

Screening methods to assess cocoa genotypes for resistance to black pod rot disease

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Introduction

Cocoa black pod rot disease (BPRD) caused by *Phytophthora palmivora* is one of the most economically important and widespread diseases of cocoa in the world including India and the loss due to BPRD exceeded \$400 million worldwide (ICCO, 2013). The symptom appears as a small water-soaked lesion on any part of the pod, at any stage of pod development (Guest, 2007). Breeding for black pod rot disease resistant is one of the most economically and environmentally friendly accepted management practices (Iwaro and Singh, 2004). The field tolerance of cocoa varieties may be correlated with resistance under controlled conditions. Under field conditions, the parameters used includes the number of pods infected per tree, lesion areas etc. The most widely used screening method under laboratory conditions is the detached pod assay which considers the number of lesions on the pod to grade pod for resistance/susceptibility (Nyadanu, 2009).

1. Field screening

Grading method for assessment of cocoa genotypes for field tolerance to black pod rot disease (BPRD)

Black pod rot disease (BPRD) caused by *Phytophthora palmivora* is causing severe damage during monsoon as well as post monsoon seasons in high rainfall zones. If the cocoa canopy is not properly pruned systematically, annually, the infection load is observed to be high. The main harvest season May-August coincides with monsoon and the inter-pod contamination is more, since cocoa yield is cauliflorous as well as cushion bearing. In general, while taking pod count, healthy and infected pods are being recorded for harvest data, which give high percentage of damage. Pods with >1 cm husk thickness prevents the disease from spreading to beans. If beans are not blackened, they can be used for fermentation. And so, the grading of infection in husk and beans are being followed to work out the grade index in the scale of 0-5, in which 0 for no infection and

Damage in pods



1	2	3	4	5
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Damage in beans



1	2	3	4	5
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Grade index	Classification
0	Highly resistant
> 0 - 1	Resistant
> 1 - 2	Moderately resistant
> 2 - 3	Moderately susceptible
> 3 - 4	Susceptible
> 4 - 5	Highly susceptible

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5 for severe infection. For example, if a tree is having a total of 40 pods, of which 30 pods are free of BPRD, 7 pods at grade 1, 2 pods at grade 2 and 1 pod at grade 3, the calculation will be $(30 \times 0) + (7 \times 1) + (2 \times 2) + (1 \times 3) = 14/40$ (total no.

of pods) and grade index will be 0.35. It denotes that most of the damage lies between 0-1 (Anon., 2024). The varieties showing yield throughout the year or late season or second season bearing are escaping from pod rot infection.

Reactions of cocoa parental clones and varieties / hybrids to black pod rot disease

S.No.	Parental clones	Grade index	Category	Varieties/ Hybrids	Grade index	Category
1	VTLC 11 x VTLC 5	1.50 1.30	Moderately resistant Moderately resistant	VTLCH 1	1.10	Moderately resistant
2	VTLC 61 x VTLC 66	0.50 1.00	Resistant Resistant	VTLCH 2	0.35	Resistant
3	VTLC 5 x VTLC 19	1.30 3.00	Moderately resistant Moderately susceptible	VTLCH 3	1.25	Moderately resistant
4	VTLC 5 x VTLC 19	1.30 2.00	Moderately resistant Moderately resistant	VTLCH 4	1.30	Moderately resistant
5	VTLC 64 x VTLC 69	1.20 2.50	Moderately resistant Moderately susceptible	VTLCH 5	0.61	Resistant
6	VTLC 32	1.10	Moderately resistant	VTLCC 1	1.00	Resistant
7	VTLC 1	1.00	Resistant	VTLCS 1	0.55	Resistant
8	VTLC 57	1.00	Resistant	VTLCS 2	0.60	Resistant

2. Lab screening

Detached pod method to assess the resistance in lab condition

Uniform-size pods of the same age (four to five month old) should be collected in early morning from 7.0 am to 10.00 am. Harvested pods must be kept carefully inside a transparent polythene bag. Not more than two pods are kept together to avoid surface damage to the pod during transit. Rinse the pods with running tap water, followed by washing them with sterile distilled water and set-out them on a tray lined with moist paper towels. Cover the tray with another tray to create humidity. Another method of incubation is the box method, which is ideal for

screening cocoa varieties for black pod rot disease resistance.

Box method of incubation



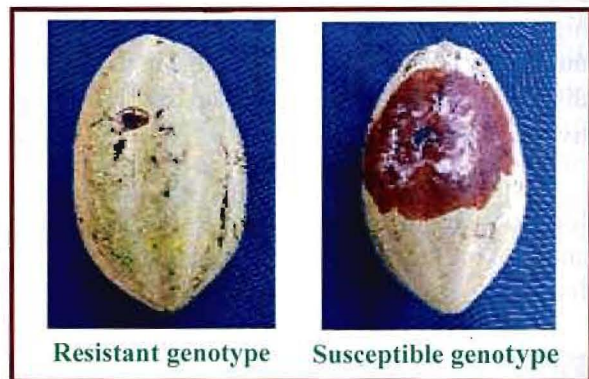
Disease rating and classification

Disease rating	Infection level	Classification
1	No symptom	Highly resistant
2	1-5 localized lesions	Resistant
3	6-15 localized lesions	Moderately resistant
4	>15 localized lesions	Partially resistant/ resistant to spreading of lesion alone
5	1-5 expanding lesions	Partially resistant/resistant to penetration alone
6	6-15 expanding lesions	Moderately susceptible
7	>15 expanding lesions	Susceptible
8	Coalesced lesions	Highly susceptible



Preparation of inoculum and inoculation

A virulent isolate of *P. palmivora* should be used. The culture should be grown on carrot agar medium for 10 days to obtain zoospore suspension. Inundate the culture plate with 10 ml sterile distilled water (chilled to 10°C), and refrigerate it for 25 minutes at 5°C followed by incubation in the dark at 25°C for 30 minutes. Ascertain the concentration of zoospore in the suspension



using haemocytometer and adjust it to 1,00,000/ml (Eskes *et al.*, 1998). Washed and dried pods were kept in a slanting position and sprayed with zoospore suspension using an atomizer from a 30 cm distance to avoid runoff. Place inoculated pods in a tray lined with a moist paper towel and cover it with a lid. Incubate the inoculated pods at 25°C inside a BOD incubator for four days and observe for lesion development.

Integrated black pod rot disease management

Apart from identifying resistant genotypes, integrated disease management practices should be adopted by preventing inoculum built up in the plot and its movement from soil to plant.

1. Following appropriate spacing and density in all cropping models.
2. Shade and canopy management through structural pruning practices to allow more sun light and airflow.
3. Leaf litter mulch and pruned branches should not be heaped over trunk to prevent stem canker.
4. Frequent harvesting at regular intervals during monsoon.
5. Sanitary pruning, removal and disposal of wilted cherelles, infected pods, pod husks, ant nests, soil tunnels built by ants on the trunk, weeds *etc.*
6. After pruning the cut ends should be swabbed with 10% Bordeaux paste/ *Trichoderma* paste and if required the exposed

trunk, branches and canopy may be sprayed with 1% Bordeaux mixture depend on the environmental conditions and as per the regional preferences



Susceptible genotype



Trichoderma pasting

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