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RESEARCH ARTICLE

Variability analysis of *Phytophthora meadii* - A major causal agent of capsule rot (Azhukal) disease in small cardamom

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Abstract

Capsule rot disease, caused by *Phytophthora* spp., pose a significant challenge to the cultivation of cardamom and other spices and plantation crops, leading to considerable yield losses. In the present study, the impact of *P. meadii* on small cardamom was assessed in the primary cardamom-growing regions of Tamil Nadu and Kerala through a roving survey conducted during 2023–2024. The survey revealed varying levels of disease severity, ranging from 8.23% to 52.80%. The highest incidence was recorded in the Udumbanchola region (52.80%), while the lowest was observed in Thandikudi (8.32%). A total of eight isolates of *Phytophthora* (designated as PHY-1 to PHY-8) were collected from diseased samples and purified using the single hyphal tip method. Pathogenicity studies were conducted to evaluate the virulence of these isolates through two different methods: a detached capsule assay and an *in-planta* assay. Among the 8 pathogenic isolates, PHY-4 exhibited the highest level of virulence (75%) and presented typical symptoms of capsule rot disease. The isolates were analyzed for cultural and morphological variability. All eight isolates displayed distinct variations in growth patterns and sporangial morphology. Optimal growth and development of *P. meadii* were observed at temperatures ranging from 25–30°C. Scanning electron microscopy (SEM) analyses revealed specific morphological characteristics of the isolates, including hyaline, coenocytic mycelium and distinct sporangial structures.

Keywords

capsule rot disease; cardamom; *in-planta* assay; *Phytophthora meadii*; SEM; variability studies

Introduction

Cardamom (*Elettaria cardamomum* (L.) Maton), commonly known as small cardamom, green cardamom, or true cardamom, is cultivated in several countries, including India, Guatemala, Sri Lanka, Indonesia, Nepal, Tanzania and Mexico (1). Often referred to as the "Queen of Spices," it is renowned for its numerous health and culinary benefits. Belonging to the family *Zingiberaceae,* cardamom is a balanced tetraploid with chromosomal numbers of 2n = 48 and 2n = 52.

India's humid climate, loamy soil enriched with organic matter, substantial rainfall and unique cultivation and processing techniques contributes to the superior quality of Indian cardamom. These factors impart distinctive characteristics such as its unique aroma, size, flavour and vibrant colour. Globally, India is the largest producer of small cardamom, with an annual production of 41000 tonnes, followed by Indonesia and Guatemala. However, in terms of export, Guatemala ranks first, with India following. Within India, Kerala leads national production with 15.54 tonnes, followed by Sikkim, Nagaland, Arunachal Pradesh, Karnataka, West Bengal, Tamil Nadu and Uttarakhand (2). The primary cardamom-growing regions in India are Kerala, Karnataka and Tamil Nadu, covering 40345, 25135 and 4930 ha, respectively. The total production from these regions is approximately 24464 tonnes (3). Cardamom has various applications ranging from common dietary usage to significant pharmacological benefits. It is widely used in various sweets and confectionery (4).

Small cardamom is highly susceptible to various diseases caused by fungi, bacteria and viruses. Currently, over twenty -six diseases have been identified, which are categorized as either major or minor based on their severity and impact on yield loss (5). Major diseases include capsule rot (azhukal) caused by *Phytophthora* spp., rhizome/clump rot caused by *Pythium* and *Rhizoctonia* spp., as well as various forms of leaf blights, blotches and spots. Minor diseases include anthracnose, sooty mold and browning (5). Cardamom is also affected by several viral diseases, including Katte (Mosaic) disease caused by Cardamom mosaic virus, Nilgiri necrosis virus, Vein clearing virus (*Kokke kandu*) and Banana bract mosaic virus (1). Among the major diseases, capsule rot caused by *Phytophthora* spp. poses a significant threat to cardamom cultivation.

Capsule rot, caused by *Phytophthora* spp*.,* is considered one of the most fatal disease in cardamom fields, leading to severe yield losses and a reduction in crop quality (6). The disease manifest as water-soaked lesions on immature leaves and capsules, which progress to necrotic areas, resulting in rotting and shedding of leaves. Infected capsules turn dull brown, emit a foul odor and eventually fall off. In advanced stages, the infection spreads to the entire plant, which results in the complete destruction of the plant (7).

The taxonomy of *Phytophthora* has undergone significant evolutions over a period of time. The first species to be identified and renamed was *Phytophthora infestans* (8). To date, 12 clades have been recognized based on nuclear and mitochondrial characteristics. This framework provides a systematic approach to classifying and understanding the diversity within the genus. However, many natural ecosystems remain unexplored and new species are yet to be described (9).

The genus *Phytophthora* tends to infect a wide range of spice and plantation crops, posing significant challenges to agriculture, forestry and horticulture (10). For example, foot rot disease caused by *Phytophthora* spp. can devastate up to 95% of black pepper vines, leading to considerable yield losses (11). In cinnamon plantations, root rot can result in losses of up to 40% (12). Globally, black pod disease caused by *Phytophthora* spp. is responsible for an estimated 44% reduction in cocoa production (13). In arecanut plantations, fruit rot can cause up to 90% yield losses, while bud rot impacts around 15% of crops (14). Similarly, capsule rot disease of small cardamom, caused by *P. meadii*, becomes more severe under heavy rainfall, resulting in crop losses of up to 40% (15). The first reported case of capsule rot disease was from the Idukki district of Kerala (16), a region where cardamom is regarded as a promising crop.

Capsule rot disease is closely associated with the southwest monsoon, during which most parts of Kerala experience continuous heavy rainfall. Environmental factors such as high soil moisture, low temperatures, high relative humidity and prolonged rainfall create favourable conditions for *P. meadii* infection (17). The disease has been recorded to cause productivity losses of up to 30% (18).

This study aims to provide a deeper understanding of the unique nature and biology of *P. meadii* in cardamom fields, enabling better identification and development of mitigation strategies. These efforts are crucial for margining capsule rot disease, which poses a significant threat to the sustainable cultivation of small cardamom.

Materials and Methods

Survey and assessment of capsule rot disease in small cardamom

A roving survey was conducted between 2023 and 2024 in the major cardamom-growing regions of Kerala and Tamil Nadu to assess the impact of capsule rot disease in cardamom. The survey covered eight villages across these regions. In each village, four fields were selected and within each field, four plots with an average size of ten square meters were randomly marked for observation. The number of infected plants showing typical symptoms was recorded in each plot.

Disease severity was computed using the Percent Disease Index (PDI), where infected capsules were scored based on visual observations using a scale of 0–5 as follow: 0 = Free from infection, 1 = 1–10% of tillers bearing infected capsules, 2 = 11–20% of tillers bearing infected capsules, 3 = 26–50% of tillers bearing infected capsules, 4 = 51–75% of tillers bearing infected capsules, 5 = >75% of tillers bearing infected capsules due to capsule rot infection. Samples were collected from plants with infected capsules and stored in sealed bags for further studies.

Isolation and purification of the pathogen

Cardamom capsules exhibiting early symptoms of rotting were collected and thoroughly rinsed with sterile water. Infected tissues, along with small portions of adjacent healthy tissues, were cut into pieces for analyses. These pieces were rinsed three times with sterilized distilled water to eliminate external contaminants. Once dried, the moisture-free samples were aseptically transferred to Petri dishes containing Corn Meal Agar (CMA) and V8 agar medium. The medium was supplemented with propiconazole 25% EC (2 mL), vancomycin chloride (200

mg), rifampicin (10 mg) and PCNB (100 mg) to inhibit bacterial contamination and enhance fungal growth. The plates were incubated at a controlled temperature of 25 ± 2° C and a relative humidity of 85 ± 5% to promote mycelial growth and sporulation (19).

Pathogenicity study

Formation and release of zoospores

To induce zoospore release, mycelia containing sporangia were washed three times with sterile deionized water. The samples were then chilled at approximately 5°C for 15–20 min before being returned to a temperature of 24°C. Zoospores were typically released within an hour following the chilling process. The zoospore suspension was subsequently collected from the culture plates and applied to raised cardamom seedlings grown in pot culture to initiate infection.

Mass multiplication of the inoculum

Mass multiplication of *P. meadii* was conducted by transferring 6 mm fungal discs into 100 mL of sterilized V8 juice broth contained in 250 mL flasks. The flasks were incubated at 30°C under alternating 12 hr cycles of light (2000 Lux) and darkness for 2 weeks. These conditions were optimized to promote fungal growth and sporangial production (20).

Preparation of sporangial suspension

The sporangial suspension was prepared by gently scraping the surface of a 10-day-old culture grown on cornmeal agar using a sterilized blade. The scrapings were transferred into Petri dishes containing an adequate amount of 0.01 M KNO_3 to keep the culture moist without submerging it. The plates were incubated at 25 ± 1°C for 72 hr. Once the sporangia had developed, they were collected by flooding the culture plates with 10 mL of sterilized distilled water, followed by two washes. The resulting sporangial suspension was filtered through cheesecloth and its concentration was adjusted to 9 × 10^4 sporangia per milliliter using a haemocytometer for measurement (21).

Proving of pathogenicity

Pathogenicity studies were successfully conducted using both detached and *in-planta* assays. In these experiments, a sporangial suspension was applied to the Njallani Green Gold variety, demonstrating effective infection in both assay methods.

Detached assay

For proving pathogenicity through the detached assay, sporangial suspensions were prepared in a known volume of sterile distilled water. The suspension (10^5 sporangia/mL) was used for inoculation. Healthy, infection-free green capsules were selected, washed with sterile water and blot-dried for a few minutes. The capsules were then inoculated with 1 mL of zoospore suspension and transferred to humid chambers to facilitate symptom expression (22).

In-planta assay

The pathogenicity of all *P. meadii* isolates was tested on 2–3month-old cardamom seedling, using the Njallani Green Gold variety, which is susceptible to *Phytophthora* infection. Successful inoculation was achieved through the soil drenching method (23). The seedlings were inoculated by pouring 100 mL of inoculum around their bases and were frequently watered to facilitate infection. Disease progression and the virulence of the pathogenic isolates were monitored. The observed symptoms were compared to the original symptom, followed by re-isolation of the pathogen from the artificially infected plants. The re-isolated culture was compared to the original culture, thereby confirming Koch's postulates. The isolates that caused the highest disease intensity were identified as the most virulent.

Variability studies

Studying the cultural and morphological variability

Since *Phytophthora* survives in infected soils as hyphae and sporangia for a certain period (5), the study of cultural and morphological variability would help researchers accurately identify the pathogen responsible for capsule rot disease. By examining these aspects, researchers can gain a better understanding of how *P. meadii* adapts, survives and impacts cardamom plantations.

To study cultural variability, mycelial plugs (6 mm) from the advancing edges of 5-day-old cultures were placed at the centre of carrot agar (CA) plates. These plates were incubated at 28 ± 2°C and colony characteristics were studied after seven days (24). For morphological variability, 5 mm mycelial plugs were removed from the periphery of 72 hr old cultures, immersed in plates containing sterile water and exposed to white light for 24–48 hr. The plates were then examined for sporangial production and the morphological characteristics were documented (22). All isolates were phenotypically identified based on colony and sporangial features by comparing them with the lineage standard isolate (25).

Effect of temperature on the growth and development of *P. meadii*

The temperature studies will provide a detailed insight into the pathogen's behavior and adaptability under varying growth conditions. Understanding these dynamics will help in predicting the growth, survival and infection rates of *P. meadii,* aiding in the development of successful mitigation strategies. To study the effect of temperature, 3 mm diameter mycelial discs were cut from the actively growing region of a 72-hr old culture and inoculated into the Petri dishes containing 15 mL of medium. The inoculated plates were then incubated in BOD incubators at varying temperatures (5°, 10°, I5°, 20°, 24°, 28°, 32°, 36° and 40°C). The radial growth of the fungus was recorded at 24 hr intervals over a period of four days (26).

Scanning electron microscopic (SEM) analysis

Fourteen days old cultures of *P. meadii* were mounted onto an aluminium stub for the scanning electron microscope using double-sided adhesive tape and sputter-coated with gold particles. The gold particles were ionized using an ion coater prior to SEM imaging. Photographs were taken using the TESCAN VEGA3 SBH model at the laboratory of Archbishop Casimir Instrumentation Center (ACIC), St.

Joseph's College, Trichy. The morphological features of the mycelium and sporangia were investigated and examined (27).

Results

Survey and assessment of capsule rot disease in small cardamom

A roving survey was conducted during 2023–2024 to assess the severity of capsule rot disease in various regions of Kerala and Tamil Nadu. The survey was carried out in the following places: Mavady, Chemmanar, Vattapara, Udumbanchozha, Poopara, Bodimettu, Perumbarai and Thandikudi. The PDI was calculated for each location. The survey highlighted the occurrence of the disease at varying levels of severity, ranging from 8.32% up to 52.80%. The highest disease severity was observed in Udumbanchozha (52.80%), followed by Vattapara (41.46%), while the least severity was observed in the Thandikudi region (8.32%). A total of eight places were surveyed and the results are summarized in Table 1. An overview of the field affected by capsule rot disease shows typical symptoms (Fig. 1).

Isolation and purification of the pathogen

Capsules showing typical capsule rot symptoms was obtained from infected fields in Kerala and Tamil Nadu and used for successful pathogen isolation. A total of eight pathogenic *Phytophthora* isolates were obtained using CMA and V8 supplemented with suitable antibiotics. These isolates were designed as PHY-1, PHY-2, PHY-3, PHY-4, PHY-5, PHY-6, PHY-7 and PHY-8 for further studies.

Pathogenicity studies

Pathogenicity studies of all isolates were carried out using both detached assay and *in-planta* methods to prove Koch's postulates and assess the virulence of the pathogenic isolates. To demonstrate pathogenicity, the cardamom variety Njallani Green Gold was employed. Both assays

Table 1. Survey and assessment of the impact of capsule rot disease in cardamom

S. No	Isolate code	Location	District and state name	Geo coordinates		PDI*
1	PHY-1	Mavady	Idukki, Kerala	9.92893°	76.8447°	34.24^{cd} (35.79)
2	PHY-2	Chemmanar	Idukki, Kerala	9.90297°	77.17671°	31.34^{d} (34.04)
3	PHY-3	Vattapara	Idukki, Kerala	10.03279°	77.12631°	41.46^{b} (40.08)
4	PHY-4	Udumbanchozha	Idukki, Kerala	9.87858°	77.1932°	52.80^{a} (46.60)
5	PHY-5	Poopara	Idukki, Kerala	10.03279°	77.12632°	37.85^{bc} (37.95)
6	PHY-6	Bodimettu	Theni, Tamil Nadu	10.02383°	78.22352°	25.83^{e} (30.52)
7	PHY-7	Perumbarai	Dindugul, Tamil Nadu	10.36732°	77.98028°	15.37^{f} (23.01)
8	PHY-8	Thandikudi	Dindugul, Tamil Nadu	10.30969°	77.64305°	8.32^{g} (16.58)
CD (p=0.05)						3.377

*PDI - Per cent disease Index
Values within parentheses are arc sine transformed values.

Fig. 1. An overview of cardamom field incited with capsule rot disease. a) an overview of the cardamom field b) cardamom plants infected with capsule rot disease c) typical capsule rot symptom

concluded that PHY-4 exhibited the higher level of virulence (>75%), showing typical symptoms and was therefore selected as the most virulent isolate. In the detached assay, the capsules initially showed water-soaked lesions, which were followed by complete rotting. In the *in-planta* assay, conducted on 2–3month-old seedlings, disease progression was observed as water-soaked lesions followed by necrosis, which later led to rotting and destruction of the plant. Fig. 2 & 3 illustrates the results of the detached and *in-planta* assays, respectively.

Fig. 2. Pathogenicity study using detached assay. a) Healthy capsules free from infection b) Capsules exhibiting rotting symptoms on artificial inoculation

Fig. 3. Pathogenicity study using *in-planta* assay. a) Healthy cardamom seedling b) Artificially inoculated seedling showing death of the plant

Variability studies

Studies on cultural variability

The eight pathogenic isolates collected from various locations were subjected to a study of cultural variability. To assess the cultural differences among the isolates, characteristics such as growth pattern, growth rate, colony color and sporulation were recorded. PHY-1 and PHY-8 exhibited a floral growth pattern with whitish to dull white, dense mycelium. PHY-2 displayed a petaloid growth pattern with dense, pure white mycelium. A plain growth pattern with irregular concentric rings was observed in PHY-3 and PHY-6, producing thin, whitish mycelium with sharp margins. PHY-4 and PHY-5 showed no definite growth pattern but had abundant, pure white mycelium. PHY-7 exhibited a slightly striated growth pattern with less aerial mycelium, which was dull white in color. Sporulation studies revealed that PHY-2, PHY-4, PHY-5 and PHY-7 exhibited faster sporulation, followed by moderate sporulation in PHY-1 and PHY-8, while less sporulation was observed in PHY-3 and PHY-6. The cultural variability of *P. meadii* is detailed in Table 2 and depicted in Fig. 4.

Studies on morphological variability

A total of eight isolates of *P. meadii* were studied in detail for morphological characterization. Variability within species was noted in the size and shape of the morphological structures. PHY-1 and PHY-8 produced globose sporangia with an L/B ratio of 1.28–1.33, while PHY-2 and PHY-4 produced ovoid sporangia with an L/B ratio of 1.23–1.40. PHY-3 produced ovoid-obpyriform sporangia with an L/B ratio of 1.86. Furthermore, PHY-5 and PHY-6 produced obpyriform sporangia with L/B ratios varying from 2.47–2.64. PHY-7 produced ellipsoidal sporangia with an L/B ratio of 1.41. All isolates were found to be highly caducous, with varying pedicel lengths. The morphological variability of *P. meadii* is represented in Table 3 and depicted in Fig. 5.

Impact of temperature on the growth and development of *P. meadii*

All isolates of *P. meadii* were studied for their growth and development at varying temperatures from 10°C–40°C. The results, based on mycelial growth, revealed that maximum growth occurred at temperatures between 25–30°C, followed by moderate growth at temperatures between 15–20°C. Slow growth was observed at temperatures below 15° C and above 30°C. All isolates of *P. meadii* failed to grow at temperature exceeding 35°C. The effect of varying temperatures on the growth of *P. meadii* is shown in Table 4 and graphically represented in Fig. 6.

Scanning electron microscopy (SEM) analysis

The investigations conducted using SEM revealed distinct

Table 2. Growth characteristics of *P. meadii* in relation to its cultural variability

S. No	Isolates	Growth pattern of mycelium	Colony colour	Growth rate	Sporulation
1	PHY-1	Floral	White	Moderate	Moderate
2	PHY-2	Petaloid	Pure white	Slow	Fast
3	PHY-3	Plain with irregular concentric rings	Pure white	Slow	Slow
4	PHY-4	No definite pattern	Pure white	Fast	Fast
5	PHY-5	No definite pattern	white	Fast	Fast
6	PHY-6	Plain with irregular concentric rings	Pure white	Slow	Slow
7	PHY-7	Slightly striated	Dull white	Moderate	Fast
8	PHY-8	Floral	Dull white	Moderate	Moderate

Table 3. Morphological variability of *P. meadii*

S. No	Isolates	Sporangial shape	Sporangial length (µm)	Sporangial breadth (µm)	L/B ratio	Nature of caudicity
1	PHY-1	Globose	42.74±0.41	33.21±0.12	1.28	Highly caducous
2	PHY-2	Ovoid	37.90±0.85	30.78±0.50	1.23	Highly caducous
3	PHY-3	Ovoid obpyriform	53.52±1.14	28.66±0.49	1.86	Highly caducous
4	PHY-4	Ovoid	56.51±1.13	40.12±0.08	1.40	Highly caducous
5	PHY-5	Obpyriform	49.70±0.98	18.76±0.40	2.64	Highly caducous
6	PHY-6	Obpyriform	48.92±0.43	19.74±0.29	2.47	Highly caducous
7	PHY-7	Ellipsoid	42.12±0.48	29.72±0.71	1.41	Highly caducous
8	PHY-8	Globose	41.71±0.89	31.29±0.20	1.33	Highly caducous

Table 4. Effect of temperature on the growth and development of *P. meadii*

S. No	Growth rates (mm)/day							
	10°C	15°C	20°C	24°C	28°C	32°C	36°C	40°C
PHY-1	1.72	5.64	6.55	7.53	7.76	1.77	0.73	0.0
PHY-2	1.75	4.55	6.02	7.12	7.24	1.72	0.51	0.0
PHY-3	2.09	4.86	6.97	7.80	7.95	1.68	0.55	0.0
PHY-4	2.17	5.69	7.95	8.67	8.88	2.11	0.76	0.0
PHY-5	1.79	5.60	6.81	7.00	7.08	1.81	0.61	0.0
PHY-6	2.11	4.97	7.83	8.11	8.26	2.90	0.63	0.0
PHY-7	1.68	4.78	6.71	7.36	7.49	1.86	0.69	0.0
PHY-8	1.56	4.66	6.87	7.39	7.52	1.79	0.71	0.0

morphological characteristics of *P. meadii,* such as

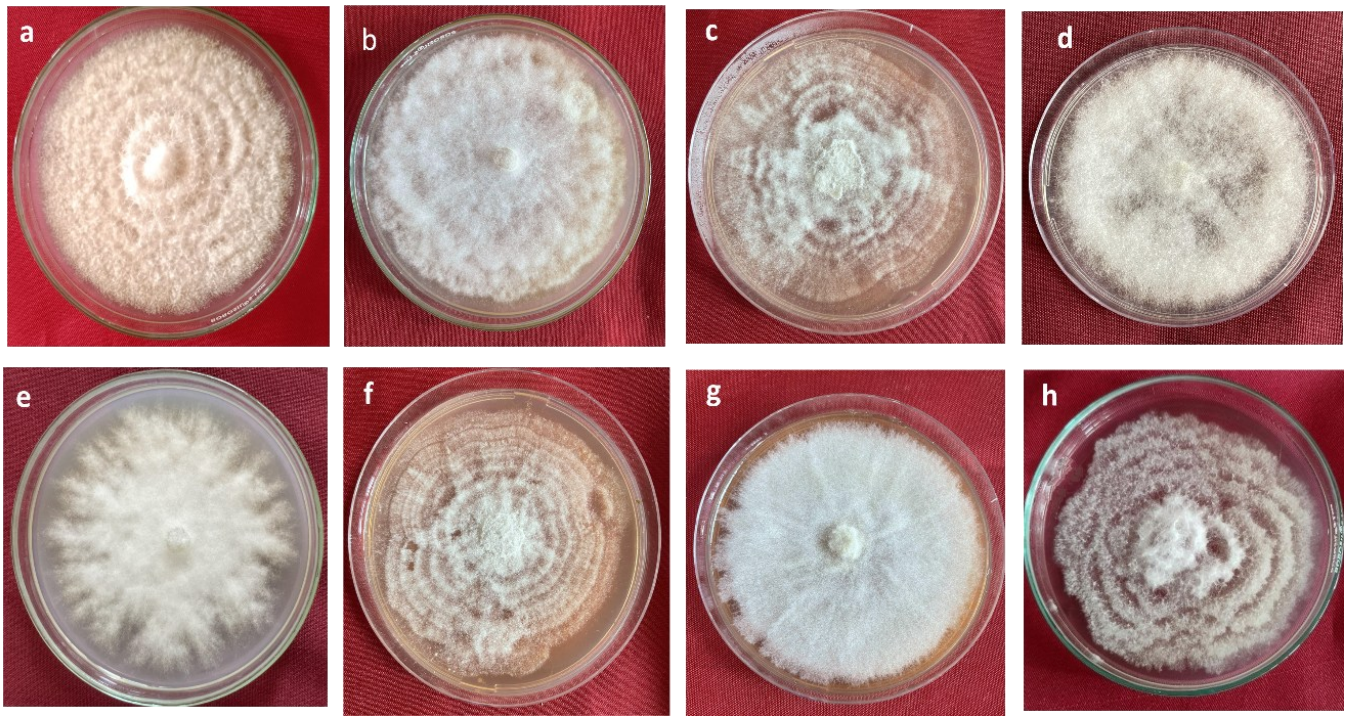

Fig. 4. Pathogenicity study using *in-planta* assay. a) Healthy cardamom seedling b) Artificially inoculated seedling showing death of the plant

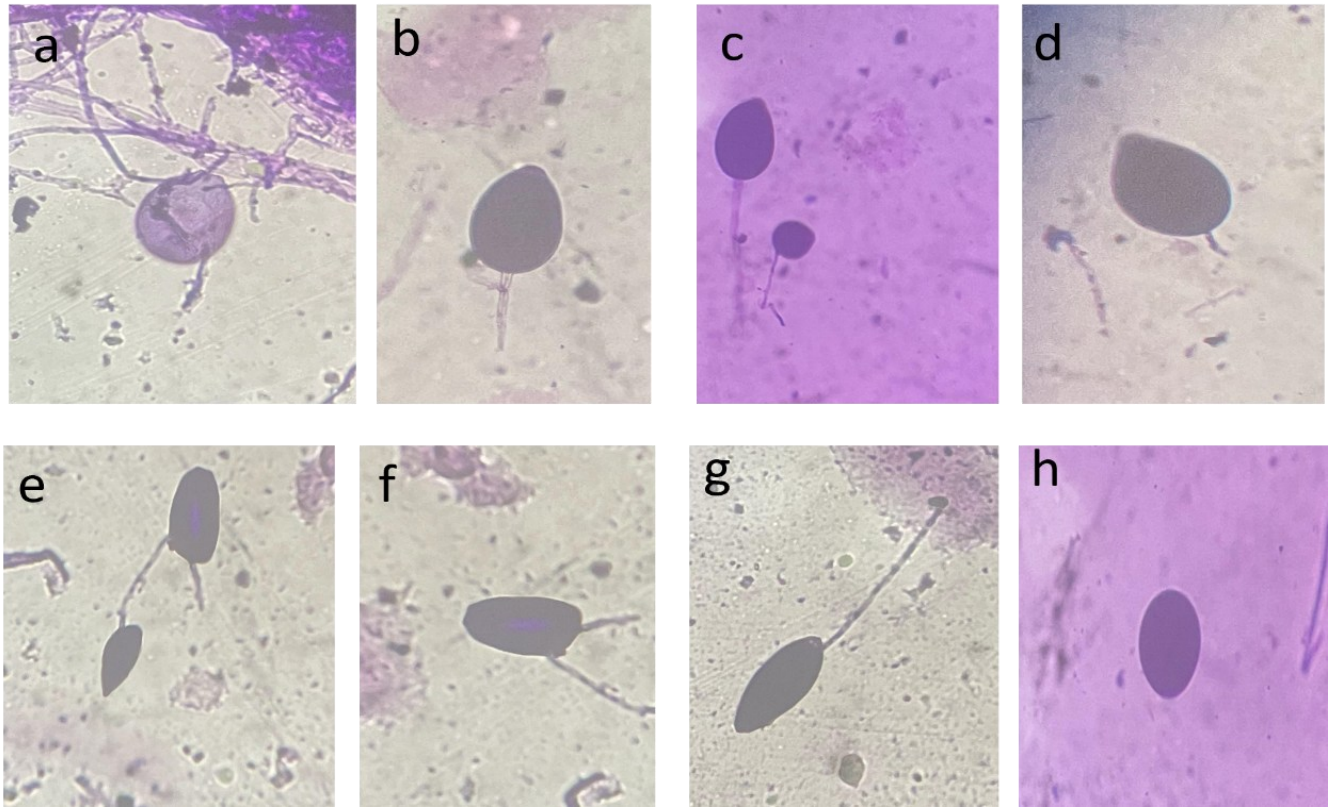

Fig. 5. Morphological variations in the sporangial structures of *P. meadii*. a) Globose b) Ovoid c) Ovoid-obpyriform d) Ovoid e) Obpyriform f) Obpyriform g) Ellipsoid h) Globose

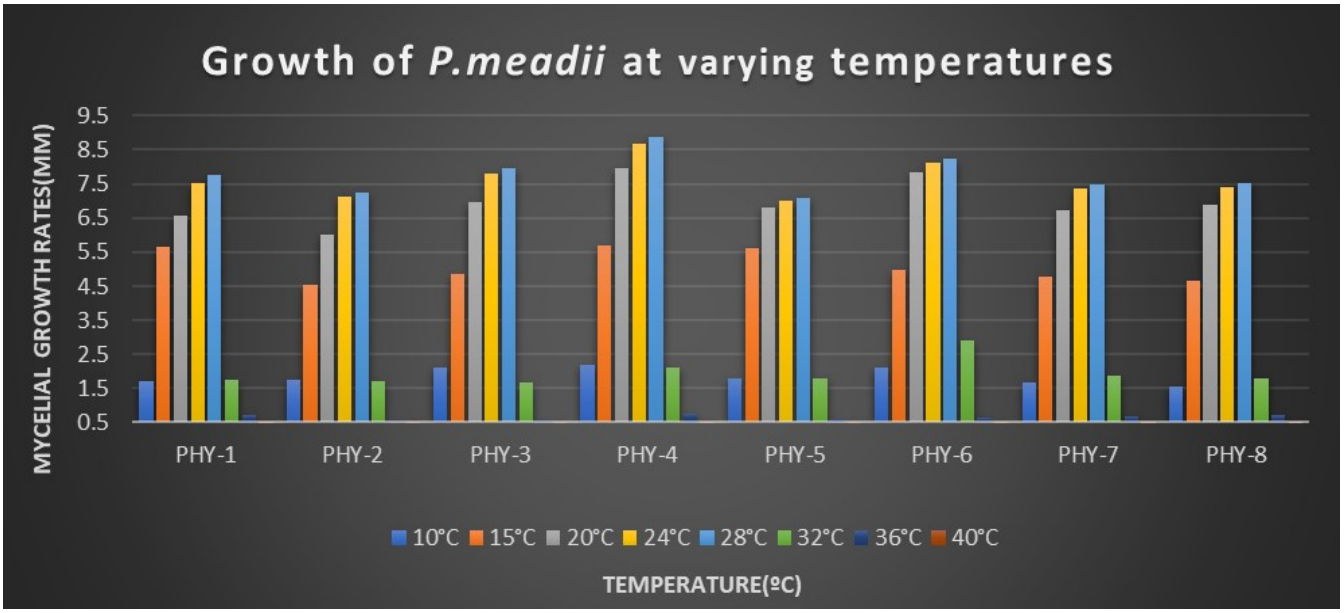

Fig. 6. Graphical representation on the mycelial growth (mm) of *P. meadii* at varying temperatures ranging from 10°C–40°C

coenocytic mycelium and variations in sporangial structures, ranging from ovoid to globose sporangia, as shown in Fig. 7.

Discussion

Phytophthora, a menacing pathogen infecting various crops worldwide, remains a persistent threat to sustainable cardamom cultivation. The present research aimed to investigate the variability of *Phytophthora* spp., responsible for capsule rot disease, in the predominant cardamom growing areas. Cardamom cultivation is primarily concentrated in the Idukki district of Kerala, where the first report of capsule rot disease was recorded (16). The southwest monsoon plays a significant role in the initiation and progression of capsule rot disease due to heavy, continuous rainfall that enhances high zoospore production. The survey assessed disease severity and crop loss in Kerala and Tamil Nadu, revealing crop loss ranging from 10% to 50%. Similarly, a yield loss of 50% was recorded during the heavy rainfall season (6). These findings align with previous reports of crop losses reaching up to

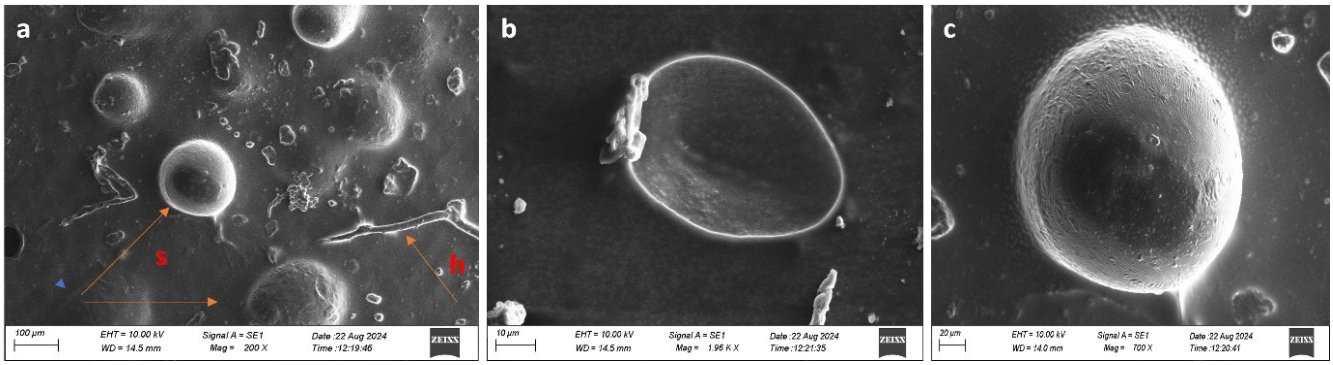

Fig. 7. Scanning electron microscopy (SEM) images depicting the morphological features of *P. meadii*. a) Varying sporangia with coenocytic hyphae b) Highly caducous and ovoid sporangia c) Globose sporangia with short pedicel

40% during extended periods of heavy rainfall (15) and a productivity loss of up to 30% has also been documented (18).

The oomycete pathogen *Phytophthora* can be isolated through various methods. In this study, the tissue segment method of isolation was employed to isolate *Phytophthora* spp. from infected cardamom capsules, a method similarly used by others for isolating pathogens from diseased cardamom, cocoa and pepper (28). The successful isolation of *P. meadii* was achieved from scrapings and pinhead-like pustules on rot-affected vanilla beans (29). Similarly, in nutmeg crops, this method was utilized for isolation (30). For the current study, CMA and V8 media with the respective antibiotics were used, as similar media with suitable antibiotics have been successful in isolating *P. cinnamomi* from avocado crops (31).

Zoospores are regarded as crucial for the initiation and spread of infection in *Phytophthora* spp., playing a major role in establishing pathogenicity. Several researchers have successfully used zoospore suspensions to prove the pathogenicity of *Phytophthora* spp. on various hosts (32–34). In this study, both detached and *in-planta* assays were used to demonstrate the pathogenicity of virulent *P. meadii* isolates. Similar methods have been employed to test pathogenicity in vanilla plantations (35) and bell pepper (33).

The soil drenching method with zoospore suspensions was employed for successful inoculation in cardamom plantations. This methodology used for inoculation aligns with the work put forth by others (36). A similar study on the pathogenicity of *Phytophthora* spp. causing leaf and fruit fall in nutmeg plantations successfully demonstrated the pathogen's ability to infect various plant parts and re-isolation confirmed Koch's postulates (22).

Cultural and morphological variability studies were conducted to assess variability within the species isolated from distinct locations. Various isolates of *P. meadii* collected from different regions of Kerala and Tamil Nadu were studied for their cultural and morphological variability. This study aligns with the previous research, which identified the oomycetous fungus *P. meadii* through phylogenetic and haplotype analysis as a causative agent of fruit rot in areca nut in India (24). The present study found remarkable differences in cultural characteristics, such as growth pattern, colony colour, growth rate and sporulation of *P. meadii,* followed by morphological variations in sporangial shape, size, papillation and caudicity. Based on these variations, the isolates were grouped into six distinct morphotypes based on colony characteristics (24).

Temperature is recognized as a key determinant influencing the growth and development of *Phytophthora* spp. Optimal temperature conditions can accelerate spore germination and increase their ability to infect host plants. Conversely, temperatures outside their preferred range can inhibit growth and reduce pathogenic potential. Fluctuations in temperature may also affect the life cycle stages, such as sporangia formation and zoospore release, further impacting disease spread and severity. Studies conducted to assess the role of temperature on the growth and development of *P. meadii* revealed significant results, with maximum growth observed between 25–30°C, which is considered ideal for most *Phytophthora* spp. (37, 38). This was followed by minimal growth at temperatures between 20–35°C. No growth was observed at temperatures beyond 35°C. These results align with the findings, where maximum growth was observed at 28°C and no growth occurred at 40°C (26). Similarly, *P. capsici* exhibited evident growth and sporulation at 28°C (39). *Phytophthora* isolates from nutmeg grew well at 25°C but failed to grow at temperatures of 10°C and 37°C (22). *P. meadii* isolates from rubber crops grew rapidly at 25°C, with slower growth at 15–20°C in darker conditions (40). *P. cinnamomi* exhibited optimal growth between 24–26°C, with minimal growth at 10°C in cinnamon crops (12).

The escalating variability among oomycetes is a subject of great interest, as it contributes to their evolving resistance over time (41). Factors such as nutrients, moisture, humidity, sporulation and temperature influence the cultural and morphological variability of these species. Elevated soil moisture promotes sporangia production and zoospore release, while extreme temperatures induce the formation of resting spores, leading to dormancy. Temperature and climate are key factors in the progression of capsule rot disease, as they tend to aggravate its severity. The present research provides key findings with practical implications, such as an assessment of the impact of *P. meadii* on small cardamom cultivation in Kerala and Tamil Nadu. The survey highlighted significant variability in disease severity across regions, with Udumbanchola experiencing the highest incidence. Pathogenicity studies

identified PHY-4 as the most virulent isolate, demonstrating typical symptoms of capsule rot disease. Since the pathogen survives as spores and resting structures that serve as the primary inoculum, it contributes to the exacerbation of capsule rot disease. Studies on the variability of *P. meadii* will aid in understanding the pathogen's adaptability and its various strains, enabling the development of future management strategies and the precise application of control measures to improve crop yield sustainably. This study partially highlights research gaps in assessing the impact of capsule rot disease and provides an in-depth examination of the characteristics of *P. meadii*.

Conclusion

Cardamom, often referred to as the "emerald jewel of spices," is highly susceptible to the devastating damage caused by the *Phytophthora* genus. This study investigates the virulent nature of the plant-damaging pathogen *P. meadii* in cardamom fields. Cardamom cultivation is frequently threatened by capsule rot disease, which results in significant yield losses. The present study aims to explore variations in disease severity and prevalence, along with a detailed investigation of *P. meadii* by examining its cultural and morphological variability across different geographical locations. Since capsule rot is favored by persistent inoculum, continuous rainfall and high soil moisture, effective sanitary practices could help reduce the initiation and spread of the disease. A comprehensive understanding of the nature and biology of *Phytophthora* species responsible for capsule rot in small cardamom provides valuable insights for accurate identification and effective disease management. Future research should focus on exploring the genetic variability of *P. meadii* to better understand its adaptations and resistance mechanisms using advanced molecular tools, thereby promoting environmental resilience. Cardamom, often referred to as the "emerald jewel of spices," is highly susceptible to the devastating damage caused by the *Phytophthora* genus. This study investigates the virulent nature of the plant-damaging pathogen *P. meadii* in cardamom fields. Cardamom cultivation is frequently threatened by capsule rot disease, which results in significant yield losses. The present study aims to explore variations in disease severity and prevalence, along with a detailed investigation of *P. meadii* by examining its cultural and morphological variability across different geographical locations. Since capsule rot is favored by persistent inoculum, continuous rainfall and high soil moisture, effective sanitary practices could help reduce the initiation and spread of the disease. A comprehensive understanding of the nature and biology of *Phytophthora* species responsible for capsule rot in small cardamom provides valuable insights for accurate identification and effective disease management. Future research should focus on exploring the genetic variability of *P. meadii* to better understand its adaptations and resistance mechanisms using advanced molecular tools, thereby promoting environmental resilience.

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Authors' Contributions

JCW and KK conceived the idea, analysed, drafted, and formatted the manuscript. JCW, KK, YI and EAAK participated in the sequence alignment and edited the manuscript. MK, RJ, SKG and MML assessed the data provided. RC, AM, JCW and KK provided the methodology for writing the manuscript. JCW and KK carried out the reference management of the manuscript. All authors read and approved the final manuscript.

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While preparing the manuscript, the authors used Grammarly to improve the language and readability. After using this tool/service, the authors reviewed and edited the content as needed and took full responsibility for the publication's content.

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