

COLLETOTRICHUM GLOEOSPORIOIDES CAUSING INFLORESCENCE DIEBACK,
BUTTON SHEDDING, AND NUT ROT OF BETEL NUT PALM

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ABSTRACT

A severe disease on betel nut inflorescences that adversely affected fruit setting and development was observed in several plantations in India. The causal agent was isolated from the rachis, buttons, husk, and kernels and identified as Colletotrichum gloeosporioides. Pathogenicity of the isolate(s) of C. gloeosporioides was tested on the above parts and on green and yellow whole betel nuts in the laboratory as well as on different stages of the intact inflorescence in the field by artificial inoculations. Symptoms of the disease are described. This is the first report of C. gloeosporioides causing inflorescence dieback, button shedding, and nut rot of betel nut palm.

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Betel nut palm (Areca catechu), a perennial tropical crop that is cultivated in an area of about 174,300 hectares in India, is of considerable economic importance to this country. In 1973-74 we observed in several plantations of Karnataka and Kerala States -- the major betel nut growing States of India -- that about 60% of betel nut palms were infected by a disease on the nut inflorescences which adversely affected fruit setting and development. The disease appeared first on the rachis as brownish patches which soon spread and caused the rachis to wither. Shedding of young 50- to 60-day-old fruits, or buttons, followed. Rotting of mature nuts occurs at later stages. They appear shriveled and wrinkled and remain on the inflorescence or drop. The disease is present throughout the year, but is most serious during the dry period (February to May).

The purpose of this investigation was to determine the etiology of inflorescence dieback, button shedding, and nut rot of betel nut palm.

MATERIALS AND METHODS

Isolations: Brown and dried parts of the rachis were collected from betel nut palms, washed in tap water, blot-dried on filter papers, and cut into 5-mm pieces. Pieces were surface sterilized in 0.1% mercuric chloride solution for 1 minute and later washed in several changes of sterile distilled water to remove excess mercuric chloride. Pieces were then plated on oatmeal agar and potato-dextrose agar media and incubated at 30 + 2°C for 4-6 days.

Isolations were also made from buttons collected in polythene sheets spread around the base of palms, and from rotting husks of nuts and discolored kernels obtained from the intact inflorescence. Isolates so obtained were numbered from 1 to 10, 11 to 34, 35 to 42, and 43 to 50 to correspond to the rachis, buttons, husk, and kernels, respectively. They were identified and their single-spore cultures prepared for use in the later studies.

Laboratory inoculation test: One set consisting of 10-cm bits of rachis, whole buttons, ripened betel nut husk, and kernels (80 of each), and another set containing green-developing and yellow ripened whole betel nuts (20 of each) were surface sterilized as above and kept in humid chambers (85-90% relative humidity) in the laboratory. The surfaces of host parts in the first set were slightly injured with fine sterilized entomological pins and inoculated separately with conidial suspension (5000 spores/ml) of the isolates 1, 2, 3, 4, 11, 12, 13, 14, 35, 36, 37, 38, 43, 44, 45, and 46 (represent 4 isolates from each host tissue) obtained from 10-day-old cultures

growing on Richard's liquid medium at $30 \pm 2^\circ\text{C}$. Betel nuts in the second set were inoculated with conidial suspension of isolate 1 with and without injuring their surfaces. Suitable controls were maintained.

Field inoculation test: Field inoculation trials were carried out during March-April 1974 in the betel nut garden of Central Plantation Crops Research Institute, Regional Station, Vittal by selecting palms about 20 years old. Inflorescences at three stages of development, namely male flowers unopened (first phase), male flowers opening but female flowers still unopened (second phase), and female flowers just opened (third phase), were marked for studies. A conidial suspension of isolate 1 containing about 5000 spores per ml was prepared as above and sprayed with an atomizer on the selected inflorescences after washing their surfaces with sterile distilled water and injuring the rachis and the base of buttons lightly with sterilized pins. The inflorescences were then covered for 48 hr with polythene bags containing moistened cotton pads at the base of the bags to ensure high humidity. Controls were wounded in a similar way, but sprayed with sterile distilled water. A total of 10 inflorescences were maintained for each treatment.

All of the inoculated parts were observed daily for signs of disease development. Whenever disease developed, reisolations were made from infected portions to determine whether the inoculated organism was the one producing the observed symptoms of the disease. Pathogenicity tests were repeated to confirm the results.

RESULTS AND DISCUSSION

One organism was isolated constantly from diseased organs, namely the rachis, buttons, betel nut husk, and the discolored kernels. The isolate could be obtained readily from the basal and stylar ends as well as from the middle portions of the buttons. Fifty isolates were obtained and all were identified as Colletotrichum gloeosporioides Penz., the conidial state of Glomerella cingulata (Ston.) Spauld. & Schrenk. Identification was confirmed by the Commonwealth Mycological Institute, Kew, U. K.

In laboratory inoculations, the four isolates of C. gloeosporioides that we tested from the rachis, buttons, ripened betel nut husk, and kernels (namely isolates 1 to 4, 11 to 14, 35 to 38, and 43 to 46, respectively) all infected the above host tissues; this indicates that the isolates did not differ from one another in their pathogenicity. Symptoms produced by these isolates were similar to those described below for other inoculations. Isolate 1 produced water-soaked lesions on both injured and noninjured yellowish ripened nuts, but lesion development was more rapid in the former (lesion size 10-30 mm in 72 hr vs. lesion size 10-30 mm in 6 days in the latter). Later, a pinkish mass of growth in concentric rings was formed on the surface, and the husk shriveled (Fig. 1). The internal kernels were discolored to dark brown. No symptoms were noticed on the green unripe nuts, indicating that such nuts were resistant to fungal infection. On reisolations from the infected parts, pure cultures of C. gloeosporioides were obtained.

In field inoculation tests, brown discoloration and growth of the fungal mycelium were noticed at the injured sites of rachis and buttons only, when the inoculation was done during the first phase of the inflorescence. For inoculations made during the second and third phases of the inflorescences, the rachis became yellow, then turned brown, and finally dried up from the top downward with severe shedding of the buttons (Fig. 2). Shedding took place within 5-7 days when inoculation was made during the third phase, whereas it took 13-15 days when inoculations were made during the second phase of the inflorescence. A hyaline mycelium with abundant conidia and setae was noticed on the rachis as well as on the buttons. The stigmatic ends of buttons turned brown. Reisolations from the infected tissues always gave pure cultures of the fungus.



FIGURE 1. Ripe betel nut exhibiting shriveled husk and concentric growth of Colletotrichum gloeosporioides on the surface. Right -- Healthy ripe betel nut.

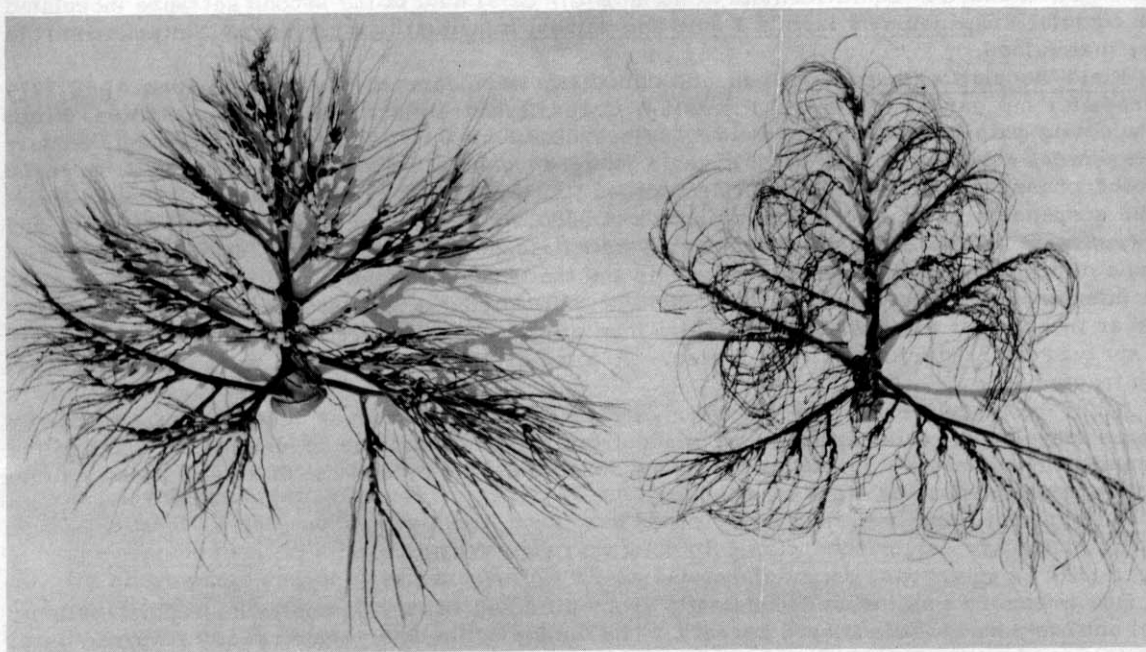


FIGURE 2. Left -- Healthy betel nut inflorescence. Right -- Dieback, caused by Colletotrichum gloeosporioides, of betel nut inflorescence showing brown rachis with fewer buttons.

Under natural conditions the fungal spores may gain entry into the parts of the inflorescence either through the scars left after the shedding of the male flowers or through the stigmatic ends of the opening female flowers. Therefore, it is possible that the shedding may be quicker if the spores are deposited on the stigma itself.

The fungus C. gloeosporioides is known to infect a number of economic as well as wild host plants in India (1, 2, 3, 4). Thomas (5, 6) recorded a Gloeosporium species as causing shedding of female flowers in betel nut; however, this is the first report to describe the role of C. gloeosporioides in causing inflorescence dieback, button shedding, and nut rot of betel nut palm.

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