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A Rot of Detached Durian Fruits Caused by *Sclerotium rolfsii*

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ABSTRAK

Satu penyakit reput buah durian yang baru bagi buah-buahan yang gugur telah diperhatikan di ladang buah-buahanUniversiti Pertanian Malaysia, Serdang, Selangor, semasa musim durian masak pada bulan Oktober, 1988. Penyebab reput ini membentuk lapisan bebenang miselium kasar yang padat, berwarna putih, berbentuk kipas yang tumbuh di atas tompok nekrotik perang berair pada buah yang bersentuhan dengan sisa rumpai yang mereput di atas tanah. Penyebab penyakit telah dikenalpasti sebagai kulat Sclerotium rolfsii. *Kehadiran pertumbuhan rumpai yang tebal di bawah pokok durian dan sisa daun rumpai yang mereput, serta keadaan lembab dan panas adalah faktor-faktor perangsang yang penting, yang menggalakkan perkembangan reputan tersebut.*

ABSTRACT

A new fruit rot of fallen durian fruits was observed in the Uiversiti Pertanian Malaysia fruit orchard in Serdang, Selangor during the fruit ripening month of October, 1988. The rot was characterized by a dense, white, fan-shaped mat of coarse mycelial strands of the causal fungus growing on a water-soaked, brown necrotic patch on the fruit in contact with decaying weed vegetation on the ground. The causal organism was identified as Sclerotium rolfsii. *Presence of a thick weed undergrowth and its decaying leaf debris, and warm, moist conditions were shown to be important predisposing factors conducive to the development of the rot.*

INTRODUCTION

Up to 1980, forty-five different microorganisms have been listed as associated with multitudinous disorders and diseases of durian, *Durio zibethinus* Murr. throughout Peninsular Malaysia (Johnston 1960, Singh 1980), Sarawak (Turner 1971), and Sabah (Williams and Liu 1976). Only one microorganism, the fungus *Rhizopus artocarpi* Racib., was listed as associated with a fruit rot of durian (Johnston 1960). Recently, a serious fruit rot of durian was reported to be caused by *Phytophthora palmivora* Butler (Lim and Chan 1986). This fruit rot was observed on intact unripe and ripe fruits and was particularly severe and rampant in several orchards in Selangor, Pahang and Johore in Peninsular Malaysia.

Lately, during the fruit ripening month of October, 1988, a new and interesting fruit rot of durian was encountered on fallen durian fruits in one of the University's durian orchards. Fruits which had fallen into the thick, profuse undergrowth of *Asystasia intrusa* Bl. ('Rumput Bunga Putih') a weed of the family Acanthaceae, around the base of durian trees and left uncollected for a few days were found to be infected by a soil-borne fungus. This paper reports on studies made on the isolation, identification, and pathogenicity of the fungus. Predisposing factors influencing the development of the fruit rot were also studied. Symptoms of the fruit rot and methods to avoid the fruit rot are also elaborated.

MATERIALS AND METHODS

Isolation

Diseased durian fruit tissue (5 x 3 x 1 mm) were taken from the advancing edge with a sterilised

scalpel after removal of the fruit epidermis and plated on potato dextrose agar (PDA). Mycelial strands of the fungus growing on the fruit surface were taken and plated directly on PDA. The PDA plates were incubated in the dark at 28° C. Hyphal tip transfers were made to obtain pure cultures from two-day old PDA cultures and pure cultures were maintained on PDA slants for use in the studies.

Cultural Studies

The following temperature regimes were used to determine the optimum and cardinal growth temperatures of the fungus viz. 8, 15, 20, 24, 28, 32, 35, and 40°C. The study was conducted using a completely randomized design and four replicate plates were used for each temperature level. Six mm diameter PDA discs taken from three-day old cultures were separately placed centrally on PDA plates and incubated at the above temperatures. Colony diameters were measured daily for four days and the data analysed using analysis of variance. Slides of the vegetative hyphae of the fungus were prepared for measurement. Sclerotia development and measurements were recorded after a week's incubation.

Pathogenicity Studies

Durian. Mature blemish-free durian fruits (cv. D 8) freshly fallen from the tree the night before were used. The surface of the fruit was surfaced-sterilised with 95% alcohol and inoculated with a 8 mm PDA disc of the test fungus taken from a four-day old culture. Fruits in the check treatment were similarly inoculated but with blank PDA disc without the test fungus. Inoculated fruits were placed in a) humid chamber with relative humidity (R.H.) of >95%, kept in a room of 28°C and b) room with a temperature of 28°C and R.H. of 75%. Eight fruits were used for each of the incubation conditions and the check treatments. Lesion developments were recorded up to six days after inoculation. Reisolation of the fungus was done on PDA.

Three–week–old durian seedlings were transplanted into sterilised and non-sterilised soil which were artificially inoculated with mycelial fragments and sclerotia of the fungus by incorporation into the medium. Another batch of seedlings transplanted into non-inoculated, sterilised soil served as check treatment. The seedlings were kept in a moist chamber at 28°C for three weeks and observed for development of disease.

Asystasia Intrusa Weeds. Asystasia weeds growing beneath the durian tree were uprooted and transplanted into pots filled with sterilised and non-sterilised soil taken from the durian orchard. Prior to transplanting, the soils were inoculated with the test fungus as in the case of the durian fruit above. Some pots were not inoculated and served as the check treatments. All the pots were kept in a humid chamber at 28°C. The trial was carried out in completely randomised design and replicated four times. Disease development was monitored for two weeks.

In addition, detached *Asystasia* plant parts were inoculated with the fungus in moist, covered, plastic containers and observed for colonisation of plant parts by the fungus.

RESULTS

Isolation and Cultural Studies

Isolation from diseased durian fruit tissues and fungal mycelial strands yielded the same fungus. The fungus was identified as *Sclerotium rolfsii* Sacc. – sclerotial state of *Corticium rolfsii* Curzi as it exhibited all characteristics of this fungus as described by Aycock (1966) and Mordue (1974). It grew readily on PDA, 20-25 mm/day and attained a diameter of 90 mm within four days. It produced a white, radiating colony of tough mycelial strands with a white colony reverse on PDA. The optimum temperature for growth was determined to be around 32°C; no significant difference in growth was found between 32°C and 28°C (Table 1). No growth was observed at 8°C and 15°C, and at 40°C the fungus grew sparingly. Abundant sclerotia (1-2 mm in diameter) were produced in more than a week old PDA cultures. The sclerotia were white at first, turning light brown and subsequently dark brown, and each had a characteristic thick rind with distinct medulla and cortex. Clamp connections were observed in the primary hyphae. Diameter of primary hyphae varied from 4-7 μm, while tertiary branches were

much narrower, 2 µm. The teleomorphic state was not observed.

TABLE 1
Effect of temperature on colony growth of *Sclerotium rolfsii*

Temperature (°C)	Mean colony diameter (cm)
8	0 g[1]
15	0.39 f
20	1.57 d
25	1.80 bc
28	1.86 a
32	1.89 a
35	1.73 c
40	0.47 e

df = 14
MSE = 0.01639
LSD = 0.07

[1] Mean values followed by similar letters denote no significant difference at P = 0.05 as determined by LSD test

Pathogenicity Studies

In the durian field, this peculiar fruit rot could easily be distinguished by the white, fan-shaped tufts of thick mycelial strands of the fungus growing on the water-soaked, brown, necrotic lesion on the durian fruit (*Fig. 1*) in contact with decaying *Asystasia* weed debris on the ground. The weed debris was caused by the durian fruit falling amongst the thick *Asystasia* undergrowth. Internally, necrosis extended into the seed cavity, rotting the seed and discolouring its cotyledons (*Fig. 2*).

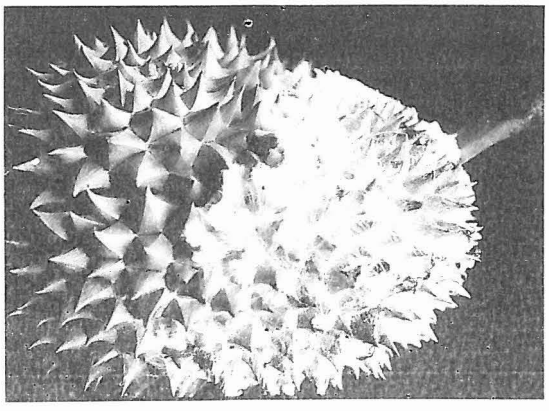

Fig. 1. Tufts of thick, white, fan-shaped mycelial strands of Sclerotium rolfsii *covering the brown, water-soaked, necrotic lesion on an infected durian fruit.*

Fig. 2. Internal necrosis caused by S. rolfsii *extending into the seed cavity, rotting the seed and discolouring the cotyledons (arrowed).*

The pathogenicity studies demonstrated that *S. rolfsii* was able to infect mature durian fruits without prior wounding, producing similar symptoms as those observed on fallen fruits in the thick *Asystasia* undergrowth. Fruits kept at both levels of relative humidity i.e. 75 and >95% at 28°C developed the rot. However, those incubated at the higher R.H. had more extensive rotting and more luxuriant growth of the fungus on the fruits. The fungus was easily isolated from infected fruits, thus satisfying Koch's postulates. The fungus did not infect the durian seedlings in both sterilised and non-sterilised soil.

S. rolfsii grew very well on the detached *Asystasia* plant parts but did not infect intact, healthy *Asystasia* plants in both sterilised and nonsterilised soils.

DISCUSSION

S. rolfsii is an ubiquitous fungus, found growing in a diverse array of soil types in the tropics and subtropics (Abeygunawardena and Wood 1957; Aycock 1966). In the soil it survives by means of its sclerotia and thick mycelial strands, growing saprotrophically on decaying plant debris and organic matter (Mordue 1974). Aycock (1966) reported that the fungus causes root and stem rot and other diseases in about 500 plant species from 100 families, most of which are dicotyledons comprising mainly composites and legumes. It is a facultative parasite, but has not been recorded to attack durian or its plant parts before. This

represents the first record of its facultative parasitism on durian.

The pathogenicity results indicate that durian is not a natural host of *S. rolfsii*. However, once its fruits became detached and fell on the ground where the fungus was present, infection of the fruit occurred especially under warm and moist conditions as was demonstrated in the pathogenicity tests on the fruits. Such a conducive environment in the orchard could be provided by the thick undergrowth of *Asystasia* weeds around the tree. Empirical evidence suggests that the falling durian fruit damaged some of the thick weed undergrowth and the damaged weed debris provided a ready food source for the luxuriant saprotrophic growth of the fungus. The fallen fruit was left *in situ* amongst the thick undergrowth, uncollected for a few days as it was hidden from the picker's view. Such a condition predisposed the fruit to infection by the fungus which was already established on the weed debris. This was also shown by infection studies on the weed: the fungus grew well on the detached *Asystasia* plant parts under warm, moist conditions but did not infect the healthy *Asystasia* under similar conditions.

To avoid *Sclerotium* rot of durian fruits, thorough and frequent (once or twice daily) picking of fallen fruits should be carried out. The base of the durian tree should be kept free from thick weed undergrowth which creates a warm and moist condition for infection of fallen fruits by soil fungi. Also, by having a thick undergrowth of weeds beneath the durian tree, many fallen fruits will remain unpicked as they will be obscured by the dense undergrowth. Besides, weeds compete with the tree for nutrients. Another way is to erect a strong canvas, nylon or wire netting beneath the durian tree to break the fall of the ripe fruits and to reduce fruit damage and mitigate the risk of infection by soil fungi. This can be a practical proposition especially with high priced cultivars like D 2, D 24, D 98, D 99 and D 114 where each fruit can fetch a price of more than $12 ringgit in the retail market.

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