant Science and Landscape Architecture, University of Maryland, College Park, MD, USA

Email: *sakthipriya161098@gmail.com

How to cite this paper: Sakthi Priya, P., Angidi, S., Thera, U.K., Nandeesh, S.V. and Rajesh, T. (2024) Management of Strawberry Grey Mold Disease Using Bio-control Agents and Plant Extracts. *American Journal of Plant Sciences*, 15, 538-551. <https://doi.org/10.4236/ajps.2024.157037>

Received: May 9, 2024

Accepted: July 22, 2024

Published: July 25, 2024

Copyright © 2024 by author(s) and Scientific Research Publishing Inc. This work is licensed under the Creative Commons Attribution International License (CC BY 4.0). <http://creativecommons.org/licenses/by/4.0/>



Open Access

Abstract

Strawberry (*Fragaria × ananassa* Duch.) is a significant global soft fruit crop, prized for its nutrient content and pleasant flavor. However, diseases, particularly grey mold caused by *Botrytis cinerea* Pers. Fr. poses major constraints to strawberry production and productivity. Grey mold severely impacts fruit quality and quantity, diminishing market value. This study evaluated five *B. cinerea* isolates from various locations in the Ri-Bhoi district of Meghalaya. All isolates were pathogenic, with isolate SGM 2 identified as highly virulent. Host range studies showed the pathogen-producing symptoms in the fava bean pods, marigold, gerbera, and chrysanthemum flowers and in the fava bean, gerbera, and lettuce leaves. *In vitro* tests revealed that neem extract (15% w/v) achieved the highest mycelial growth inhibition at 76.66%, while black turmeric extract (5% w/v) had the lowest inhibition at 9.62%. Dual culture methods with bio-control agents indicated that *Bacillus subtilis* recorded the highest mean inhibition at 77.03%, while *Pseudomonas fluorescens* had the lowest at 20.36% against the two virulent isolates. Pot evaluations demonstrated that *B. subtilis* resulted in the lowest percent disease index at 20.59%, followed by neem extract at 23.31%, with the highest disease index in the control group at 42.51%. Additionally, *B. subtilis* significantly improved plant growth, yielding an average of 0.32 kg compared to 0.14 kg in the control. The promising results of *B. subtilis* and neem leaf extract from this study suggest their potential for eco-friendly managing grey mold in strawberries under field conditions.

Keywords

Strawberry Grey Mold, BCA, Plant Extracts, *Botrytis cinerea*

1. Introduction

Strawberry (*Fragaria × ananassa*) is a vital soft fruit crop cultivated globally on over 370,000 hectares [1]. It is a significant source of macro- and micronutrients, vitamins, and health-promoting antioxidants, contributing to a beneficial human diet [2] [3]. The strawberry plant is a perennial herbaceous species with short stems (crowns) and densely spaced leaves, producing complex accessory and aggregate fruits composed of achenes and a receptacle [4]. Achenes are small, single-seeded fruits, while the receptacle is anatomically equivalent to floral meristem tissue [5]. *Fragaria × ananassa* is an allo-octoploid ($2n = 8x = 56$), originating as a synthetic hybrid between the octoploid species *Fragaria chiloensis* and *Fragaria virginiana* [6] [7].

Strawberry fruits are rich in sugars, dietary fiber, vitamins, and amino acids and can be processed into high-value by-products [8]. However, strawberries are highly susceptible to microbial infections, both pre- and post-harvest, particularly fungal infections, which significantly impact their economic value [9]. One of the most devastating strawberry cultivation diseases is grey mold, caused by the necrotrophic fungus *B. cinerea*. This pathogen typically results in a 10% to 25% yield reduction and can cause severe losses exceeding 50% [8] [10] [11]. Additionally, *B. cinerea* can infect post-harvest strawberry fruits, leading to substantial economic losses during storage, transportation, and sales [12].

Traditionally, chemical control has been the most effective strategy for managing strawberry grey mold [13]-[15]. However, the frequent and extensive use of various fungicides has led to increasingly serious “3R” problems: Resistance, Resurgence, and Residue [13]. Consequently, there is an urgent need to explore new strategies or alternatives to chemical fungicides for controlling strawberry grey mold.

Biocontrol of *B. cinerea* using plant extracts or biocontrol agents (BCAs) has emerged as a viable alternative to chemical control due to its environmentally friendly nature and lack of cross-resistance [10] [16] [17]. Plant extracts, such as neem, aloe vera, lantana, and black turmeric, have demonstrated significant efficacy in controlling strawberry grey mold *in vitro* [18]-[20]. Various BCAs, including fungi, *B. species*, and actinomycetes, have been applied to combat strawberry grey mold through different modes of action, such as induced resistance, competition for nutrients, production of bioactive metabolites, and parasitism [11] [21].

2. Materials and Methods

2.1. Sample Collection and Pathogenicity

A total of six isolates were collected from different parts of the Ri-Bhoi district in Meghalaya. The samples were obtained from CPGSAS, Umiam, located at 25°40'52.32"N latitude, 91°54'41.04"E longitude, and 1010 meters above mean sea level. These isolates were tested on strawberries to identify virulent isolates (Table S1).

The ability of isolates to cause grey mold was assessed *in vitro* on strawberry fruits. Uniform-sized, infection-free strawberry fruits were plucked from healthy plants. After surface sterilization with 70% ethanol, the fruits were injured with a needle and inoculated with a 5 mm mycelial disc from a 10-day-old culture. Inoculated fruits were then incubated at $27 \pm 1^\circ\text{C}$, with uninoculated fruits maintained as controls. Three replications were maintained for each treatment. Symptom development, measured based on the size of the infection area (cm^2), was monitored regularly. The symptoms of artificially inoculated fruits were compared with those from naturally infected plants.

2.2. Host Pathogenicity Test

A host pathogenicity test was conducted to determine whether *B. cinerea* causes infections on other crops grown near the strawberry farm. The most virulent isolate, SGM 4, was selected for this study (Table S2). Leaves and flowers of five different plants were tested by inoculating them with a conidial suspension from a 10-day-old pathogen culture [22].

Plant parts were surface sterilized using 1% sodium hypochlorite and washed with sterile distilled water. After air drying on sterilized filter paper under laminar airflow, they were inoculated and placed in humid chambers at room temperature. Observations were made after 3 days of inoculation and continued up to 10 days. The total percentage of the area infected was calculated for each replication.

2.3. Molecular Characteristics of SGM2 and SGM4

Pure fungal DNA was isolated following the HiPurAR protocol (HiMedia MB543). Fungal mycelia from a 10-day-old culture were used for DNA isolation. The isolated DNA was amplified using PCR with ITS primers (ITS1F and ITS4R), and the PCR products were separated and visualized on agarose gel [23]. Sequence data obtained from 1st BASE, Eurofins Genomics India Pvt Ltd., Bangalore, were analyzed using BLASTn for homology search against reference sequences.

2.4. In Vitro Management of *B. cinerea* Using Plant Extracts and Biological Control Agents

The efficacy of five locally available botanicals and one fungicide (carbendazim) was tested against *B. cinerea* isolates SGM2 and SGM4 using the poisoned food technique [24]. Aqueous leaf extracts of the botanicals and carbendazim at different concentrations were incorporated into Potato Dextrose Agar (PDA) medium. Mycelial growth inhibition was assessed, and percent inhibition was calculated using Vincent's formula [25]. Additionally, three fungal antagonists (*Trichoderma harzianum*, *T. asperellum*, *Metarhizium anisopliae*) and two bacterial antagonists (*P. fluorescens*, *B. subtilis*) were screened for their inhibition potential against *B. cinerea* using the dual culture technique [26].

2.5. In Vivo Management of SGM in Strawberry

Based on the results from *in vitro* studies, the two most effective plant extracts

and antagonists were selected for *in vivo* tests on strawberry plants and fruits. Healthy plants were inoculated with *B. cinerea*, and the efficacy of treatments was evaluated by measuring lesion areas and fruit rot inhibition.

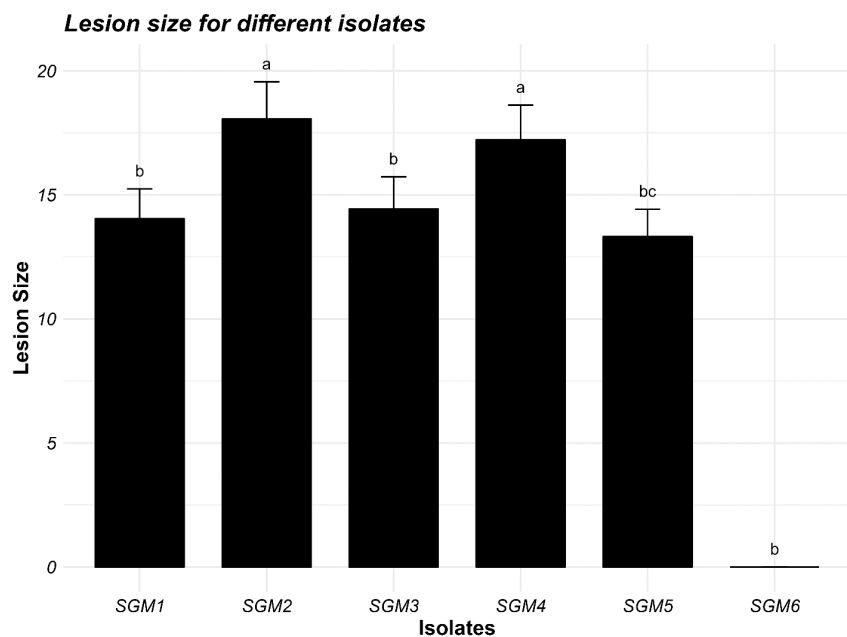
2.6. Experimental Design and Data Analysis

All experiments were conducted using one-factor and two-factor completely randomized designs (CRD). Levene's test was employed to assess the homogeneity of variance between replications, and Tukey's test was used to determine significant differences between treatments using R-software [27].

3. Results

3.1. Pathogenicity Test

Significant differences in lesion sizes were observed in the pathogenicity test to evaluate the virulence of various fungal isolates. Among the isolates tested, SGM 2 and SGM 4 exhibited the highest degree of pathogenicity. SGM 2 and SGM 4 produced the largest lesion sizes of 18.06 cm² and 17.22 cm², respectively, significantly different than other isolates (Figure 1). These isolates also demonstrated faster symptom development, producing visible greyish mycelial growth on the fruit within 7 days of inoculation. In contrast, the remaining isolates, SGM 1, SGM 3, and SGM 5, showed moderate pathogenicity with lesion sizes of 14.04 cm², 14.43 cm², and 13.32 cm², respectively (Figure 1). These findings indicate that SGM 2 and SGM 4 are more virulent than the other isolates tested, as they induced larger lesions and exhibited a quicker onset of symptoms.



In this experiment, different isolates were evaluated for their virulence on strawberries, the x-axis representing isolates, and the y-axis representing the lesion size in centimeters of the infection.

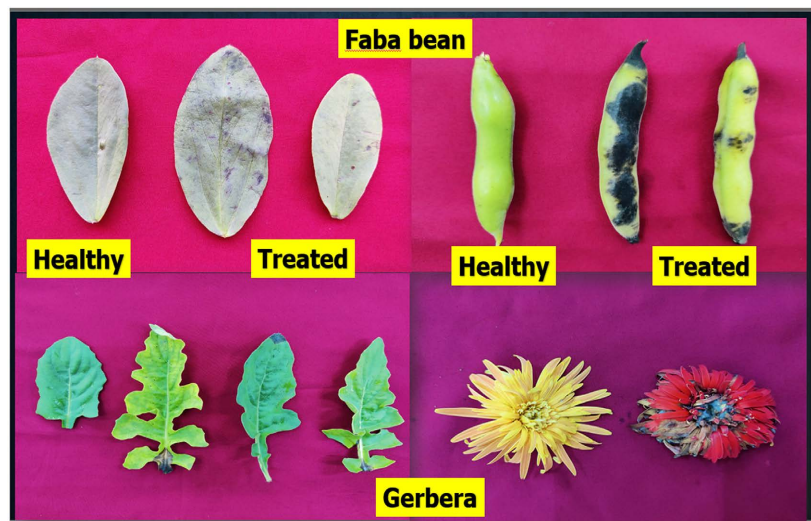
Figure 1. Pathogenic variability of different isolates of *B. cinerea* on infected fruits.

3.2. Host Pathogenicity Test

In the following study, the host range of the SGM 4 isolate was evaluated under *in vitro* conditions. Five host plants from the families Asteraceae and Fabaceae were selected. The results indicated a positive reaction on all tested plants, with varying degrees of symptom development on different parts (Figure 2). The symptoms of Faba bean pods appeared four days post-inoculation, from purple brown to blackish lesions, followed by grey mold growth covering the entire leaf (Figure 2). Faba bean leaves, brown to black elongated spore masses were observed on the midrib and veins (Figure 2). Flowers exhibited greyish fungal growth on the disc florets and coronal filaments (Figure 2). These findings demonstrate that the SGM 4 isolate of *Botrytis* has a broad host range, capable of infecting multiple plant species within the Asteraceae and Fabaceae families.



(a)



(b)

In this experiment, different hosts were inoculated with isolate SGM2 to evaluate the broad-spectrum infection. The hosts are chrysanthemums, marigolds, lettuce, faba bean leaf, pod, and gerbera.

Figure 2. (a) and (b): Symptoms of *B. cinerea* on various hosts.

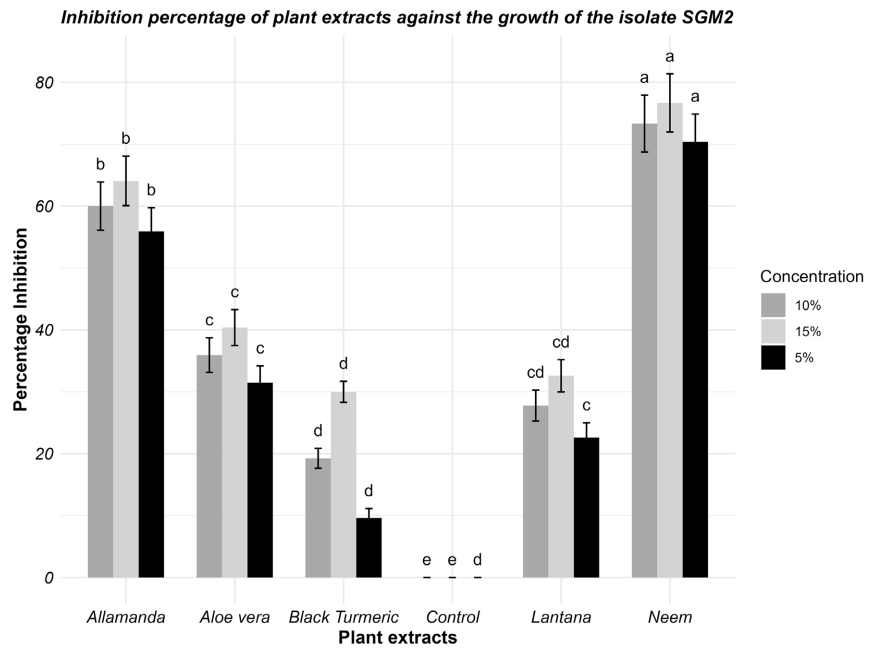
3.3. Molecular Characteristics of SGM2 and SGM4

Universal ITS oligonucleotide primers, ITS1 F and ITS4 R, were used to confirm the sequence of SGM 2 and SGM 4 isolates. The obtained sequences, identified as FI774, confirmed the pathogens as *B. cinerea*, the organism causing grey mold. In gel documentation, the isolates were amplified between 500 - 600 bp (Figure S1). The sequences obtained from the PCR amplification were compared using the BLAST analysis tool from the National Centre for Biotechnology Information (NCBI). The sequences were confirmed and deposited in the NCBI Gene Bank. It was observed that the sequences obtained were 98% similar to the reference sequences from the Gene Bank.

3.4. In Vitro Management of *B. cinerea* Using Plant Extracts and Biological Control Agents

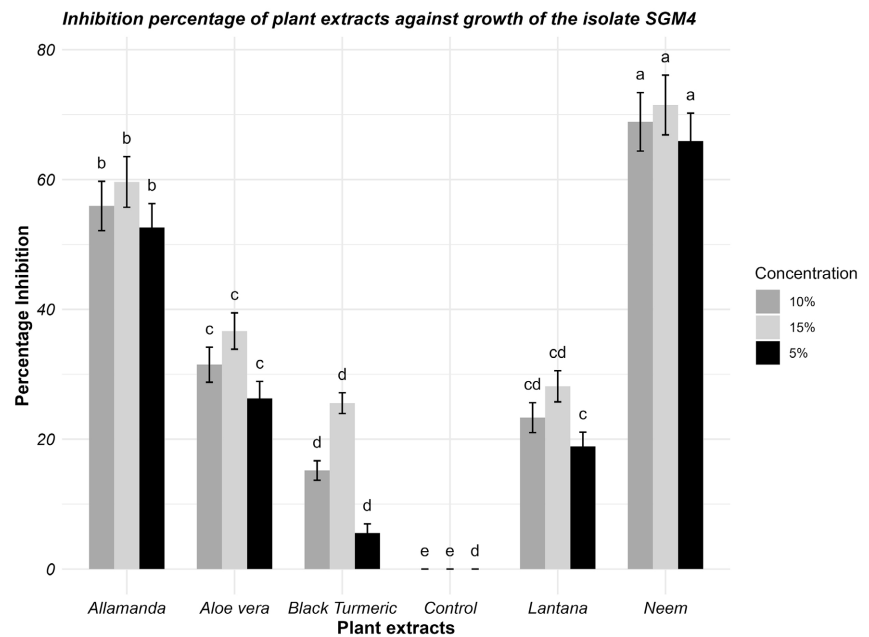
The study evaluated the antifungal activity of various plant extracts against *B. cinerea* isolates SGM 2 and SGM 4. The results indicate that neem extract consistently exhibited the highest inhibition percentages across both isolates and all concentrations tested. At 15% concentration, neem extract achieved inhibition rates of 76.67% for SGM 2 and 71.48% for SGM 4, indicating its potent antifungal properties (Figure 3 and Figure 4). Allamanda leaf extract also showed significant antifungal activity, with inhibition percentages ranging from 52.59% to 64.07% across different concentrations and isolates. Aloe vera extract exhibited moderate inhibition, while lantana and black turmeric extracts demonstrated lower effectiveness against *B. cinerea* isolates at all concentrations tested (Figure 3 and Figure 4). Overall, the findings highlight the concentration-dependent nature of plant extracts' antifungal efficacy against *B. cinerea* isolates. Neem and Allamanda extracts emerged as the most effective biocontrol agent among the plant extracts tested, suggesting their potential for use in integrated disease management strategies against grey mold in agricultural settings.

The antagonistic potential of five biocontrol agents (BCAs) *B. subtilis*, *T. harzianum*, *T. viride*, *M. anisopliae*, and *P. fluorescens*—was evaluated against two isolates, SGM 2 and SGM 4, using the dual culture technique. Significant differences ($p < 0.05$) were observed among the treatments (Figure 5). For isolate SGM 2, *B. subtilis* and *T. harzianum* exhibited the highest inhibition of mycelial growth, demonstrating strong antagonistic activity with percentage inhibitions of 72.59% and 68.14%, respectively. Similarly, for isolate SGM 4, significant differences ($p < 0.05$) were observed among the treatments. *B. subtilis* showed the highest inhibition of mycelial growth at 77.03%, followed by *T. harzianum* at 66.66%, indicating their effectiveness as biocontrol agents (Figure 5). In contrast, *P. fluorescens* exhibited the least inhibition, highlighting its lower efficacy than the other BCAs. These findings underscore the potential of *B. subtilis* and *T. harzianum* as effective biocontrol agents against isolates, with their efficacy varying significantly across different isolates.



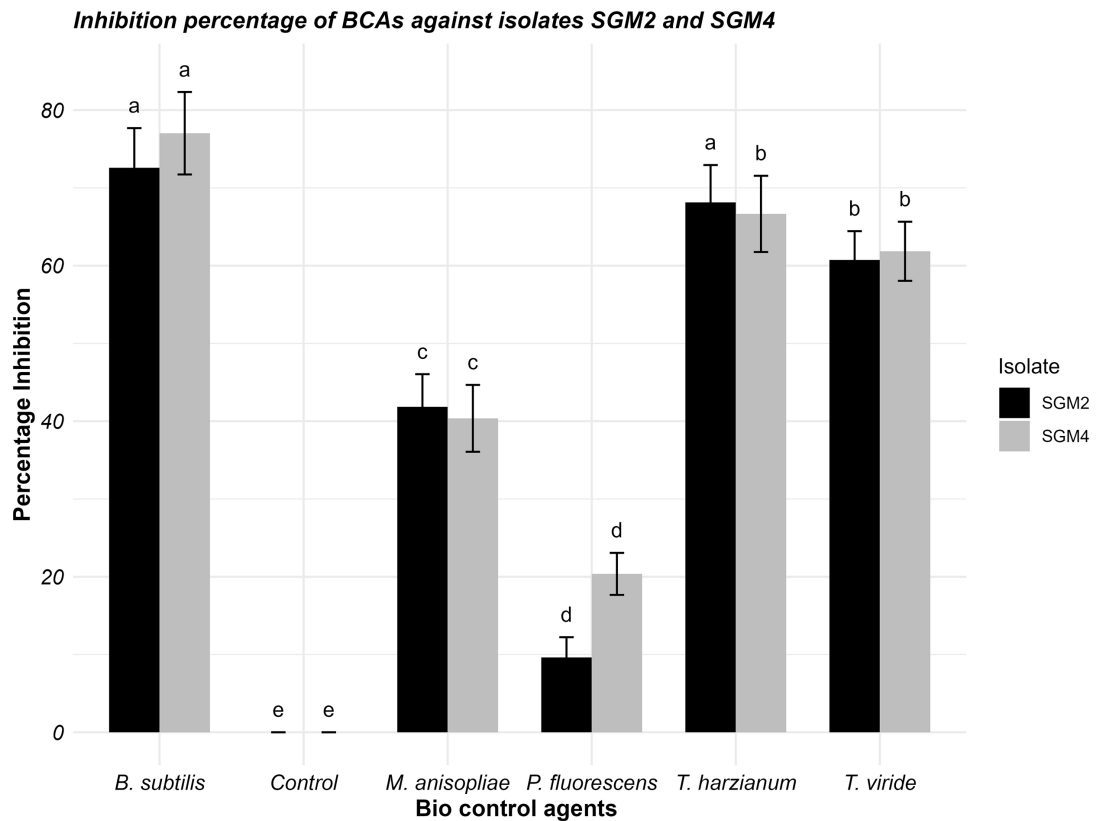
In this experiment, five biocontrol agents were evaluated against *B. cinerea* isolate SGM2, and control carbendazim in in-vitro conditions. The X-axis represents the neem, aloe vera, lantana, black turmeric, and allamanda. The Y-axis represents the percentage inhibition of the pathogen.

Figure 3. Bar diagram showing percent inhibition of plant extracts against the growth of *B. cinerea* isolates SGM2.



In this experiment, five biocontrol agents were evaluated against *B. cinerea* isolate SGM4, and control carbendazim in in-vitro conditions. The X-axis represents the neem, aloe vera, lantana, black turmeric, and allamanda. The Y-axis represents the percentage inhibition of the pathogen.

Figure 4. Bar diagram showing percent inhibition of plant extracts against the growth of *B. cinerea* isolates SGM4.



In this experiment, five biocontrol agents were evaluated against *B. cinerea* isolate SGM2, SGM4 and control carbendazim in in-vitro conditions. X-axis represents the biocontrol agents *Bacillus subtilis*, *Trichoderma harzianum*, *Trichoderma viride*, *Metarhizium anisopliae*, and *Pseudomonas fluorescens*. The Y-axis represents the percentage inhibition of the pathogen.

Figure 5. Bar diagram showing percent inhibition of BCAs against *B. cinerea* isolates SGM2 and SGM 4.

3.5. In Vivo Management of SGM in Strawberry

The *in vitro* efficacy of various treatments, including neem and allamanda botanical extracts, *B. subtilis* and *T. harzianum* biocontrol agents, and carbendazim fungicide, which was initially effective against grey mold, were assessed through a pot culture experiment. The disease progression index (PDI) values tracked the effectiveness of the treatment over three spray intervals. Initially, all treatments exhibited high PDI values, indicating significant disease presence, but subsequent sprays led to notable PDI reductions across all treatments. *B. subtilis* consistently achieved the lowest PDI values throughout, achieving the highest disease reduction (51.56%) after 2nd spray. Neem extract also demonstrated effective disease control, particularly notable in later intervals, with a disease reduction of 45.17% at 2nd spray (Table 1). Carbendazim, while effective, showed marginally higher PDI values compared to *B. subtilis* and neem extract, indicating slightly lesser efficacy in disease control. These results underscore significant differences in efficacy among the treatments, highlighting *B. subtilis* and neem extract as particularly effective options for managing grey mold in strawberry cultivation.

Table 1. *In vivo* evaluation of different plant extracts, BCA's and fungicide against *B. cinerea*

Treatments	Per cent Disease Index (PDI)				Average PDI	Reduction of Diseases
	Before spray	1 st spray	2 nd spray	3 rd spray		
T1 (<i>T. harzianum</i>)	30.46 ± 7.66 ¹ (33.49) ²	33.96 ± 4.47 (35.64)	28.21 ± 2.05 (32.08)	24.71 ± 1.94 (29.80)	29.33 ± 2.36 (32.79)	31.00 (33.83)
T2 (<i>B.subtilis</i>)	19.03 ± 5.45 (25.86)	25.28 ± 3.46 (30.18)	21.28 ± 1.88 (27.47)	16.78 ± 2.36 (24.18)	20.59 ± 1.64 (26.98)	51.56 (45.89)
T3 (Neem)	22.06 ± 4.43 (28.01)	28.06 ± 2.62 (31.99)	23.31 ± 3.24 (28.87)	19.81 ± 2.16 (26.43)	23.31 ± 1.78 (28.86)	45.17 (42.22)
T4 (Allamanda)	31.02 ± 3.72 (33.85)	34.52 ± 2.36 (35.98)	30.02 ± 1.10 (33.22)	26.27 ± 2.56 (30.83)	30.45 ± 1.83 (33.49)	28.36 (32.17)
T5 (Carbendazim)	28.89 ± 3.37 (32.51)	29.39 ± 3.21 (32.82)	25.64 ± 1.81 (30.42)	22.14 ± 2.52 (28.06)	26.51 ± 0.81 (30.99)	37.63 (37.84)
T6 (Control)	35.09 ± 3.08 (36.32)	38.09 ± 2.32 (38.11)	45.34 ± 3.83 (42.32)	51.54 ± 5.94 (46.05)	42.51 ± 1.25 (40.69)	0.00 (0)
SE(m)	2.44	1.58	1.25	1.61	0.84	0.76
CD (p = 0.05)	7.25	4.71	3.72	4.79	2.51	1.49

¹Values are mean of 3 replications and ± standard deviation, ²Values parenthesis indicate arc sine transformed value, SE (m): Mean standard error; CD: Critical difference.

4. Discussion

The present research on strawberry grey mold involved collecting and analyzing five isolates from the Ri-Bhoi district of Meghalaya. Among these, isolates SGM 2 and SGM 4 were selected for *in vitro* testing based on their pathogenicity. Eco-friendly products, including five biocontrol agents (BCAs) and various plant extracts, were tested against these isolates. The most effective BCAs and plant extracts were tested in a pot culture experiment with the highly virulent isolate SGM 2. Pathogenicity tests using an *in vitro* detached fruit technique showed variations in disease symptoms among the isolates. These studies indicated that all five isolates produced typical grey mold symptoms, with lesion sizes ranging from 13.32 to 18.06 cm². Isolate SGM 2 caused the highest lesion size of 18.06 cm², while isolate SGM 5 caused the lowest at 13.32 cm². These findings are consistent with the symptoms [28] [29].

The host range study revealed that *B. cinerea* infected all five hosts from the Asteraceae and Fabaceae families, confirming its host non-specificity, as reported by Droby and Litcher [30] and Williamson *et al.* [31].

In vitro management using plant extracts showed that neem extract at 15% concentration was highly effective, inhibiting 71% - 76% of *B. cinerea* growth. Lower concentrations (10% and 5%) also showed significant inhibition, followed by allamanda leaf extract at 15% and 10%. Black turmeric extract was the least effective, inhibiting 25.55% - 30% growth, consistent with findings by Ghosh [32] and Wahmare *et al.* [33].

The antagonistic potential of BCAs revealed that *B. subtilis* was the most effective, inhibiting mycelial growth by 72.59% - 77.03%. *T. harzianum* showed

substantial inhibition, while *M. anisopliae* and *P. fluorescens* were the least effective. These findings were consistent with Freeman *et al.* [34] and Yue *et al.* [35]. The best treatments from the *in vitro* assays were evaluated in pot culture using the most virulent isolate, SGM 2. *B. subtilis* showed the highest efficacy, with the lowest percent disease index (PDI) of 20.59% after 15 days of the third spray. Allamanda showed the highest PDI of 30.45%. Neem extract at 10% concentration reduced grey mold by 45.17%, consistent with Gholamnezhad [36] studies.

5. Conclusion

Our results indicated the potential of BCAs and plant extract methods as viable alternatives to traditional chemical fungicides, addressing the challenges of resistance, resurgence, and residue associated with chemical treatments. Implementing these eco-friendly strategies could enhance strawberry cultivation's sustainability and economic viability by reducing losses due to grey mold.

Acknowledgements

The authors extend gratitude to CPGS-AS, CAU (I) for providing the necessary facilities and constant support and encouragement throughout the research.

Authors' Contributions

SP, NSV, and RT contributed to the experimental design, data collection, and ideology of the experiment. SA and UKT analyzed the data and wrote the manuscript.

Availability of Data and Materials

All data produced during this study were included in this article.

Conflicts of Interest

The authors declare no conflicts of interest regarding the publication of this paper.

References

- [1] FAOSTAT (2018) Food and Agriculture Organization of the United Nations. <http://faostat.fao.org>
- [2] Giampieri, F., Forbes-Hernandez, T.Y., Gasparri, M., Alvarez-Suarez, J.M., Afrin, S., Bompadre, S., *et al.* (2015) Strawberry as a Health Promoter: An Evidence Based Review. *Food & Function*, **6**, 1386-1398. <https://doi.org/10.1039/c5fo00147a>
- [3] Basu, A., Nguyen, A., Betts, N.M. and Lyons, T.J. (2013) Strawberry as a Functional Food: An Evidence-Based Review. *Critical Reviews in Food Science and Nutrition*, **54**, 790-806. <https://doi.org/10.1080/10408398.2011.608174>
- [4] Darrow, G.M. (1966) *The Strawberry: History, Breeding and Physiology*. Holt, Ri-

- nehart and Winston, Inc.
- [5] Hollender, C.A., Geretz, A.C., Slovin, J.P. and Liu, Z. (2011) Flower and Early Fruit Development in a Diploid Strawberry, *Fragaria Vesca*. *Planta*, **235**, 1123-1139. <https://doi.org/10.1007/s00425-011-1562-1>
- [6] Edger, P.P., Poorten, T.J., VanBuren, R., Hardigan, M.A., Colle, M., McKain, M.R., *et al.* (2019) Origin and Evolution of the Octoploid Strawberry Genome. *Nature Genetics*, **51**, 541-547. <https://doi.org/10.1038/s41588-019-0356-4>
- [7] Rousseau-Gueutin, M., Lerceteau-Kohler, E., Barrot, L., Sargent, D.J., Monfort, A., Simpson, D., Denoyes-Rothan, B., *et al.* (2008) Comparative Genetic Mapping between Octoploid and Diploid *Fragaria* Species Reveals a High Level of Colinearity between Their Genomes and the Essentially Disomic Behavior of the Cultivated Octoploid Strawberry. *Genetics*, **179**, 2045-2060.
- [8] Hassan, E.A., Mostafa, Y.S., Alamri, S., Hashem, M. and Nafady, N.A. (2021) Bio-safe Management of Botrytis Grey Mold of Strawberry Fruit by Novel Bioagents. *Plants*, **10**, Article 2737. <https://doi.org/10.3390/plants10122737>
- [9] Garrido, C., Carbú, M., Fernández-Acero, F.J., González-Rodríguez, V.E. and Cantoral, J.M. (2011) New Insights in the Study of Strawberry Fungal Pathogens. *Genes Genomes Genomics*, **5**, 24-39.
- [10] Nguyen, X.H., Naing, K.W., Lee, Y.S., Moon, J.H., Lee, J.H. and Kim, K.Y. (2014) Isolation and Characteristics of Protocatechuic Acid from *Paenibacillus elgii* HOA73 against *Botrytis cinerea* on Strawberry Fruits. *Journal of Basic Microbiology*, **55**, 625-634. <https://doi.org/10.1002/jobm.201400041>
- [11] Zhang, Y., Wang, C., Su, P. and Liao, X. (2015) Control Effect and Possible Mechanism of the Natural Compound Phenazine-1-Carboxamide against *Botrytis cinerea*. *PLOS ONE*, **10**, e0140380. <https://doi.org/10.1371/journal.pone.0140380>
- [12] Chen, P., Chen, R. and Chou, J. (2018) Screening and Evaluation of Yeast Antagonists for Biological Control of *Botrytis cinerea* on Strawberry Fruits. *Mycobiology*, **46**, 33-46. <https://doi.org/10.1080/12298093.2018.1454013>
- [13] Moura, G.G.D.D., Barros, A.V.D., Machado, F., Martins, A.D., Silva, C.M.D., Durango, L.G.C., *et al.* (2021) Endophytic Bacteria from Strawberry Plants Control Gray Mold in Fruits *via* Production of Antifungal Compounds against *Botrytis cinerea* L. *Microbiological Research*, **251**, Article ID: 126793. <https://doi.org/10.1016/j.micres.2021.126793>
- [14] Fontana, D.C., Neto, D.D., Pretto, M.M., Mariotto, A.B., Caron, B.O., Kulczynski, S.M., *et al.* (2021) Using Essential Oils to Control Diseases in Strawberries and Peaches. *International Journal of Food Microbiology*, **338**, Article ID: 108980. <https://doi.org/10.1016/j.ijfoodmicro.2020.108980>
- [15] Zhou, L., Song, C., Muñoz, C.Y. and Kuipers, O.P. (2021) *Bacillus* *Cabrialesii* BH5 Protects Tomato Plants against *Botrytis cinerea* by Production of Specific Antifungal Compounds. *Frontiers in Microbiology*, **12**, Article 707609. <https://doi.org/10.3389/fmicb.2021.707609>
- [16] Šernaitė, L., Rasiukevičiūtė, N. and Valiuškaitė, A. (2020) The Extracts of Cinnamon and Clove as Potential Biofungicides against Strawberry Grey Mould. *Plants*, **9**, Article 613. <https://doi.org/10.3390/plants9050613>
- [17] Iqbal, M., Jützel, M., França, S.C., Wäckers, F., Andreasson, E. and Stenberg, J.A. (2022) Bee-Vectored *aureobasidium Pullulans* for Biological Control of Gray Mold in Strawberry. *Phytopathology*, **112**, 232-237. <https://doi.org/10.1094/phyto-05-21-0205-r>

- [18] Yang, C., Lu, J., Xu, M., Shi, X., Song, Z., Chen, T., et al. (2022) Evaluation of Chitosan Coatings Enriched with Turmeric and Green Tea Extracts on Postharvest Preservation of Strawberries. *LWT*, **163**, Article ID: 113551. <https://doi.org/10.1016/j.lwt.2022.113551>
- [19] Hernández-Ceja, A., Loeza-Lara, P.D., Espinosa-García, F.J., García-Rodríguez, Y.M., Medina-Medrano, J.R., Gutiérrez-Hernández, G.F., et al. (2021) *In Vitro* Antifungal Activity of Plant Extracts on Pathogenic Fungi of Blueberry (*Vaccinium* sp.). *Plants*, **10**, Article 852. <https://doi.org/10.3390/plants10050852>
- [20] De Rodríguez, D.J., Hernández-Castillo, D., Rodríguez-García, R. and Angulo-Sánchez, J.L. (2005) Antifungal Activity *in Vitro* of Aloe Vera Pulp and Liquid Fraction against Plant Pathogenic Fungi. *Industrial Crops and Products*, **21**, 81-87.
- [21] Spadaro, D. and Droby, S. (2016) Development of Biocontrol Products for Postharvest Diseases of Fruit: The Importance of Elucidating the Mechanisms of Action of Yeast Antagonists. *Trends in Food Science & Technology*, **47**, 39-49.
- [22] Rathi, R. and Tripathi, R.S. (1991) Host Range of *B. cinerea* Pers. ex. Fr. on Different Host Plants. *Indian Journal of Mycology and Plant Pathology*, **21**, 60-63.
- [23] HiMedia Laboratories (n.d.). HiPurAR Genomic DNA Miniprep Kit. <https://www.himedialabs.com/TD/GM540.pdf>
- [24] (n.d.). Eurofins Genomics. India Pvt Ltd. <https://www.eurofinsgenomics.com/en/>
- [25] Nene, Y.L. and Thapliyal, P.N. (1993) Fungicides in Plant Disease Control. Oxford & IBH Publishing Co. Pvt. Ltd.
- [26] R Core Team (2020) R: A Language and Environment for Statistical Computing. R Foundation for Statistical Computing. <https://www.R-project.org/>
- [27] Vincent, J.M. (1947) Distortion of Fungal Hyphae in the Presence of Certain Inhibitors. *Nature*, **159**, 850-850. <https://doi.org/10.1038/159850b0>
- [28] Skidmore, A.M. and Dickinson, C.H. (1976) Colony Interactions and Hyphal Interference between *Septoria nodorum* and Phylloplane Fungi. *Transactions of the British Mycological Society*, **66**, 57-64. [https://doi.org/10.1016/s0007-1536\(76\)80092-7](https://doi.org/10.1016/s0007-1536(76)80092-7)
- [29] van Kan, J.A.L., Shaw, M.W. and Grant-Downton, R.T. (2014) *Botrytis* Species: Relentless Necrotrophic Thugs or Endophytes Gone Rogue? *Molecular Plant Pathology*, **15**, 957-961. <https://doi.org/10.1111/mpp.12148>
- [30] Droby, S. and Lichter, A. (2007) Post-Harvest Botrytis Infection: Etiology, Development and Management. In: Elad, Y., Williamson, B., Tudzynski, P. and Delen, N., Eds., *Botrytis: Biology, Pathology and Control*, Springer, 349-367.
- [31] Williamson, B., Tudzynski, B., Tudzynski, P. and Van Kan, J.A.L. (2007) *Botrytis Cinerea*: The Cause of Grey Mould Disease. *Molecular Plant Pathology*, **8**, 561-580. <https://doi.org/10.1111/j.1364-3703.2007.00417.x>
- [32] Ghosh, A. (2018) Bioformulation of Antifungal Herbal Extract from *Curcuma caesia* Roxb. and *Ixora coccinea* L. against *Botrytis cinerea* Pers. *The Journal of Phyto-pharmacology*, **7**, 56-59. <https://doi.org/10.31254/phyto.2018.7112>
- [33] Wahmare, P., Chavan, A. and Shinde, R. (2011) *In Vitro* Evaluation of Fungicides, Bioagents and Plant Extracts against *B. cinerea* Pers. Inciting Gray Mold of Grapevine. *African Journal of Microbiology Research*, **5**, 2544-2550
- [34] Freeman, S., Minz, D., Kolesnik, I., Barbul, O., Zveibil, A., Maymon, M., et al. (2004) Trichoderma Biocontrol of *Colletotrichum acutatum* and *Botrytis cinerea* and Survival in Strawberry. *European Journal of Plant Pathology*, **110**, 361-370. <https://doi.org/10.1023/b:ejpp.0000021057.93305.d9>

- [35] Yue, H., Mo, Y., Zhao, M., Liu, W. and Li, H. (2023) Inhibition Effects of *P. fluorescens* on *B. cinerea* *in Vitro*. *Journal of Plant Protection*, **43**, 21-28.
- [36] Gholamnezhad, J. (2019) Effect of Plant Extracts on Activity of Some Defense Enzymes of Apple Fruit in Interaction with *Botrytis cinerea*. *Journal of Integrative Agriculture*, **18**, 115-123. [https://doi.org/10.1016/s2095-3119\(18\)62104-5](https://doi.org/10.1016/s2095-3119(18)62104-5)

Supplementary

Table S1. List of isolates collected.

Isolate code	Name of the place	Latitude	Longitude
SGM 1	Umktieh	25.68102°N	91.910726°E
SGM 4	Umeit	25.82105°N	91.96780°E
SGM 2	Sohliya	25.865471°N	91.654786°E
SGM 3	Umran	25.736332°N	91.879747°E
SGM 5	Umdiker	25.744149°N	91.821929°E

Table S2. Results of host range study conducted on five different hosts of grey mold pathogen, *B. cinerea*.

Sl. No.	Host plant	Botanical name	Plant part used	Reaction
1	Chrysanthemum	Chrysanthemum indicum L.	Flower	+
2	Faba bean	Vicia faba L.	Leaf and Pod	+
3	Gerbera	Gerbera jamesonii L.	Leaf and Flower	+
4	Lettuce	Lactuca sativa L.	Leaf	+
5	Marigold	Tagetes erecta L.	Flower	+

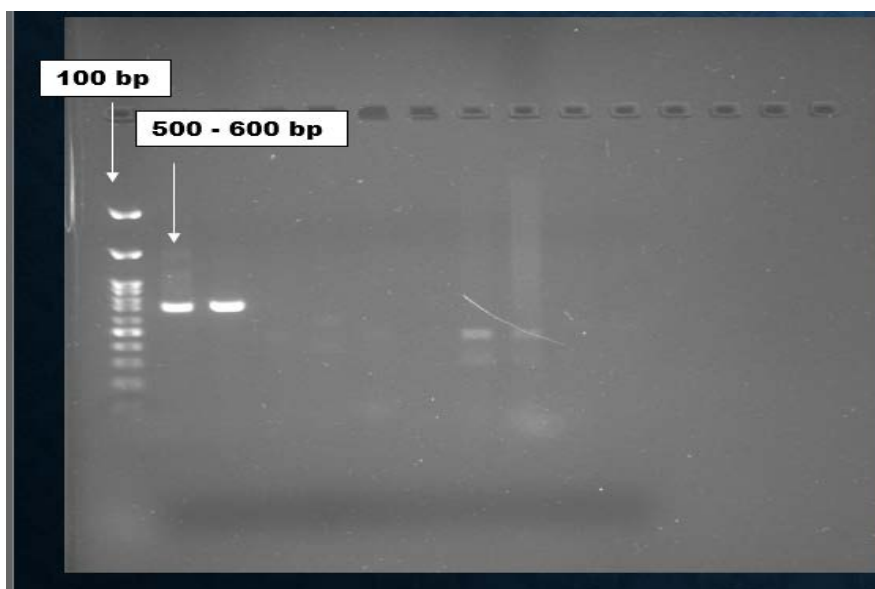


Figure S1. PCR amplification of the Isolate SGM 2 and SGM 4 with 100 bp ladder.