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Pathogenic Variability of *Lasiodiplodia theobromae* (Pat.) Griffon and Maubl. Causing Mango Dieback and Screening of Mango Germplasm for Disease Resistance

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ABSTRACT

Background: Employing resistant germplasm is one of the most practical and cost efficient approach for disease management. Neverthless, use of these resistant cultivars turns to be fruitless due to persistence of diversity among pathogen populations across different agroecologies. These variations facilitate the pathogen to adapt and thrive in diverse environmental conditions. The variants within a population can alter the pace of disease progression and potentially trigger disease outbreaks in other host lines, which could undermine the cultivar stability. Furthermore, pathogenic variability studies indirectly aid in discriminating between resistant and susceptible accessions.

Methods: Five distinct isolates of *Lasiodiplodia theobromae*, associated with mango dieback and derived from five different mango varieties and five different districts in Kerala, were inoculated on six differential mango cultivars. Pathogenic attributes including PDI, PDS, incubation days, lesion size and virulence index were assessed. The coefficient of infection was determined to discern the landraces for their level of resistance/susceptibility towards mango dieback.

Result: Different virulence patterns were discerned for the isolates. The range for various pathogenic parameters, including PDI, PDS, incubation days, lesion size and virulence index across different isolates, was recorded as follows: 33.33-100 per cent. 13.33-73.33 per cent, 9-17.33 days, 0.28-0.95 cm and 2.78-4.19 respectively. The parameter, virulence index indicated that the isolates KoKBn, KPB, and WMnMa were more virulent. The coefficient of infection calculated based on PDI and PDS identified the varieties Neelam, Bennet Alphonso, and Banganapally as susceptible and the remaining varieties Priyur, Muvandan, and Imam Pasand as moderately susceptible.

Key words: Diversity, Mango cultivar, Mango variety, Response, Variability, Variation, Varietal resistance, Varietal screening.

INTRODUCTION

The disease mango dieback is incited by a necrotrophic and wound pathogen *Lasiodiplodia theobromae*, (Pat.) Griffon and Maubl. The fungus is plurivorous, ubiquitous, polyphagous, ecologically diverse, opportunistic pathogen (Punithalingam, 1980) which is categorized within Ascomycota phylum and is a member of the family Botryosphaeriaceae. This family encompasses species that may thrive as saprotrophs or seed endophytes (Crous *et al*., 2006).

The term dieback means that the plant dies from top downwards (Prakash and Misra, 2001). According to Khanzada *et al*. (2004), the leaves acquire brownish tinge and get rolled at margins. Later, leaf falls and it look like a dead branch. Other branches of the tree also get affected giving a fire scorch appearance. Sometimes perishment of trees also happens. During initial stages of gummosis, it was seen as a droplet; under severe stages larger portion of the tree got affected. A range of symptoms has been documented for the disease by various authors which include discolouration of vascular systems, rolling up of affected leaves, leaf fall, oozing of gums, cracking of bark *etc*. (Saeed *et al*., 2017; Kwon *et al*., 2017; Honger *et al*., 2018).

The pathogen attacks stressed plants, causing up to 40% loss in Pakistan (Mohali *et al*., 2005; Alam *et al*., 2017). It has been noted that the disease is prevalent worldwide. In India, disease has been reported in Uttar Pradesh, Punjab, Hyderabad, Andhra Pradesh, Tamil Nadu. In Punjab, incidence was 100 per cent (Fateh *et al*., 2022). Owing to its broad host spectrum and distribution, this complex pathogen manifests variability in morphology (Dheivam *et al*., 2020; Renganathan *et al*., 2020; Logeshwari *et al*., 2022), pathogenicity (Rodríguez-Gálvez *et al*., 2017; Ullah *et al*., 2017) and genotype (Xie *et al*., 2016 and Sathya *et al*., 2017).

Cultural, biological and chemical interventions have been used to mitigate dieback in mango agroecosystems.

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However, host plant resistance remains as the most potent and viable option in combating the disease. However, the durable use of novel mango cultivars resistant to dieback necessitates consideration of the pathogenic variability of the causal organism. The availability of pathogenically diverse isolates aggressive on different resistant cultivars is instrumental in locating the resistant loci in the host. Integration of genes from multiple resistance sources is vital to provide comprehensive resistance to an array of pathotypes prevalent in an area.

The characterization of diverse populations of *Lasiodiplodia theobromae* could improve the knowledge about genetic structure of the pathogen population, epidemiological studies, gene flow, population biology and its interaction with mango in order to produce mango cultivars with stable and durable resistance to dieback. Indirectly, pathogenic variability study would aid in identifying resistant and susceptible accessions offering the most sustainable disease control strategy.

Morphological, cultural and genetic variability of *Lasiodi-plodia theobromae* have been extensively studied worldwide (Xie *et al*., 2016; Sathya *et al*., 2017; Dheivam *et al*., 2020; Renganathan *et al*., 2020; Logeshwari *et al*., 2022). No studies have yet addressed the pathogenic variability of the dieback pathogen *Lasiodiplodia theobromae*. Furthermore, research on screening mango germplasm for resistance to mango dieback remains scarce. Therefore, the present study was undertaken with a goal to characterize *Lasiodiplodia theobromae* isolates collected from the prominent mango growing areas in Kerala for virulence diversity and choose geographically and pathologically diverse isolates for screening of mango accessions.

MATERIALS AND METHODS

The experiment was conducted at the Department of Plant Pathology, College of Agriculture, Vellanikkara, Kerala Agricultural University, Thrissur, Kerala during the year 2024. A total of 37 isolates of *Lasiodiplodia theobromae* were obtained from 17 mango varieties and 23 locations spread across five districts of Kerala. From these 37 isolates, 10 most virulent isolates were selected based on incubation days and size of lesion. Details of the isolates have been represented in Table 1.

These 10 isolates were subjected to cluster analysis based on morphological and cultural characters using PAST 4.03 (Paleontological Statistics Software Package for Education and Data Analysis) (Hammer and Harper, 2001) software and five isolates of *Lasiodiplodia theobromae* were chosen based on location, variety, clusters formed and cultural characters of the isolates.

Later, these five representative isolates were inoculated on six popular mango varieties *viz.,* two traditional varieties including Muvandan and Priyur and four commercial varieties Neelam, Bennet Alphonso, Banganapally and Imam Pasand. Mycelial Bit Innoculation Method (MBIM) was used for inoculating these five isolates (Rocha *et al.,* 1998). Healthy mango seedlings were used for this purpose. Selected healthy twigs were sterilized with alcohol and later washed with sterile water.

A 5 mm diameter mycelial PDA plug was kept on a 5 mm sized cut made on the bark of the twig with the mycelium facing the cambium (Ismail *et al*., 2012). Each plant was covered with a polythene cover for the maintenance of humidity. Plants were monitored regularly for the development of symptoms. For each isolate, three replications and a control were kept. The experiment was done in randomised block design (RBD). Different observations like the size of the lesion, the days taken for symptom development, PDI (Per cent Disease Incidence) and PDS (Per cent Disease Severity) were noted down.

PDS was calculated using 0-5 scale given by Gupta and Prasad (1984) as represented in Table 2.

Virulence index (VI) was calculated for different isolates according to the formula given by Ahmad (2016).

$$\text{Virulence index} = \frac{\text{Per cent disease intensity}}{\text{Latent period}}$$

Per cent disease intensity and latent period is identical to per cent disease severity and incubation days respectively.

In order to categorize the varieties for their level of resistance/susceptibility towards mango dieback, coefficient of infection (CI) was calculated based on the formula given by Datar and Mayee (1981).

$$\text{Coefficient of infection} = \frac{\text{PDI} \times \text{PDS}}{100}$$

Where
PDI = Per cent disease incidence.
PDS = Per cent disease severity.

Different mango varieties were categorized based on CI value according to Sangeetha (2008) as given in Table 3.

RESULTS AND DISCUSSION

Cluster analysis

Cluster analysis of the 10 most virulent isolates of *Lasiodiplodia theobromae* yielded two major clusters, I and II. The major cluster I had two subclusters, Ia and Ib while major cluster II had only one subcluster. Two isolates each from subcluster Ia and Ib and one isolate from major cluster II were selected. Cluster analysis has been depicted in Fig 1.

The subcluster Ia was divided into two groups; From the first group containing the isolates KoKBn and PVaT, KoKBn was selected being the only one isolate from Kottayam. PMuNe was selected from the second group being the only variant present in that group. The subcluster Ib had two groups; First group of I b was divided to two subgroups; PMuM from first subgroup was avoided as one isolate from Palakkad has been selected earlier. From second subgroup containing KPKa and KPB, KPB was

selected based on earliness in pycnidia production which could be related to higher virulence. From the second group containing TMNe and WMnMa, WMnMa was selected due to variety specificity. From cluster II containing TNaM and WAmMa, TNaM was selected due to location specificity. Therefore, selected isolates were TNaM, WMnMa, KPB, PMuNe and KoKBn. Hence, these representative isolates were collected from five different districts and different varieties.

Evaluation of pathogenic variability of different isolates of the pathogen

All the five isolates caused 100 per cent incidence on Banaganapally variety. PDI ranged between 66.67-100. Among the isolates, isolate TNaM caused highest PDS of 73.3% on Neelam variety whereas PMuNe showed lowest PDS of 13.33% on Imam Pasand. Largest lesion (0.95 cm) was caused by KPB on Muvandan while smallest lesion (0.28 cm) was caused by PMuNe on Imam Pasand. KPB

Table 1: Details of 10 most virulent isolates of *Lasiodiplodia theobromae*.

District	Location	Variety	Isolate code	Latitude/Longitude
Thrissur (T)	Muringoor (M)	Neelam (Ne)	TMNe	10.28425/76.330734
	Nandipulam (Na)	Muvandan (M)	TNaM	10.39165/76.319452
Palakkad (P)	Muthalamada(Mu)	Muvandan (M)	PMuM	10.564587/76.76548
		Neelam (Ne)	PMuNe	
	Vadakkanchery (Va)	Totapuri (T)	PVaT	10.3530/76.2734
Wayanad (W)	Ambalavayal(Am)	Mallika (Ma)	WAmMa	11.617714/76.21427
	Mankombu (Mn)	Mallika (Ma)	WMnMa	11.616119/76.222066
Kasargod (K)	Padanakkad (P)	Kalapadi (Ka)	KPKa	12.255804/75.117235
		Banganapally (B)	KPB	
Kottayam (Ko)	Kumarakom(K)	Bennet alphonso (Bn)	KoKBn	9.624327/76.43567

Table 2: Disease severity score chart for pathotypic variability studies.

Score	Category	Description of symptom
1	Resistant (R)	Few pin head sized spots
2	Moderately resistant (MR)	Not more than one cm area affected from cut end
3	Moderately susceptible (MS)	1 to 2 cm area infected from cut end
4	Susceptible (S)	More than 2 cm area infected from cut end
5	Highly susceptible (HS)	Cut end completely affected and died, infection passes to main stem

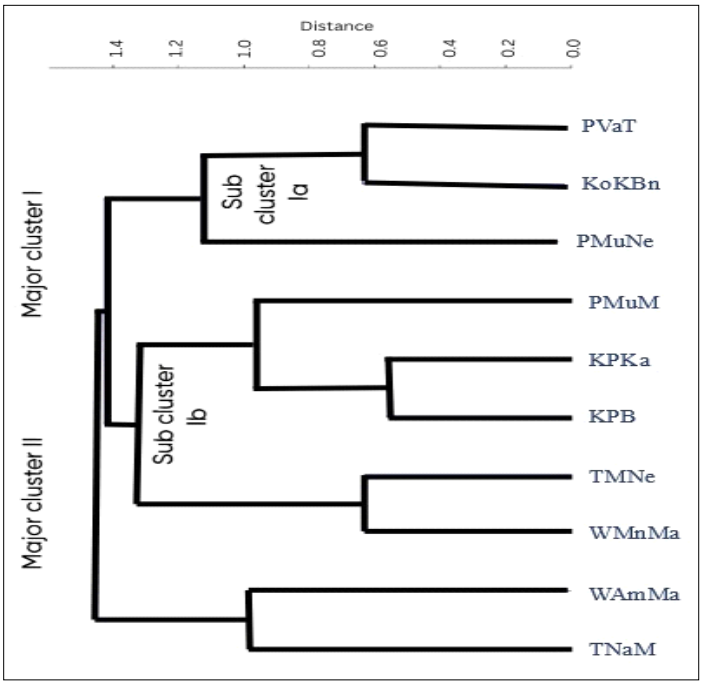


Fig 1: Cluster analysis of ten isolates of *Lasiodiplodia theobromae*.

took minimum number of incubation days (9) on Muvandan while WMnMa and PMuNe took maximum number of incubation days (17.33) in Neelam and Imam Pasand respectively. Different symptoms caused by the isolates include black discolouration on inoculated area and cracking of outer cortex. Some isolates were able to cause drying of mango seedlings; *i.e.* TNaM, WMnMa, KPB and KoKBn caused drying of twigs in Neelam and KoKBn caused drying of twigs in Priyur. On Neelam variety, KPB caused drying of twigs 29 DAI (days after inoculation); WMnMa 34 DAI; KoKBn 37 DAI and TNaM caused drying 44 DAI. On Priyur variety, KoKBn caused drying of twigs 41 DAI. At the outset, lesions formed on the plants and progressively spread across the entire stem; subsequently leaves started showing wilting and yellowing symptoms. A week later, the leaves turned brown in colour and by the following week, leaves started falling. Experimental layout has been represented in Fig 2 and pathogenic variability evaluation of *L. theobromae* isolates on different mango varieties has been represented in Fig 3i and 3ii.

Variations were discerned significantly different among the isolates for different pathogenic parameters. PDI, PDS, size of lesion, days taken for symptom development (incubation days/latent period), virulence index for different isolates, CI value for different varieties and categorization of mango varieties based on CI value have been furnished in Table 4, 5, 6, 7, 8, 9 and 10 respectively.

The isolate TNaM produced substantial variation for different pathogenic parameters like PDI, PDS, lesion size and incubation days on different mango varieties. Range of PDI was 66.67-100 per cent with maximum and minimum disease incidence of 100 per cent on Bennet Alphonso, Neelam and Banganapally and 66.67 per cent on Priyur, Muvandan and Imam Pasand respectively. PDS ranged between 26.64-73.33 per cent with highest of 73.33 per cent on Neelam and lowest of 26.64 per cent on Priyur and Imam Pasand. Lesion length ranged between 0.4-0.76 cm with largest and smallest lesion on Banganapally (0.76 cm) and Priyur (0.4 cm) respectively. Incubation days ranged between 11.67-16 with minimum of 11.67 days on Bennet Alphonso and maximum of 16 days on Priyur and Imam Pasand.

Table 3: Categorization of mango varieties based on CI value.

CI value	Category
0-4	Highly resistant (HR)
4.1-9	Resistant (R)
9.1-19	Moderately resistant (MR)
19.1-39	Moderately susceptible (MS)
39.1-69	Susceptible (S)
69.1-100	Highly susceptible (HS)



Fig 2: Experimental layout.

Appreciable variation was discerned for different pathogenic traits like PDI, PDS, incubation days and lesion size by the isolate PMuNe. Range of PDI was 33.33-100 per cent with highest and lowest incidence of 100 per cent on Bennet Alphonso, Imam Pasand, Banganapally and Priyur and 33.33 per cent on Muvandan respectively. PDS ranged between 13.33-53.28 per cent with maximum on Bennet Alphonso and Banganapally (53.28%) and minimum on Imam Pasand (13.33%). Lesion length ranged between 0.28-0.88 cm with largest and smallest lesion on Bennet Alphonso (0.88 cm) and Imam Pasand (0.28 cm) respectively. Range of latent period was 10-17.33 with minimum and maximum days of 10 on Bennet Alphonso and 17.33 on Imam Pasand respectively.

On different mango varieties, the isolate, WMnMa displayed considerable diversity variation for different pathogenic attributes like PDI, PDS, incubation days and lesion size. PDI ranged between 66.67-100 per cent. Cent per cent disease incidence was exhibited by WMnMa on Neelam, Imam Pasand and Banganapally and lowest incidence (66.67%) on Priyur, Muvandan and Bennet Alphonso. Range of PDS was 26.64-66.67 per cent with highest and lowest on Neelam (66.67%) and Bennet Alphonso (26.64%) respectively. Lesion length ranged between 0.41-0.78 cm with largest and smallest lesion on Banganapally (0.78 cm) and Priyur (0.41cm) respectively. Incubation days ranged between 10.33-17.33 with minimum and maximum of 10.33 on Banganapally and 17.33 on Priyur respectively.

Clear variation was detected for different pathogenic parameters like PDI, PDS, incubation days and lesion size by the isolate, KPB. PDI ranged between 66.67-100 per cent with maximum and minimum incidence on Priyur, Muvandan, Neelam and Banganapally (100%) and Imam Pasand and Bennet Alphonso (66.67%) respectively. PDS ranged between 26.64-70 per cent with highest and lowest on Neelam (70%) and Imam Pasand (26.64%) respectively. Range of lesion length was 0.56-0.95 cm with maximum and minimum lesion size on Muvandan (0.95 cm) and Bennet Alphonso (0.56 cm) respectively. Latent period ranged between 9-16 with minimum and maximum number of days on Muvandan (9) and Imam Pasand (16) respectively.

The isolate KoKBn exhibited evident variation for different virulence traits like PDI, PDS, incubation days and

lesion size on different mango varieties. PDI ranged between 66.67-100 per cent with maximum disease incidence (100%) on Priyur, Imam Pasand, Bennet Alphonso and Banganapally and minimum incidence (66.67%) on Muvandan and Neelam. Range of PDS was 33.3-66.67 per cent with maximum and minimum on Priyur (66.67%) and Muvandan (33.3%) respectively. Lesion length ranged between 0.31-0.9 cm with largest and smallest lesion on Bennet Alphonso (0.9 cm) and Neelam (0.31 cm) respectively. KoKBn took minimum number of days for symptom initiation of 10 on Bennet Alphonso and maximum number of days of 17.33 on Neelam and incubation days ranged between 10-17.33.

Different works have been done on pathogenic variability of different pathogenic fungi while the pathogenic variability of *Lasiodiplodia theobromae* has not been investigated yet. The variation observed in the current study is in congruence with the variation observed by Vitthal (2015) who studied pathogenic variability among 12 isolates of *Colletotrichum capsici* responsible for leaf spot of turmeric. According to his work, incubation days and number of spots ranged from 7 to 13 and 3 to 6 respectively. Range of disease incidence and severity was 35-66 per cent and 30.51-38.32 per cent respectively. Ahmad (2016) identified the isolate Kti 10 with average disease intensity of 10.3 per cent as most virulent among 12 isolates of *Exserohilum turcicum* when artificially inoculated on 12 maize genotypes. In another experiment, he studied variability of these 12 isolates on another maize cultivar, Pahalgam local cultivar wherein range of different virulence traits like incubation days, PDI, breadth and length of lesion was 4.07-7.74, 20.23-30.5 per cent, 0.48-0.95 cm and 2.88-8.42 cm respectively. Lakshmi Prasad *et al.* (2020) categorized 220 isolates of *Alternaria helianthi* into three different groups *viz*., low,

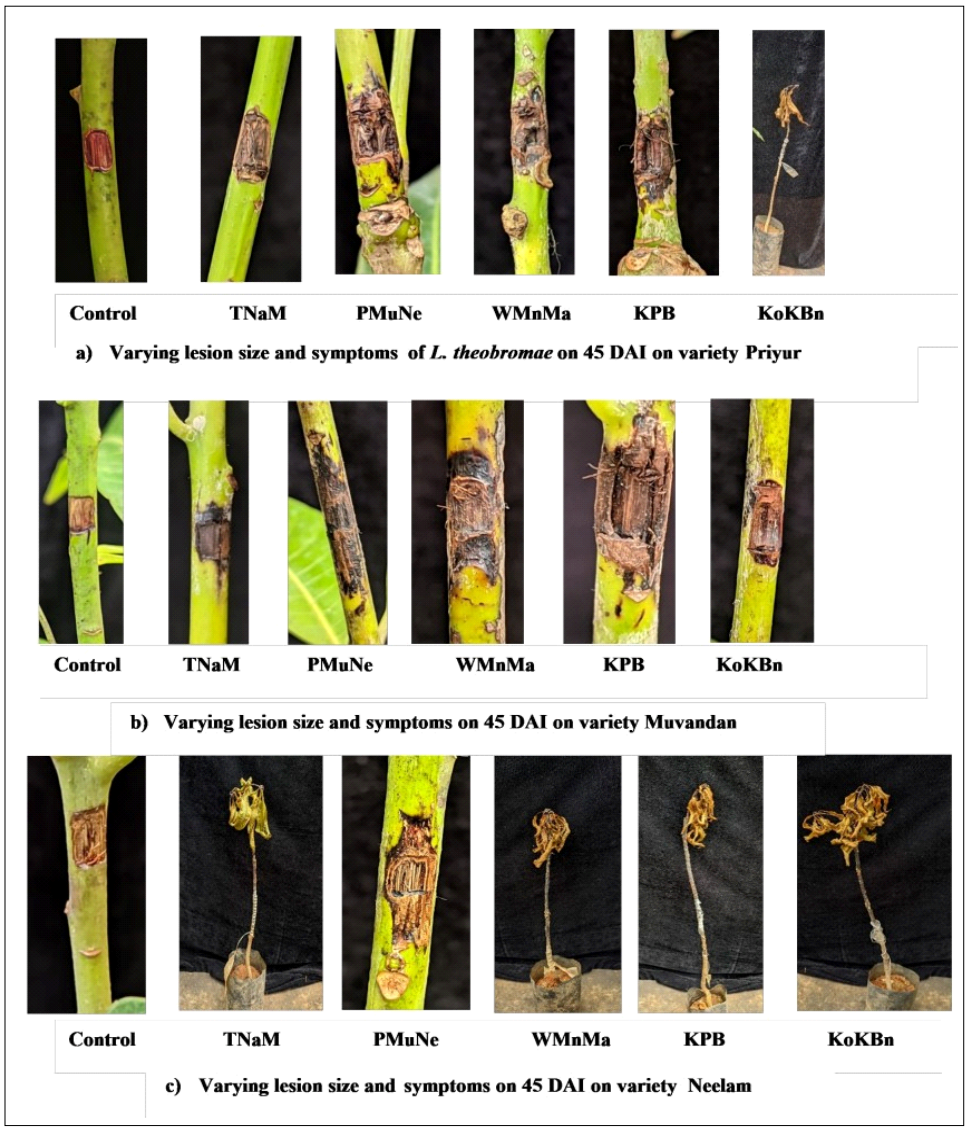


Fig 3i: Evaluation of pathogenic variability of *L. theobromae* isolates on different mango varieties.

medium and high aggressive groups based on disease reaction on six sunflower accessions. More number of isolates fell under medium aggressive group.

The pathogenic variability reported in this study in also in consonance with the variation observed by Mohammed *et al.* (2022) in isolates of *Alternaria porri* collected from different parts of Uganda which produced PDS ranging between 16.7-56.3% and the onion variety Red coach was found to be resistant. Das *et al.* (2023) also observed variation in nine isolates of *Fusarium oxysporum* in three wilt susceptible cultivars of lentil wherein PDI ranged between 75-100% and isolate W7 was found to be most aggressive. Pathogenic variability was evaluated for ten isolates of *Alternaria cyamopsidis* causing *Alternaria* blight

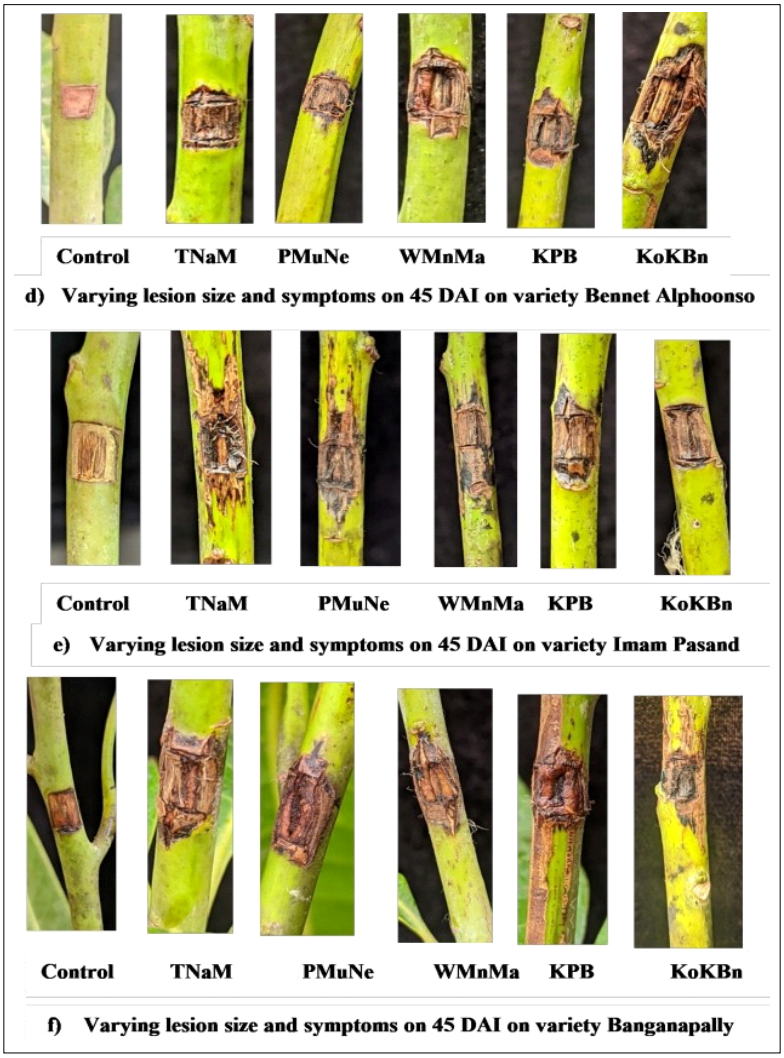


Fig 3ii: Evaluation of pathogenic variability of *L. theobromae* isolates on different mango varieties.

Table 4: PDI of selected five isolates against six varieties of mango.

Variety \ Isolates	I_1 (%)	I_2 (%)	I_3 (%)	I_4 (%)	I_5 (%)	Mean
Priyur	66.67	100.00	66.67	100.00	100.00	86.66
Muvandan	66.67	33.33	66.67	100.00	66.67	66.67
Neelam	100.00	66.67	100.00	100.00	66.67	79.99
Imam pasand	66.67	100.00	100.00	66.67	100.00	86.66
Bennet alphonso	100.00	100.00	66.67	66.67	100.00	86.64
Banganapally	100.00	100.00	100.00	100.00	100.00	100.00
Mean	83.33	83.33	83.33	88.88	83.33	

I_1- TNaM, I_2- PMuNe, I_3- WMnMa, I_4- KPB and I_5- KoKBn.

in clusterbean whose disease intensity ranged between 34.69-65.50%. The most and least virulent isolate was AlcyJp1 (disease intensity of 65.50%) and AlcyBk2 (disease intensity of 34.69%) respectively (Sharma *et al*., 2024). Pathogenic variability was evaluated based on size of necrotic lesion produced by *Macrophomina phaseolina* by Amrate *et al*. (2024). They concluded that the isolates Mp-1 to be most virulent (15.3 cm) and Mp-8, Mp-10 and Mp-12 to be least virulent (3 mm). Similar results were obtained in the area under the lesion progression curve also. Errampalli *et al*. (2024) categorized six isolates as highly virulent with >50% average mean over all the cultivars among 89 isolates of *Fusarium oxysporum f. sp. ricini* collected from 11 Indian states. Jojy *et al*. (2024) also noticed diversity in isolates of *Colletotrichum capsici* in pathogenic characters like incubation days, lesion size, and PDI which ranged between 1-3, 0.17-1.13 cm and 16.67-45.33% respectively.

The variation in symptom expression of different isolates on different varieties may be due to variation in climatic conditions or variation in inherent resistance of different varieties or variation in genotype of isolates. Genetic variation of *Colletotrichum* isolates inciting mango anthracnose (Bincader *et al*., 2022) and *Alternaria* isolates causing cotton leafspot (Sampathkumar and Raghavendra, 2024) was established using SSR primers and ISSR-PCR respectively. Variation may arise from interaction of above-mentioned factors also. Since India is one of the centres of origin of mango, it could be the centre of diversity of its pathogen *Lasiodiplodia theobromae* also. This can lead to pathogen variability because diversity of *Lasiodiplodia theobromae* has been larger in areas where natural host population exist than the regions where the host has been introduced.

Variability in phytopathogens is driven by various phenomena such as host specialization, selection, mutation,

Table 5: PDS of selected five isolates against six varieties of mango.

Variety \ Isolates	I_1 (%)	I_2 (%)	I_3 (%)	I_4 (%)	I_5 (%)	Mean
Priyur	26.64	39.96	33.30	46.62	66.67	38.62
Muvandan	39.96	19.98	39.96	66.60	33.30	39.96
Neelam	73.33	26.64	66.67	70.00	60.00	47.95
Imam pasand	26.64	13.33	39.96	26.64	46.62	30.63
Bennet alphonso	46.62	53.28	26.64	33.30	59.94	43.95
Banganapally	53.28	53.28	53.28	46.62	46.62	50.61
Mean	41.07	36.63	43.29	49.95	41.07	

I_1- TNaM, I_2- PMuNe, I_3- WMnMa, I_4- KPB and I_5- KoKBn.

Table 6: Lesion size* produced by selected five isolates of *L. theobromae* on six varieties of mango (45 DAI).

Variety \ Isolates	I_1 (cm)	I_2 (cm)	I_3 (cm)	I_4 (cm)	I_5 (cm)	Control	CV %	CD %
Priyur	0.40(a)	0.58(a)	0.41(a)	0.65(a)	0.70(a)	0.00(b)	46.71	0.38
Muvandan	0.58(ab)	0.30(bc)	0.61(ab)	0.95(a)	0.45(abc)	0.00(c)	58.24	0.51
Neelam	0.71(a)	0.38(b)	0.58(ab)	0.61(ab)	0.31(bc)	0.00(c)	42.08	0.33
Imam pasand	0.41(bc)	0.28(cd)	0.63(ab)	0.60(ab)	0.73(a)	0.00(d)	37.52	0.30
Bennet Alphonso	0.73(ab)	0.88(bc)	0.46(ab)	0.56(ab)	0.90(a)	0.00(c)	50.0	0.46
Banganapally	0.76(a)	0.73(a)	0.78(a)	0.78(a)	0.73(a)	0.00(b)	25.37	0.29

Table 7: Number of days* taken for symptom initiation/latent period/incubation days of selected five isolates of *L. theobromae* on six varieties of mango.

Variety \ Isolates	I_1	I_2	I_3	I_4	I_5	Control	CV %	CD %
Priyur	16(ab)	15(b)	17.33(a)	12(c)	11.33(d)	0.00(e)	6.42	1.39
Muvandan	15(a)	15.67(a)	12.67(b)	9(c)	15.67(a)	0.00(e)	4.55	0.93
Neelam	15(bc)	15.67(b)	14(c)	12.33(d)	17.33(a)	0.00(e)	4.5	1.03
Imam Pasand	16(b)	17.33(a)	13.33(c)	16(b)	11.67(d)	0.00(e)	3.50	0.79
Bennet Alphonso	11.67(c)	10(d)	16.67(a)	14.33(b)	10(d)	0.00(e)	4.16	0.79
Banganapally	12.67(c)	11.67(c)	10.33(d)	14.33(b)	15.67(a)	0.00(e)	5.26	1.03

I_1- TNaM, I_2- PMuNe, I_3- WMnMa, I_4- KPB and I_5- KoKBn.

* Mean of three replications.

recombination *etc.* Strong directional selection intensifies when a resistant gene becomes common in an area. This favours the increase in virulent mutant so that break down of resistance happens. The extent to which selection occurs is determined by degree of host specialisation resulting in increased variations in pathogen population.

Ability of *Lasiodiplodia theobromae* isolates collected from dragon fruit to infect other fruit crop such as mango, guava and banana (Briste *et al*., 2022) indicates that the fungus can infect multiple host species. Under natural conditions, this can lead to the change of pathogenicity level, contributing to pathogenic variability. Mango intercropping and mango trade across the state Kerala introduces diverse genotypes of the pathogen. Additionally, intercropping facilitates maintenance and continuous spread of distinct pathogen strains every season.

In the current investigation, the mean virulence index was found highest for the isolates KoKBn (4.19), KPB (4.08) and TNaM (3.35). Thus, these isolates were the most virulent among the five isolates. Range of virulence index was 2.78-4.19. This finding is in harmony with Vaniya *et al.* (2022) who reported that range of virulence index was 2.18-10 in her studies on variability of *Sclerotium rolfsii* isolates inducing stem rot of Indian bean.

Varietal screening for dieback resistance

In the present study, lowest CI was observed for Imam Pasand and highest for Banganapally. Range of CI was 27.08-50.61. This finding is in line with Sangeetha (2008) who screened thirteen mango varieties to dieback disease and reported that the range of CI was 0.63-31.88.

In the present investigation, based on CI value the varieties Neelam, Bennet Alphonso and Banganapally were categorized as susceptible (S) to mango dieback whereas the varieties Priyur, Muvandan and Imam Pasand were categorized as moderately susceptible (MS) to mango dieback (Table 10). This corroborates the reports by Jain and Depale (2023) who observed that maximum lesion size (69.99 cm^2) was produced in Neelam variety and it was categorized as susceptible to stem end rot of mango caused by *Botryodiplodia theobromae.* Findings in our present work is in slight conformity with Khanzada *et al.* (2015) who reported that the mango variety Neelam was showing moderate susceptibility to *Lasiodiplodia theobromae* inciting mango decline. In the current study, all the isolates produced 100 per cent incidence on the variety Banganapally, which proves its susceptible nature.

Differential response of different mango cultivars to mango dieback may also be related to variation in amount of different metabolites of the pathogen like defense related enzymes, antifungal compounds, phenols, pectin *etc* which has to be studied in detail. Different compounds like lasiodiplodin and jasmonic acid (Salvatore *et al.*, 2020), virulence factor of the pathogen like NmrA-like proteins (Peng *et al.,* 2022) have been isolated from *Lasiodiplodia theobromae.* Different varieties may be having varied amount of above mentioned compounds which might be the reasons for varying resistance and this aspect also has to be investigated in future research. According to some reports, the resistance of different mango varieties to dieback disease was related to presence of chitinase enzyme. According to Karunanayake *et al*. (2014) elevated

Table 8: Virulence index for different isolates of *Lasiodiplodia theobromae.*

Isolates / Variety	I_1	I_2	I_3	I_4	I_5
Priyur	1.66	2.66	1.95	3.88	6.06
Muvandan	2.66	1.33	3.33	7.40	2.22
Neelam	4.88	1.77	4.76	5.83	3.52
Imam Pasand	1.66	0.78	3.07	1.66	4.23
Bennet Alphonso	4.23	5.32	1.66	2.37	5.99
Banganapally	4.44	4.84	5.32	3.33	3.10
Mean	3.26	2.78	3.35	4.08	4.19

I_1- TNaM, I_2- PMuNe, I_3- WMnMa, I_4- KPB and I_5- KoKBn.

Table 9: Coefficient of infection for different varieties evaluated under pathogenic variability.

Isolates / Variety	I_1	I_2	I_3	I_4	I_5	Mean
Priyur	17.75	39.96	22.19	46.62	66.66	38.63
Muvandan	26.63	6.65	26.63	66.60	22.19	29.74
Neelam	73.33	17.75	66.67	70.00	19.99	49.54
Imam pasand	17.75	13.33	39.96	17.75	46.62	27.08
Bennet alphonso	46.62	53.28	17.75	22.19	59.94	39.95
Banganapally	53.28	53.28	53.28	46.62	46.62	50.61

I_1- TNaM, I_2- PMuNe, I_3- WMnMa, I_4- KPB and I_5- KoKBn.

Table 10: Categorization of different mango varieties based on CI value.

Mango varieties	Category
Priyur	Moderately susceptible (MS)
Muvandan	Moderately susceptible (MS)
Neelam	Susceptible (S)
Imam pasand	Moderately susceptible (MS)
Bennet alphonso	Susceptible (S)
Banganapally	Susceptible (S)

proportion of chitinase in mango cultivars Rata and Kohu contributed resistance to the infection caused by *Lasiodiplodia theobromae* inciting stem end rot in mango. Different scientists have documented differences in the accumulation of defense molecules in various cultivars. Varying level of phenol (Alañón *et al*., 2021; Mandal *et al*., 2021), terpenoids (Shimizu *et al*., 2021), phenol and antioxidant capacity (Troiani *et al*., 2022) and terpenoids and phenols (Tandel *et al*., 2023) have been reported in different mango cultivars. This variation in different compounds may be accounted for differential response of mango germplasm to mango dieback disease, necessitating further research. The differential reactions of the germplasm might be important to manage mango dieback through gene deployment.

CONCLUSION

Intraspecific variation in aggressiveness was observed for the isolates collected from diverse mango agroecologies of Kerala emphasising that the disease management strategies must be tailored accordingly. Although the study could not meet its goal of identifying resistant sources, the susceptible varieties could be either excluded from cultivation or genetically modified through resistance breeding to incorporate dieback resistant genes. Further exploration of pathogenic variability studies could involve more differential set of cultivars and more diverse array of pathotypes.

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Disclaimers

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Conflict of interest

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