

## STANDARDISATION OF SCREENING TECHNIQUES AGAINST 'AZHUKAL' DISEASE OF SMALL CARDAMOM (*Elettaria cardamomum* MATON)

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### ABSTRACT

Screening techniques were standardised for evaluating the susceptibility of cardamom cultivars against 'Azhukal' disease caused by *Phytophthora meadii*. A number of screening experiments were carried out in the laboratory and in the field using different forms of inocula such as zoospores, mycelial culture discs and infected cardamom seeds. In young seedlings, zoospore sprayings was found to be superior to seedling dip method. Mature plant parts such as leaves, rhizomes, panicles and capsules were inoculated with zoospores, mycelial culture discs and artificially infected cardamom seeds in laboratory and field conditions. Among the various methods tested, inoculation on cut tips of capsules of detached panicles with mycelial culture disc was found to be an easy and effective method of screening. Inoculation of panicles with artificially infected cardamom seeds was also found to be effective but inferior to culture disc method.

### INTRODUCTION

Small cardamom (*Elettaria cardamomum* Maton) is cultivated on a large scale in the tropical evergreen forests of Western Ghats. Among the many diseases that the crop, 'Azhukal' or capsule rot is the most serious one (Menon et al, 1972). The disease occurs in the high ranges of Kerala and brings about crop losses upto 30% (Nambiar and Sarma, 1976). Recent studies on the etiology of the disease showed that it is *Phytophthora meadii* Mc Rae of A<sub>2</sub> mating type (Anonymous, 1986). The field control of the disease through fungicides alone is not possible always since azhukal incidence is closely linked with heavy and continuous rainfall. It has been reported that crop resistance together with plant sanitation and fungicidal application plays an important role in achieving satisfactory disease control (Jacob, Okaisabor and Adebayo, 1973).

The objective of the present study was for evolving an effective and easy method for testing the resistance/tolerance of cardamom cultivars against *P. meadii* infection.

### MATERIALS AND METHODS

Pure culture of *P. meadii* was obtained from 'azhukal' affected capsules following the selective isolation technique described by Tsao and Guy (1977). For standardizing the techniques cv. Malabar LBC was used throughout.

#### Preparation of inocula

*P. meadii* was grown on carrot agar in darkness for 48 hours and the cultures were incubated in light for 24 hours at 25°C to induce sporulation. Zoospore suspension was prepared from sporulated culture discs by cold shock treatment (Tsao and Garber, 1970). For the culture disc inoculation

method, 10 mm sized mycelial discs were cut from 48 hrs. old culture using a cork borer. Seed inoculum was prepared by artificially inoculating green capsules with culture bits and incubating them for 7 days under humid conditions. Seeds extracted from fully infected capsules were used for inoculation.

### Methods of inoculation

(a) *Seedling dip method*: Four months old seedlings grown under sterile conditions in green house were removed, washed thoroughly and kept in a beaker containing 25 ml zoospore suspension at a concentration of 1.5 lakh spores/ml. Assessment of infection was made as percentage mortality after 4 days.

(b) *Zoospore spray method*: Spraying with zoospore suspension was done in 4 months old potted seedlings as well as on panicles of mature plants using an atomiser. Zoospore spraying was done on detached panicles in the laboratory and also on intact panicles in the field. One set of panicles sprayed in the field was kept in polythene cover to retain humidity while other set was kept without cover. The seedlings and panicles sprayed in the laboratory were also kept in moist chamber at 24-25°C with an R. H. of 75-80%.

Leaf and rhizome bits were also inoculated by placing a drop of zoospore suspension on them using a syringe.

(c) *Culture disc inoculation*: Inoculation of detached panicles was done in the laboratory as follows: A 5 mm culture disc was cut from actively growing culture and placed on the panicles at 10 cm distances at the nodal regions. A piece of moistened cotton was also kept and the inoculated panicles were placed in polythene bags to retain humidity.

Individual capsules on the panicles were inoculated by cutting their tips and placing

2 mm size culture disc as described by Weststeijn (1969). The inoculated panicles were incubated in moist chamber.

(d) *Inoculation with infected seeds*: Inoculation of panicles and capsules with artificially infected seeds was done as follows: Each nodal portion of the panicle or capsule tip was inoculated with infected seeds and incubated in moist chamber. Five to six panicles were screened each time and the experiments were repeated until consistent results were obtained. In all cases, the percentage of capsule infected was taken as the criteria for assessing the tolerance.

### RESULTS

Observations on seedling mortality and percentage of capsules/leaf bits infected were recorded upto 15 days after inoculation. Results of the screening methods in seedlings are presented in Fig. 1. In the seedlings, spraying with zoospores gave a high percentage of infection than the seedling dip method. In the dip method, infection was noticed as early as 4 days after inoculation. In the zoospore spray method the infection began on the 8th day and thereafter steadily increased upto 90% on 14th day.

Inoculation by zoospore spraying on detached and intact panicles also gave a high percentage of infection. In detached panicles, early symptom development was seen on the 4th day after inoculation while in the field the symptoms appeared after 8 days. Fig. 2 shows comparison between laboratory and field inoculations by spraying and also the difference in percentage infection between open and covered panicles in the field. Infection percentage was generally high in detached panicles. In field, a slightly higher percentage infection was noticed in covered panicles.

Inoculation on rhizomes with drops of zoospore suspension did not give infection to

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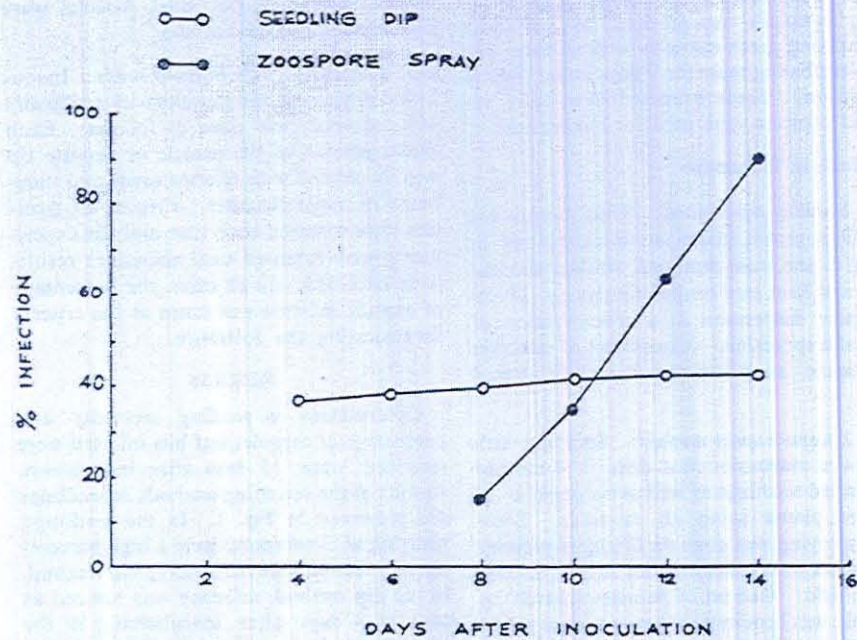


Fig. 1. Percentage infection of cardamom seedlings under seedling dip and zoospore spray methods.

detectable levels. In leaf bits, upto 35% infection was noticed within 5 to 6 days after inoculation. However, no consistent results were obtained with leaves and rhizomes. Results of culture disc and seed inoculation methods are presented in Table I. Inoculation on detached panicles with 5 mm size mycelial culture discs showed infection on the panicles and capsules. On the 8th day upto 24% infection was seen on the capsules. Lesions developed on panicle axis and these extended both upwards and downwards. A high percentage of capsule infection and early symptom development was noticed when cut tips of capsules were inoculated with 2 mm culture discs. Lesions appeared on the 4th day after inoculation from the cut portions and slowly extended towards the pedicel. Inoculation of panicles and capsule tips with

infected cardamom seeds also resulted in considerable extent of infection but only second to that of culture disc inoculation.

#### DISCUSSION

Different methods of inoculation tried showed considerable variation in the percentage infection. It has been shown that for a comprehensive assessment of resistance/susceptibility status of a cultivar, two or more methods of screening is desirable in addition to field resistance (Kularatne and Jacob, 1980). In seedlings, spray inoculation with zoospores was found to be more effective than seedling dip method.

Inoculation in the laboratory and field showed that in detached panicles the percentage of infection and severity are much higher

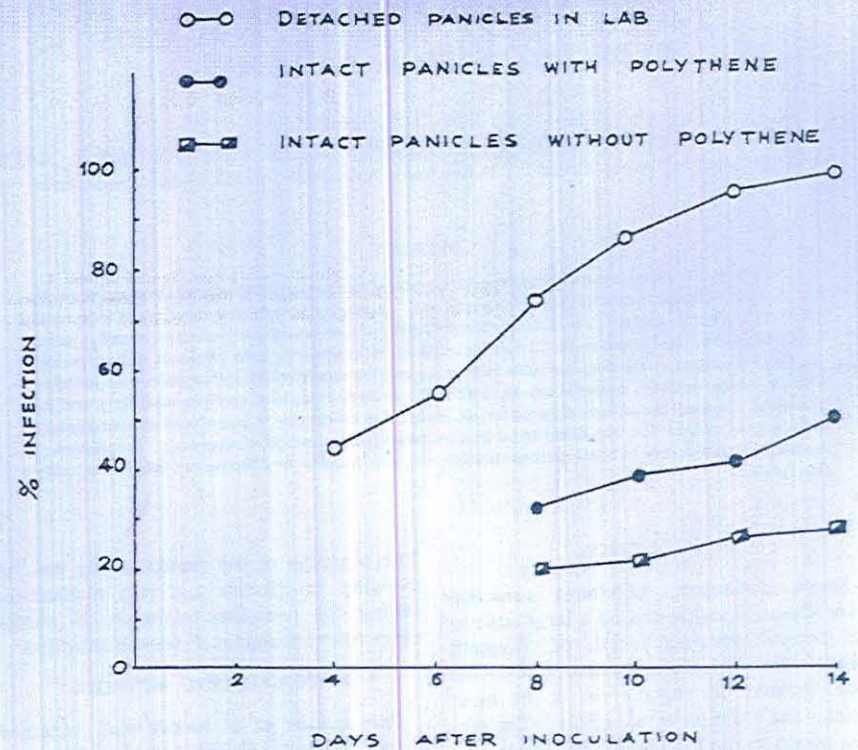


Fig. 2. Percentage infection of cardamom panicles in detached and intact conditions

Table I. Screening of bearing panicles of cardamom using different forms of inoculum

Treatment	No. of Panicles	No. of capsules inoculated	No. of capsules infected	Per cent infection on 8th day	Time taken for rotting
Zoospore spray	8	190	123	64.7	4-8 days
Culture disc (5 mm size) on panicles	6	112	27	24.0	8-10 days
Culture discs (2 mm size) on cut tips of capsules	8	190	126	66.5	4-5 days
Infected seed on panicles	6	178	72	40.5	8-10 days
Infected seed on cut tips of capsules	6	160	38	32.7	10-14 days
Infected seed inoculation on panicles in the field	12	227	46	20.30	14 days



than those in attached panicles. Similar differences have been observed in the case of cacao (Blaha, 1967). However, the degree of resistance in detached panicles under laboratory conditions could be used as an indication of a higher resistance that can be expected under field conditions (Lellis and Peixoto Filho, 1960).

Though spraying of seedlings and panicles with zoospore suspension showed higher disease incidence, the preparation of inoculum is much cumbersome and time consuming for large scale inoculations. For the quick production of large quantities of inoculum in the shortest period and getting maximum infection percentage, the culture disc inoculation method is superior to other methods. Placing culture bits on cut tips of capsules is advantageous in getting early and uniform capsule infections which can be rated as per severity. Though production of large quantities of inoculum as infected seeds is much easier than other methods, the seed inoculation method is time consuming and less effective as compared to the former.

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## A COMPARISON OF ISOLATES OF *KOLEROGA NOXIA* ON *COFFEA ARABICA*

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#### ABSTRACT

A comparison of isolates of *Koleroga noxia* Donk. (= *Pellicularia koleroga* Cooke, *Corticium koleroga* (Cooke) V. Hohnel, *Ceratobasidium* sp.) from three different zones of Chikmagalur District, causing black-rot disease of coffee, showed variation in their morphology *in vitro* and in basidial characteristics.

The pathogenicity tests carried out on *Coffea arabica* L. under modified propagation chamber in the nursery, using pure cultures of these isolates revealed difference in their virulence.

#### INTRODUCTION

Black-rot or koleroga disease on *Coffea arabica* L. by *Koleroga noxia* is prevalent in most of the coffee growing regions of the world and considered to be the second important disease in India. Blackening and rotting of the affected leaves, twigs and developing berries are the common symptoms. Affected leaves get detached from the branches and hang down by means of slimy fungal strands. The fungus develops on the surface as a film, which can be easily peeled off when wet. Continuous south-west monsoon without a long dry spell, atmosphere saturated with 95-100% relative humidity, thick over-head shade, sheltered from sun light and wind in valleys, frequent or continuous mist during monsoon are essential for the outbreak of black-rot.

Cooke (1876 a,b) first described the disease from the material collected in Mysore State and identified it as *Pellicularia koleroga*, as a hyphomycetous fungus. Hohnel (1910) re-

examined the collection of Cooke and identified basidia and basidiospore and re-named the pathogen as *Corticium koleroga* (Cooke) v. Hohnel. Coleman, Venkata Rao and Narasimhan (1923) isolated the fungus first time and proposed the name *Hypochnus koleroga*. Roger (1943) suggested for the revival of old name *P. koleroga*. Venkatarayan (1949) rejected the Cooke's nomenclature and opined that coffee black-rot fungus should be known as *Botryobasidium koleroga* (Cooke) Venkatarayan. Donk (1958) proposed new genus and species and re-named it as *Koleroga noxia* Donk. Talbot (1965) considered *Koleroga noxia* as synonym of *Ceratobasidium* sp. Talbot. Opinion differed between the investigators on the morphology of this fungus. Coleman et al. (1923), Narasimhan (1933), Mathew (1954 a) and Muthappa (1979) described the sclerotial stage apart from basidial or pellical stage. But Donk (1958) described it without sclerotia. Talbot (1965) observed the repetitive

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