surface concolorous with pileus, fibrillose, rugulose; stuffed. *Context* thin, indigo, changing pale yellowish-orange when bruised. Spore print reddish-pink. *Spores* 9.9–13.2 × 11.8–13.2 μm, tetrahedrally angular, often twinned tetrahedrons (one behind the other), hyaline, smooth, thin-walled, guttulate. *Basidia* 35.2–44.2 × 8.8–13.2 μm, clavate, hyaline, two-four-sterigmate. *Lamella-edge* fertile. Cystidia not seen. *Hymenophoral trama* irregular with narrow mediostratum, of thin-walled, hyaline hyphae 7.7–11 μm diameter. *Pileipellis* of repent, hyaline to pale brownish hyphae 7.7–16.2 μm diameter. *Stipe tissue* of moderately thin-walled hyphae 6.6–21.6 μm diameter with few refrative contents. Clamp connections lacking.

Basidiomata solitary among leaf litter under *Bambusa arundinacea* Willd. and *Dendrocalamus strictus* Nees, Kalinga, alt. 940 m, 2nd August 1983, HCIO No. 36811. The species hitherto unrecorded from India. The twinned tetrahedric spores are characteristic of it. The species also forms the first record of the genus from Orissa.

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OCCURRENCE OF DRY ROOT ROT OF *CARICA PAPAYA* L. CAUSED BY *MACROPHOMINA PHASEOLINA* (TASSI) GOID. IN TAMIL NADU, INDIA

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MACROPHOMINA PHASEOLINA (Tassi) Goid. causes the disease commonly referred to as dry root rot or charcoal rot under conditions of high temperature and drought stress[1]. In many cases, the disease develops in plants with poor vigour or at flowering and seed development stages. Many economically important plants, including cereal, legume, vegetable, fruit and fibre crops are attacked by this anamorphic fungus[1]. During January 1988, *Carica papaya* (var. Co 2), a well-known fruit crop of our country, affected by dry root rot following natural infection, was observed at the University Orchard, Coimbatore. Nearly 35.5% of the plants at full bearing stage were affected and suffered significant yield losses of up to 60%. Total damage occurred in plants that were subjected to moisture stress.

The first symptom of the disease was yellowing of leaves. Within a day or two, such leaves dropped off (figure 1). Dramatically sudden and more or less complete lodging observed in the field seemed to be a typical disease trait. Infection occurred exclusively on the secondary and tertiary roots; the fungus then moved into the primary root and then into the crown region. A red to black wet rot developed just before death and rapidly disappeared. Thereafter sclerotial formation took place, and measured 105–132 μm in diameter. In all severe cases, disintegration of the tissues in the lower stem portion, especially near the soil level, was noticed. On such plants, pycnidia developed at the collar and on the stem 0.5–1 inch above ground level. Pycnidia

Figure 1. Severely affected plant whose leaves have dropped off.

appeared as raised grey to black bodies. Microscopically, pycnidia were erumpent, oval to round with clear ostiole, and measured 72–187 μm (average 135 μm). Pycniospores were glassy, hyaline, elongate, and measured 9–22 × 5–8 μm (average 18 × 5.5 μm).

The pathogen was isolated on potato dextrose agar from the infected tissues of the roots and collar region. Pycnidia were induced successfully in a sand maize medium incubated at 30±2°C for ten days. The pathogen has been identified as *Macrophomina phaseolina* (Tassi) Goid., as the pycnidial and pycniospore characters closely resembled those described by Kulkarni *et al*[2], and later confirmed by the Commonwealth Mycological Institute, Kew, England (IMI No. 322857).

The pathogenicity of the fungus was tested on the papaya cultivar Co 2 at bearing stage by introducing an infested toothpick into the stem near soil level[3]. For an inoculum substrate, toothpicks (5 × 7 cm) were boiled for an hour in water, rinsed and autoclaved in potato dextrose broth. After a week at 28 C, the broth was decanted and a 5 ml suspension of sclerotia was transferred to the toothpicks. They were then incubated at room temperature. When the sclerotia became dense, the toothpicks were removed from the culture flask and dried at 28°C. Twenty-five inoculated plants and ten uninoculated plants were maintained. Twenty-five days after inoculation, all the inoculated plants showed yellowing and dropping of leaves. The control plants were free from the disease. The pathogen was reisolated from the infected tissue and found to be similar to those isolated from naturally infected tissues.

The role of pycniospores in fruit infection was studied *in vitro*. Pycniospore suspension was prepared as described by Ekundayo and Haskins[4]. Pycnidia were picked with an inoculating loop from ten-day-old cultures, dispersed in sterile distilled water and crushed to release the pycniospores. Fragments of pycnidia were removed by filtration through a loose non-absorbent cotton plug. The spore concentration was determined usjng a haemocytometer and adjusted to 5×10^5 spores/ml with sterile distilled water. The spore suspension was sprayed on unripe papaya fruits (var. Co 2) after pin-prick injury. Typical fruit rot symptoms appeared seven days after inoculation. Initially small, water-soaked spots appeared as circular specks on the fruit surface. The spots rapidly enlarged and became sunken (figure 2). The pathogen advanced deep into the fruit and caused rotting and disintegration (figure 3). With dry sclerotia from a ten-day-old-culture, similar fruit rot

Figure 2. Spots on fruit after infection with spore suspension.

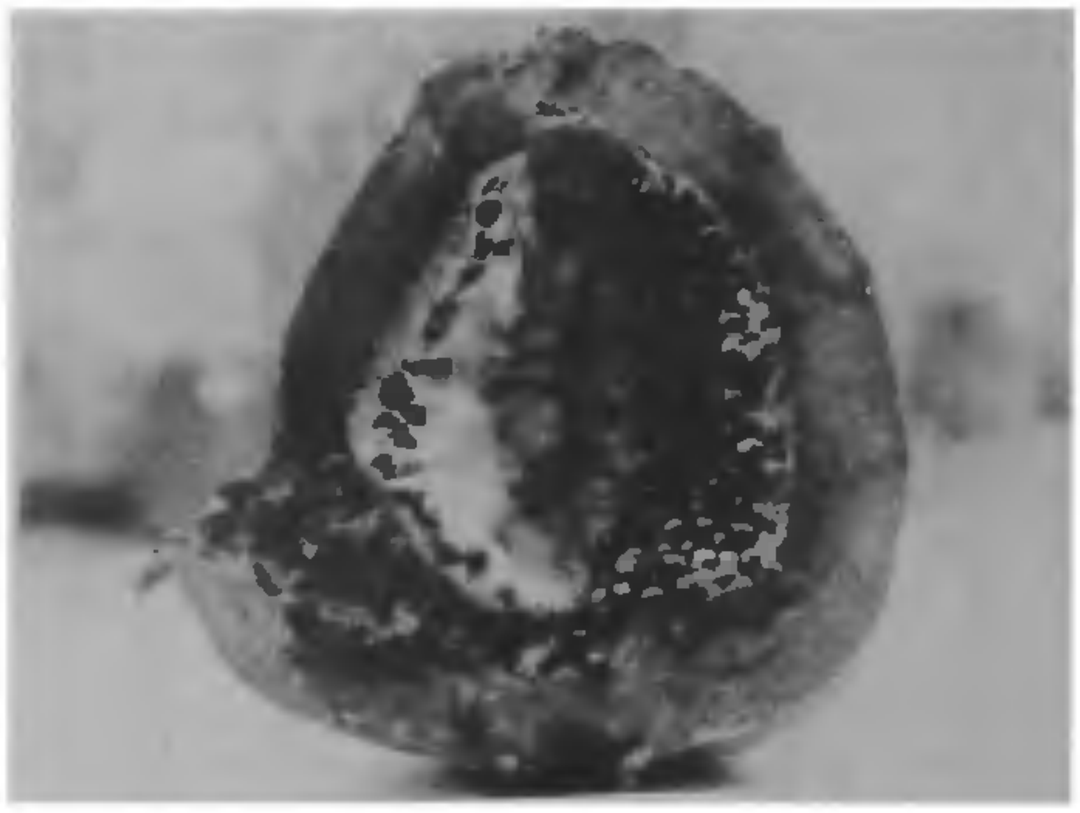

Figure 3. Rotting and disintegration of papaya fruit after artificial inoculation.

symptoms appeared twenty days after inoculation. Similar disease symptoms caused by *M. phaseolina* on papaya fruits were also observed by Kapur and Chohan in Punjab[5].

Although the natural host range of *M. phaseolina* appears to be quite extensive[1], artificial inoculation has not been successful in all the hosts. In a preliminary experiment, the seeds of *Sorghum bicolor* (L.) Moench. *Zea mays* L., *Pennisetum typhoides* (Burnm. f.) Stapf & Hub., *Triticum aestivum* L., *Cajanus cajan* (L.) Millsp., *Cicer arietinum* L., *Phaseolus vulgaris* L., *Dolichos lablab* L., *Vigna sinensis* Endl., *Phaseolus aureus* Roxb., *Phaseolus mungo* L., *Glycine max* Merr., *Arachis hypogea* L., *Gossypium hirsutum* L., *Ricinus communis* L. and *Capsicum frutescens* L. were sown separately in

steam-sterilized soil in greenhouse pots. Approximately 100 ml of a concentrated aqueous suspension of sclerotia from twenty-day-old petri dish cultures were added to each pot. In each crop, ten pots with five plants per pot were maintained. Plants that received water without sclerotia served as control. At flowering stage, the underground stems of the plants were injured with a sterile needle. Seven days after making the stem injury, the roots of all the plants were examined under a dissecting microscope and the infection diagnosed by the presence of distinct sclerotia. Laboratory isolation from these diseased roots confirmed the visual evaluation. Except *T. aestivum*, *C. cajan* and *D. lablab*, all the other hosts tested showed the presence of sclerotia with typical root rot symptoms. Though this pathogen has been reported on more than a hundred plant species from many parts of the world, there has been no report of this pathogen causing dry root rot disease on *C. papaya*.

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DETECTION OF INDIAN CASSAVA MOSAIC VIRUS BY ELISA

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MOSAIC disease is a major constraint in the cultivation of cassava, causing considerable yield loss[1]. It is caused by a geminivirus known as Indian cassava mosaic virus (ICMV)[2]. A similar disease in Africa is caused by African cassava mosaic virus (ACMV). ACMV and ICMV have been shown to differ in their reaction to monoclonal antibodies produced against ACMV[3]. As cassava is propagated vegetatively the disease spreads mainly due to inadvertent use of diseased stems. The damage caused by the disease can be checked to a large extent if the farmers are provided with certified healthy planting material. Main pre-requisite for such an effective screening is a reliable diagnostic method. With this aim double anti-body sandwich enzyme linked immunosorbent assay (DAS–ELISA) was tried to detect ICMV.

Antiserum to ICMV was obtained by immunizing a rabbit with ICMV purified from infected *Nicotiana benthamiana* following the method of Sequiera and Harrison[4]. DAS–ELISA tests were done as outlined by Clark and Adams[5]. Purified IgG from homologous antiserum to ICMV was used both as coating and conjugate antibody at 10^{-3} dilution. For conjugation, alkaline phosphatase (Sigma VII. P. 5521) was used in the presence of 0.05% glutaraldehyde at a concentration of 5000 units of enzyme/mg of IgG. Leaf samples were extracted with 0.01 M Tris-EDTA buffer with 0.1% Tween 20 and 2% polyvinyl pyrrolodine (MW 44,000), and two dilutions of each sample (10^{-1}, 10^{-2}) were used. For IgG coating, 200 μl of diluted preparation was put in each well of Dynatech microtitre plates and incubated at room temperature for 4 h. Plates were washed with washing buffer (PBS with 0.02% Tween 20). Immediately after washing, plates were coated with 200 μl of test samples per well. For each dilution two replicates were maintained and incubated overnight at 4°C. The plates were again washed with washing buffer before coating them with 200 μl/well conjugated antibody and incubated at room temperature for 4 h. After another round of washing 300 μl of *p*-nitrophenyl phosphate in diethanolamine buffer at pH 9.8 was added in each well and absorption read at 405 nm in a Dynatech microplate Reader-II after 1 h and overnight incubation.

Young and fully opened leaves from naturally infected plants of cassava cultivars, Kalikalan, H-226 and H-1687, showing severe symptoms were tested. In order to check whether the symptom-free plants are virus-free or not, leaves of meristem derived apparently healthy plants in the field were also assayed. Meristem derived glasshouse grown healthy cassava plants and uninoculated *N. benthamiana* were used as negative control. An absorption value