

Screening small cardamom (*Elettaria cardamomum* Maton) field gene bank accessions for phenotypic characters, yield potential and disease resistance

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Abstract

Evaluation of 117 field gene bank accessions of small cardamom for phenotypic characters along with yield potential and disease resistance (rhizome rot and leaf blight) at ICAR-Indian Institute of Spices Research Regional Station, Appangala revealed significant variation with respect to phenotypic characters. The highest plant height (350 cm) and number of bearing tillers (36.4) were recorded in the accessions, field gene bank (FGB) 65 and FGB 16, respectively. The highest fresh weight of capsules (399.61 g) and maximum number of capsules (244.20) plant⁻¹ were recorded in the accession, FGB 13. Further, based on screening for disease resistance, 35 and 15 accessions of cardamom were identified as resistant to leaf blight and highly resistant to rhizome rot, respectively. Based on the multivariate cluster analysis, 117 FGB accessions were classified into 5 clusters *viz.*, clusters 1 and 2 encompassing equal number of accessions (44), cluster 3 with one accession (FGB 10), clusters 4 and 5 with 8 and 20 accessions, respectively.

Keywords: characterization, cluster, hotspot, leaf blight, rhizome rot, yield

Introduction

The major challenges in the cultivation of small cardamom are the prevalence of various diseases incited by pathogenic oomycetes, fungi, viruses and nematodes. Among the fungal diseases, leaf blight caused by *Colletotrichum* species (Govindaraju *et al.* 1998; Chethana *et al.* 2016) and rhizome rot incited by *Rhizoctonia solani*-*Pythium vexans* complex, are the most

widely spread, destructive and economically important. These diseases are cosmopolitan and prevalent throughout the cardamom growing tracts causing significant reduction in the yield by destroying the effective photosynthetic area (leaf blight) and rotting as well as subsequent toppling of the tillers (rhizome rot). Prevalence of the inoculum within the plantation and infective propagules disseminated from adjacent

fields and plantations overexposed to sunlight further favours the initial establishment and subsequent spread of leaf blight disease. Whereas, presence of inoculum in the soil as well as in the crop debris, overcrowding of plants and thick shade, favour development of rhizome rot disease. In plantations, leaf blight incidence attains maximum severity during November to February, whereas, severity of rhizome rot is found to be maximum during the month of August (Joseph & Suseela 1995).

Though these diseases could be managed by plant protection chemicals, in view of the increased demand for organically grown spices, health hazards to human beings and other mammals through biomagnifications, environmental pollution due to residual effect, enhanced concerns over the adverse effect of extensive and non-judicious application of plant protection chemicals and development of fungicide resistant variants of pathogens due to mutation, it is highly essential to evolve alternative strategies to vanquish these devastating diseases (Thomas & Bhai 2002).

Characterization of the germplasm is essential to provide information on the desirable traits of accessions, paving way for its utilization in the crop improvement programmes. Extensive exploration for natural resistance in the hotspots of genetic diversity forms the initial stage in identifying resistant sources which is the most reasonable, cost effective and sustainable strategy to manage diseases and thereafter their subsequent characterization. This strategy was successfully adopted in the identification and release of cardamom varieties resistant to rhizome rot, IISR Avinash (Venugopal *et al.* 2006), IISR Vijetha (Venugopal 2002) and Appangala 2 (Senthil Kumar *et al.* 2017) against *Katte*/mosaic disease and several accessions against leaf blight and rhizome rot diseases (Senthil Kumar *et al.* 2018; Biju *et al.* 2018). At National Active Germplasm Site (NAGS) of ICAR-IISR Regional Station, Appangala, about

600 cardamom accessions have been maintained which consist of genotypes collected from different hotspots of diversity.

Hence, the present investigation was carried out with the objectives to characterize small cardamom field gene bank accessions based on morphological characters, yield and to identify resistant sources against biotic (leaf blight and rhizome rot) stresses.

Materials and methods

Planting materials and location

Small cardamom field gene bank accessions (117 numbers) representing Malabar, Mysore and Vazhukka ecotypes maintained in the Field Gene Bank (FGB) at ICAR-Indian Institute of Spices Research Regional Station, Appangala, Karnataka comprised materials for the study. The FGB accessions were planted (five clumps per accession) during 2011 under uniform shade. Recommended plant protection operations as per package of practices of ICAR-IISR were adopted to manage insect pests and diseases other than leaf blight and rhizome rot.

Screening for disease resistance

Field screening of the cardamom germplasm for leaf blight and rhizome rot diseases was done by visual observation. Per cent disease index (PDI) was calculated by rating the severity of foliar symptoms expressed in a clump. The scoring of leaf blight incidence was based on the expression of the foliar symptoms on the inner tillers (minimum 8-12 tillers). PDI was calculated for all the plants of each accession and mean values were taken to compute PDI for each genotype (Praveena *et al.* 2013). The incidence of rhizome rot was recorded based on 1 to 5 disease rating scale (Venugopal *et al.* 2006). The disease rating scale was designed based on the number of infected tillers in a clump. For each accession, disease incidence in five clumps was

recorded and PDI was calculated based on the formula of Biju *et al.* (2018).

Statistical analysis

Observations with respect to vegetative and yield attributes were recorded for three consecutive years (2014–16) and the pooled data were subjected to multivariate cluster analysis using R version 4.0.1 with facto extra package for visualization. Intra and inter cluster distances were estimated using the clv package in R software.

Results and discussion

Phenotypic characterization

Morphological characters *viz.*, plant height, number of bearing tillers, number of capsules per plant and fresh weight of capsules exhibited significant variation among the genotypes (Table 1). Among the 117 FGB accessions evaluated, the plant height was found in the range from 100 to 350 cm. The maximum plant height (350 cm) and highest number of bearing tillers (36.4) were recorded in the accession, FGB 65 and FGB 16, respectively. Interestingly, the accession FGB 32 recorded highest number (40.3) of panicles, longest panicle (120.4 cm) and highest inter nodal length (5.65 cm). The maximum fresh weight of capsules (399.61 g), maximum number of capsules (244.20) plant⁻¹ and highest number of seeds capsule⁻¹ (26.74) were recorded in the accession, FGB 13. It is noteworthy to mention that the capsules of FGB 13 were found to be long (2.20 cm) and wider (1.45 cm) as compared to other accessions under study. High yield obtained in the small cardamom accession, FGB 13 may be attributed to the higher number of capsules plant⁻¹ as positive correlation was obtained between number of capsules plant⁻¹ and yield (Miniraj *et al.* 2000; Senthil Kumar *et al.* 2018). Akhila *et al.* (2017) and Senthil Kumar *et al.* (2018) also reported significant variation among the genotypes of small cardamom with

respect to morphological characters. These characters play an important role in identifying the desirable traits of progenitors to be included in the breeding programmes.

Among the yield characters, the highest coefficient of variation was shown by wet weight of capsules (63.26%) followed by number of capsules (55.52%) (Table 1). Among the morphological characters, number of panicles recorded highest coefficient of variation (52.05%) followed by inter node length (42.68%) whereas the lowest coefficient of variation was recorded in leaf breadth (9.80 cm). In contrary to the present findings, Akhila *et al.* (2017) reported highest coefficient of variation in number of vegetative buds (65.84%) followed by number of bearing tillers (20.18%). The significant variations obtained among the accessions for phenotypic characters in the present evaluation implied the presence of significant genotypic difference between the small cardamom accessions.

Clustering and genetic diversity analysis

Based on the multivariate cluster analysis, 117 FGB accessions were classified into 5 clusters at approximate height of 12 (Fig. 1). It was found that clusters 1 and 2 encompassed equal number of accessions (44), whereas clusters 4 and 5 consisted of 8 and 20 accessions, respectively. Interestingly, cluster 3 was solitary with a single accession (FGB 10) indicating its distinctness. Cluster 4 was unique from other clusters as it consisted of accessions (8) collected from one geographical location (Pampadumpara, Idukki, Kerala), while the other clusters consisted of collections representing various cardamom growing regions of Kerala and Karnataka. Even though, 117 FGB accessions represented Malabar, Mysore and Vazhukka ecotypes, no distinct clusters were formed based on ecotypes. The accessions from the same ecotypes were grouped into different clusters which also indicated the close relationship between these

Table 1. Mean characterization of small cardamom field gene bank accessions

Character	Range	Mean	S.D	CV	Promising genotype
Plant height (cm)	100.00-350.00	201.19	52.05	25.87	FGB 65, FGB 47, FGB 20, FGB 14
Bearing tillers	1.80-36.40	9.38	2.98	41.59	FGB 16, FGB 37, FGB 55, FGB 77, FGB 15, FGB 35
No. of leaves	14.60-241.00	124.61	44.86	36.00	FGB 73, FGB 71, FGB 77
Leaf length (cm)	32.40-66.20	57.25	5.73	10.01	FGB 72, FGB 36, FGB 18, FGB 20, FGB 26, FGB 33
Leaf breadth (cm)	7.36-11.32	9.27	0.92	9.89	FGB 73, FGB 20, FGB 22, FGB 9, FGB 52
No. of panicles	2.4-40.30	11.54	6.01	52.05	FGB 32
Panicle length	23-120.40	46.62	15.18	31.17	FGB 32
Inter node length (cm)	1.1-5.65	2.70	1.15	42.68	FGB 32
No. of capsules	11.50-244.20	179.81	99.82	55.52	FGB 13, FGB 45, FGB 46
Wet weight (g)	11.25-399.61	160.93	101.81	63.26	FGB 13
Capsule length (cm)	1.16-2.20	1.50	0.22	11.32	FGB 13
Capsule width (cm)	0.73-1.45	1.13	0.13	11.38	FGB 13
No of seeds fruit ⁻¹	7.4-26.74	15.46	3.92	25.38	FGB 13

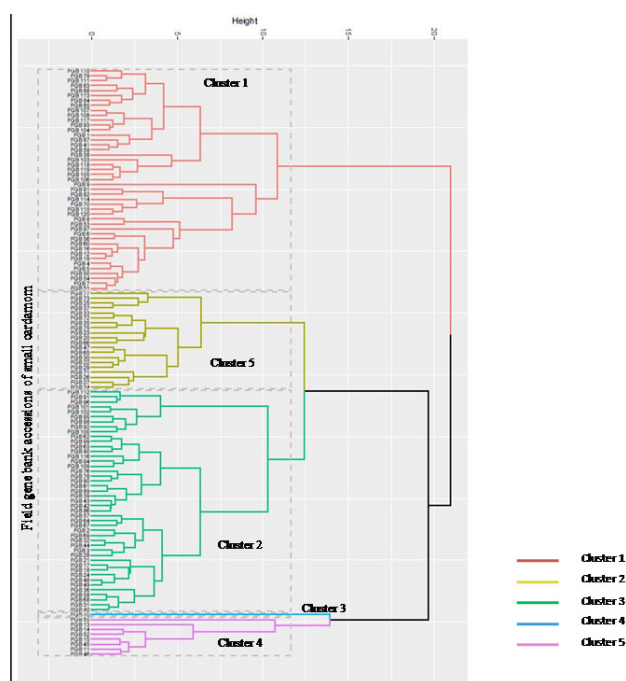


Fig. 1. Clusters of field gene bank accessions of small cardamom using Ward's method.
(The reader is referred to the web version of this article for interpretation of colours)

Table 2. Trait wise cluster means of field gene bank accessions of small cardamom

Cluster	No. of capsules	Wet weight (g)	Dry weight (g)	Plant height (cm)	No. of bearing tillers	No. of non bearing tillers	No. of panicles	No. of leaves	Leaf length (cm)	Leaf breadth (cm)
c1	66.328	50.094	16.758	200.638	5.822	7.155	4.172	88.029	54.792	9.024
c2	42.143	30.745	9.218	230.518	9.854	10.947	5.969	138.461	59.411	9.551
c3	140.800	121.000	42.000	304.000	10.600	10.800	6.800	147.200	61.000	5.450
c4	262.300	179.000	44.375	330.000	12.650	9.750	7.550	115.025	63.475	10.858
c5	71.053	54.615	16.610	278.100	14.470	10.340	9.900	165.320	64.650	10.829

Table 3. Distribution pattern of accessions into five clusters and estimate of average inter and intra cluster distances

	Inter cluster distances in Average method					No. of genotypes
	c1	c2	c3	c4	c5	
c1	0	110.5663	178.9619	304.2842	149.0731	44
c2	110.5663	0	168.7405	312.8675	104.6022	44
c3	178.9619	168.7405	0	177.4977	127.8945	1
c4	304.2842	312.8675	177.4977	0	270.8155	8
c5	149.0731	104.6022	127.8945	270.8155	0	20
Intra cluster distances in Average method						
	c1	c2	c3	c4	c5	
	106.424	65.949	0	194.885	97.118	

ecotypes. Similar inferences were also drawn by Prasath & Venugopal (2004) while categorizing 310 accessions of small cardamom into different clusters.

Trait (morphological and yield parameters) wise cluster means indicated that cluster 4 exhibited highest mean for the traits *viz.*, number of capsules, wet weight, dry weight, plant height and leaf breadth whereas the cluster 5 exhibited highest mean for the traits *viz.*, number of bearing tillers, number of panicles, number of leaves and leaf length (Table 2). These results indicated that the genotypes belonging to the clusters 4 and 5 could be the thrust areas of selection for genetic improvement. Further,

cluster 4 exhibited the highest intra cluster distance (194.88), indicating the prevalence of considerable diversity among the genotypes of the cluster whereas clusters 2 and 4 exhibited highest inter cluster distance (312.87) followed by the cluster 1 and 4 (304.28) (Table 3) indicating potentiality of the genotypes representing these clusters as parental materials for hybridization programmes. This is in line with the findings of Prasath & Venugopal (2004).

Identification of leaf blight and rhizome rot resistant sources

In the present study, attempts were made to identify resistance sources against leaf blight and

Table 4. Categorization of cardamom field gene bank accessions based on reaction towards leaf blight

Per cent disease index (%)	Category	Accessions
< 10%	Highly resistant	Nil
11 – 20%	Resistant	FGB 1, FGB 2, FGB 3, FGB 4, FGB 5, FGB 7, FGB 8, FGB 9, FGB 11, FGB 13, FGB 14, FGB 15, FGB 18, FGB 19, FGB 21, FGB 22, FGB 24, FGB 25, FGB 27, FGB 28, FGB 30, FGB 31, FGB 37, FGB 39, FGB 44, FGB 46, FGB 52, FGB 53, FGB 55, FGB 56, FGB 58, FGB 60, FGB 67, FGB 87, FGB 113
21 – 30%	Moderately resistant	FGB 6, FGB 10, FGB 12, FGB 16, FGB 17, FGB 20, FGB 23, FGB 26, FGB 29, FGB 32, FGB 33, FGB 34, FGB 35, FGB 36, FGB 41, FGB 45, FGB 47, FGB 48, FGB 50, FGB 54, FGB 57, FGB 59, FGB 63, FGB 64, FGB 65, FGB 66, FGB 68, FGB 71, FGB 72, FGB 73, FGB 75, FGB 76, FGB 78, FGB 84, FGB 86, FGB 89, FGB 90, FGB 94, FGB 97, FGB 98, FGB 118, FGB 119, FGB 120
31 – 40%	Moderately susceptible	FGB 38, FGB 40, FGB 42, FGB 43, FGB 49, FGB 51, FGB 61, FGB 62, FGB 69, FGB 70, FGB 77, FGB 80, FGB 83, FGB 85, FGB 88, FGB 91, FGB 92, FGB 93, FGB 95, FGB 99, FGB 104, FGB 105, FGB 106, FGB 107, FGB 110, FGB 112, FGB 114, FGB 115, FGB 116, FGB 117
41 – 50%	Susceptible	FGB 79, FGB 81, FGB 82, FGB 100, FGB 101, FGB 102, FGB 108, FGB 109, FGB 111
> 51 %	Highly susceptible	Nil

rhizome rot diseases among the FGB accessions (117) of small cardamom by categorizing them based on per cent disease index. The accessions exhibited variable reactions towards both rhizome rot and leaf blight diseases. It was inferred that, out of 117 accessions screened for leaf blight disease, 35 accessions were grouped under resistant category while, nine accessions were grouped under susceptible category. Forty three accessions were found to be moderately resistant whereas, 30 accessions were identified as moderately susceptible to leaf blight disease (Table 4). Among the genotypes evaluated for resistance against rhizome rot, fifteen accessions were grouped under highly resistant and twelve under resistant category. Forty seven and 41 accessions were grouped

under moderately susceptible and susceptible categories respectively whereas two accessions were found to be highly susceptible (Table 5). This was in line with the findings of Senthil Kumar *et al.* (2018) who identified 22 genotypes resistant to leaf blight and 29 genotypes highly resistant to rhizome rot. Praveena *et al.* (2013) reported two accessions *viz.*, IC-349613 and IC-349588 as highly resistant to leaf blight while Biju *et al.* (2018) reported that among the small cardamom accessions screened for resistance against leaf blight, 24 accessions of Malabar types, 24 accessions of Mysore types and 41 accessions of Vazhukka types exhibited resistant reaction to leaf blight disease.

In an attempt to identify small cardamom

Table 5. Categorization of cardamom field gene bank accessions based on reaction towards rhizome rot

Per cent disease index (%)	Category	Accessions
0.0 – 5.0	Highly resistant	FGB 1, FGB 3, FGB 8, FGB 9, FGB 13, FGB 21, FGB 22, FGB 26, FGB 28, FGB 29, FGB 30, FGB 45, FGB 52, FGB 60, FGB 118
5.1 – 10.0	Resistant	FGB 5, FGB 19, FGB 27, FGB 31, FGB 33, FGB 34, FGB 49, FGB 50, FGB 58, FGB 107, FGB 117, FGB 119
10.1 – 25.0	Moderately susceptible	FGB 2, FGB 4, FGB 6, FGB 7, FGB 10, FGB 11, FGB 12, FGB 14, FGB 15, FGB 16, FGB 17, FGB 18, FGB 20, FGB 23, FGB 25, FGB 32, FGB 35, FGB 44, FGB 46, FGB 47, FGB 48, FGB 51, FGB 53, FGB 54, FGB 55, FGB 56, FGB 57, FGB 59, FGB 70, FGB 75, FGB 76, FGB 78, FGB 81, FGB 82, FGB 83, FGB 84, FGB 87, FGB 97, FGB 105, FGB 106, FGB 108, FGB 110, FGB 112, FGB 113, FGB 114, FGB 115, FGB 116
25.1 – 50.0	Susceptible	FGB 24, FGB 36, FGB 37, FGB 38, FGB 41, FGB 42, FGB 43, FGB 61, FGB 62, FGB 63, FGB 64, FGB 65, FGB 66, FGB 67, FGB 68, FGB 69, FGB 71, FGB 72, FGB 73, FGB 77, FGB 79, FGB 80, FGB 85, FGB 86, FGB 88, FGB 89, FGB 90, FGB 91, FGB 92, FGB 93, FGB 94, FGB 95, FGB 98, FGB 99, FGB 100, FGB 101, FGB 102, FGB 104, FGB 109, FGB 111, FGB 120
> 50%	Highly susceptible	FGB 39, FGB 40

accessions that possess dual resistance against leaf blight and rhizome rot diseases, it was found that the accessions *viz.*, FGB 1, FGB 3, FGB 8, FGB 9, FGB13, FGB 21, FGB 22, FGB 28, FGB 30, FGB 52 and FGB 60 were found to have dual resistance against both rhizome rot and leaf blight diseases. Similarly, Senthil Kumar *et al.* (2018) reported thirteen genotypes of small cardamom resistant to leaf blight and rhizome rot diseases. Hence, these accessions can be considered as candidates for donor materials of resistant genes and can be incorporated in breeding programmes for developing varieties with appreciable yield potential and resistance towards both leaf blight and rhizome rot diseases.

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