

INVESTIGATIONS ON THE BUD ROT DISEASE [*PHYTOPHTHORA PALMIVORA* (Butl.)] OF COCONUT

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II SUMMARY

Survey on the incidence of bud rot disease in India shows sporadic occurrence in all the coconut growing states except Gujarat. The intensity/frequency of disease varied considerably in different areas. The disease was more rampant in the South West coastal tract than the north east which is directly related to the annual rainfall. Microclimate (relative humidity and temperature) of coconut palms also showed direct correlation with the total precipitation which in turn was more favourable to the development of disease in young palms below 20 years than in adult palms.

Phytophthora palmivora (Butl) the pathogen associated with the disease was isolated from infected tissue collected from the basal part of the affected crown which showed symptoms of dry rot. *P. palmivora* was also isolated from 'Mahali' of arecanut, leaf fall of rubber, bud rot of palmyra and oil palm and capsule rot of cardamom. Selective medium containing mycostatin and/or thiamine HCl was used for isolation. All the isolates of *P. palmivora*, except that from cardamom which was not tested, infected coconut seedlings under controlled conditions of temperature and humidity. Cross inoculation on arecanut and oil palm seedlings and twigs of rubber, *Bougainvillea sp*, *Hibiscus rosasinensis*, *Artocarpus incisa* and *A. integrifolia* gave positive results. *P. palmivora* established infection on cut pieces of tender coconut petiole under controlled conditions in 72 hr, and produced a crop of sporangia in another 48 hr, which induced secondary infection within 24 hr. Temperature (22-24°C) favoured sporulation and growth of the isolates tested (P A 1, P R 1 and PP 1). *P. palmivora* (PC 1) produced oospores in culture media as well as plant tissue. The fungus was found to survive in infected plants for over 5 months. Laboratory trials indicated that species of *Pseudomonas*, *Xanthomonas* and *Erwinia* isolated from coconut crown hastened secondary rotting.

Screening trials in the laboratory suggest that, of the fungicides tested Demosan at 1200 ppm can effectively check infection of *P. palmivora*. Failure of disease incidence in the field prevented confirmation of these results under natural conditions.

III DETAILED REPORT

INTRODUCTION

Bud rot disease of coconut, wide spread and sporadic in occurrence was attributed to *Phytophthora palmivora* by Butler (1906) from India. Elsewhere intensive work on the etiology of the disease confirmed the specific identity of the causal agent as *P. palmivora* (Butler, 1925). Comparative morphological and pathological studies on *Phytophthora* from coconut, arceanut, cocoa, rubber, colocassia etc, revealed that they are all *P. palmivora* Butl. (Ashby 1927, Thomas *et al.* 1947, Gadd, 1927 and Reinking 1919). Butler (1910) reported that the disease is closely associated with weather conditions particularly relative humidity. Thorold (1955) working on black pod of cocoa also stated that the activity of *P. palmivora* is dependent on relative humidity. Short periods of high humidity followed by low temperature and free moisture are conducive for formation of swarm spores, development and infectivity of *P. infestans* (Crosier, 1934). Menon and Pandalai (1958) observed that the disease is more common in young palms aged upto 40 years. Earlier study on the symptomatology of the disease at this station revealed that the first visual symptom is the paling and slight yellowing of the 3rd or 4th leaf in coconut (Plate 1). Expression of similar symptoms or breakdown of the spindle is the next visible indication of the progress of the disease. Dissection of the crown of a palm which succumbed to the disease showed that the bud is rotten and the infection extends down to the 7th or 8th leaf. The spadices in the respective leaf axils also showed sign of infection. The younger three leaves and the tender inflorescences in their axils were healthy. This is suggestive of axillary infection occurring below the 3rd leaf extending both inward and downward.

The seasonal occurrence of bud rot disease and the influence of environmental conditions on *Phytophthora* suggest the need for a better understanding of the biology of the pathogen and correlation of the disease with weather factors and microclimatic conditions. Detection of the disease in the early stages so as to save the palm is rather difficult if not impossible. Hence adoption of prophylactic measures may prove more effective than curative methods for checking the disease and consequential loss. In view of the morphology of the coconut crown and nature of infection of the pathogen, fungicides having systemic property may be more suited for the successful control of the disease (Plate 2).

Survey on the distribution and intensity of bud rot disease in the different coconut growing states of India

Detailed village to village survey was carried out in Kerala on the West Coast and Tamil Nadu in the East Coast. In the other coconut growing States viz. Mysore, Andhra Pradesh, Orissa, West Bengal, Assam, Maharashtra and

PLATE 1
PLATE 2

Fig. 1 Bud rot affected coconut showing the first visual symptom of yellowing in the central spindle.

Fig. 1 A palm affected by bud rot and recovered on treatment with Bordeaux paste in the early stage of disease.

Fig. 2 Longitudinal section of the crown infected by bud rot. Brownish lesions along the space between peltates indicate the path of spread of rotting down towards the vegetative bud which in this instance escaped infection.

PLATE 2

Fig. 1 A palm affected by leaf rot and recovered on treatment with Bordeaux paste in the early stage of disease.

Gujarat and the Union Territories of Goa and Mahe random survey was conducted. Percentage incidence of disease was calculated based on the infection recorded in groups of palms in half an acre plots.

RESULTS.

In general, the disease was noticed in various types of soil viz. sandy loam, reclaimed clayey, red loam and gravelly laterite distributed from seashore to hill top and on river banks. In Kerala a total of 437 Revenue villages and 2053 plots were observed (Table 1). Intensity of disease incidence varied from 0.1 to 6.5 per cent, this being high in Quilon and Alleppey Districts. Occasionally heavy incidence of disease to the extent of 35.0—40.0 per cent, was observed in a couple of gardens having large number of young palms. Along the east coast in Tamil Nadu occurrence of the disease ranged from 1.45 to 3.6 per cent. Bud rot disease is prevalent in four out of 14 coconut growing districts in Mysore. The disease was more rampant in the coastal districts of South and North Kanara than in Chitradurga and Tumkur. As observed in Kerala severe incidence of disease affecting nearly 40.0 per cent of palms in a young plantation was also noticed in Tumkur. In the Union Territory of Goa the coconut crop account for over 18,000 ha. consisting mainly of old plantations. Confined to the interior tract, bud rot disease rarely occurs in palms below 20 years old. In Maharashtra State also the disease was noticed only in areas where large numbers of young palms exist. At the Regional Coconut Research Station, Ratnagiri 31 cases of bud rot were noticed in a collection of 3000 palms aged 3 to 12 years. Similar observations were made at Vengurla also. In Kolaba and Thana Districts bud rot was not detected. In the Saurashtra area of Gujarat State coconut gardens mostly young plantations along the coast from Mangrol through Veraval to Mahuva were surveyed. Bud rot was not noticed in any of the gardens. Certain localities of the major coconut growing areas of West Godavari, Kistna and Chittoor districts of Andhra Pradesh had scattered occurrence of the disease to the extent of 0.9 to 10% in groups of 100 to 600 palms. Palmyra palms growing in the same area, particularly in Kistna district were affected severely. Unlike in coconut where young palms are more susceptible to the disease, in palmyra adult palms—30 to 50 years old are generally the victims of the disease. Symptoms of the disease consist of paling and drying of the youngest leaf which stands erect. External symptoms, as in the case of coconut, do not indicate the extent of internal damage. By the time clear symptoms of disease are visible the crown is damaged beyond recovery. Coconut growing areas in West Bengal, Assam and Orissa were found to be free from disease during the period November—December.

Occurrence of the disease is confined to the South West and North East Monsoons. Generally palms aged 5 to 15 years are more susceptible to the disease.

Studies on the meteorological and micro-climatic factors in relation to the disease.

PROCEDURE.

Data on rainfall, temperature and humidity were recorded at Kayangulam and Kasaragod Research Station observatories from May to October for five years from 1968 to 1972. Observations on the micro-climate were initiated in 1968 on a group of 20 palms belonging to 4 age groups — 3 to 5 years, 6 to 10 years, 11 to 15 years and 16 to 20 years. Temperature and relative humidity were recorded at the leaf axil between 9 and 10 am using Assman Psychrometer. During 1969 besides the 4 groups of palms selected earlier 5 palms in the age group 21-40 years were also taken for observations as rare instances of bud rot cases occurred in 30 or 35 year old palms.

In 1970, collection of data on micro-climate was modified to provide a critical assessment based on the earlier information. Observations were taken on 50 palms of five age groups from 9.30 hr to 16.30 hr adopting Latin Square Method of randomization. Relative humidity and temperature were recorded at five periods at the leaf axils of groups of 10 palms of five ages at a time and in the open air at three locations — at Kayangulam and Kasaragod in Kerala and at Muthupet in Tamil Nadu.

Since the data collected in 1970 indicated that the late forenoon and afternoon readings have no bearing on disease development, further observations were made in the early forenoon i. e. from 7.30 hr to 9.30 hr on 30 palms of different ages in a randomized manner at two locations on the West Coast (Kayangulam and Kasaragod) and one on the East Coast (Pattukottai). As the trend in microclimate in relation to age of palms was similar in the different areas, the observations were confined to Kayangulam during 1972.

RESULTS.

Pre-monsoon data, recorded in May hardly indicated any difference between the microclimate of palms of different ages. However, with the onset of S. W. monsoon, palms in the age group 5 to 10 and 11 to 15 years recorded lower temperature and higher relative humidity as compared to younger or older palms. Positive correlation with rainfall and disease incidence was observed at three locations — Kayangulam, Kasaragod and Ratnagiri (Table 2).

Reduction in rainfall was reflected in micro-climate and development of disease (Table 3). Days which recorded high humidity (95-100%) are termed as "favourable day" for infection. At Kayangulam favourable days occurred for 3 to 6 days in 1969 while it was 13 to 20 days in 1968. As against 31 cases of bud rot observed in the farm in 1968 only 13 cases were recorded during 1969. Of the 13 palms infected in June, 1969, 5 made natural recovery by August.

Incidence of disease showed an upward trend for about four weeks during the monsoon period, subsequently it decreased when reduction in precipitation and consequent fall in relative humidity occurred. Occurrence of the disease was noticed in palms aged 6 to 20 years (Tables 4 and 5).

In 1970 considerable variation in weather factors like rainfall and temperature was observed between Kayangulam and Kasaragod on the West Coast. Total rainfall received at Kasaragod was higher than at Kayangulam which influenced the duration of favourable days. This was again reflected in the incidence of disease. At Kayangulam disease was observed in only one palm while at Kasaragod 24 palms were affected (Table 6a and b).

The data on microclimate for a 10 week period from June to September (1970) reveal that 3 to 20 year old palms have higher humidity, the maximum being in the 11 to 15 year old palms. This holds good for open air data at the crown level of the palms. Duration of 'favourable days' was also higher in palms upto 20 years old than in older palms. However, the data are not statistically significant.

Analysis of the microclimatic data recorded in 1971 at Kayangulam and Kasaragod reveals significant variation between palms of different ages. On an average palms aged 16 years and more recorded significantly higher temperature (25.4°C — 25.7°C) than that of 3 to 10 year old (25.28 — 25.5°C). No significant difference was observed between the age group 3-5 and 6-10 years and between 16 to 20 years and above 20 years. At Kasaragod the average relative humidity was also significantly different in the five groups of palms, the maximum being in 3 to 5 year old and minimum in palms above 20 years. No significant difference was observed in the data recorded at Kayangulam (Table 7).

Number of 'favourable days' observed at Kayangulam was 7 to 13 and at Kasaragod 45 to 53 which is correlated with total rainfall i.e. 1619.3 mm in 63 days and 2964.3 mm in 74 days respectively. Seven palms aged 5 to 20 years were affected by bud rot at Kasaragod while only one was observed at Kayangulam.

The average temperature in the leaf axils and open air at the crown level of palms of different ages showed significant variation at Kayangulam in 1972. Both the values were found to be minimum in the palms of the age group 11-15 years. The humidity percentage on the contrary showed significant difference only in the open air but not in the leaf axils. Total rainfall during this period was only 1021.7 mm in 56 days - the duration of days having high humidity varied from 20-23 days. Maximum humidity percentage during these days was 95-97, except on a single day when it touched 99 per cent. Temperature varied from 23.0 to 26.5°C . (Table 8).

There was no incidence of disease at the Research Station farm where the microclimatic data were recorded. However occurrence of the disease was observed nearly 5 KM away in the backwater area. The coconut gardens in this area were almost inundated for most part of the year. Young palms aged 10-15 years growing on bunds were found to be affected.

The data collected at Muthupet on the East Coast in 1970 show that the microclimatic parameters hardly touched the favourable level during the period under observation. Relative humidity varied from 69 to 93 per cent at 9.30 hr. But the trend in the microclimate remained the same as on the west coast viz. 3 to 20 year old palms had higher humidity than older palms. Disease incidence was not recorded at the observational centre but rare occurrence was noticed 20 to 30 KM away.

At Pattukottai significant variation in temperature and relative humidity in palms of different age groups was observed in 1971 but neither the temperature nor relative humidity provided favourable condition for the disease.

Data collected on microclimate with thermohygrographs during July-August and October-November, 1972 was found to be erratic.

STUDIES ON THE PATHOGEN

I ISOLATION OF PATHOGEN

(a) INFECTED MATERIAL

PROCEDURE:

Several methods for the isolation of pathogen from infected material washed in running water, surface sterilized with 0.1 per cent mercuric chloride or 1/14 calcium hypochlorite, without surface sterilization or after maceration were adopted. During later isolations unsterilized infected materials were floated overnight in tap water and chilled at 5°C for 30 min. after changing the water. The water used for chilling as well as the infected tissue were cultured. Glucose nitrate medium (Hendrix, 1965) and rubber leaf extract were also used as substrate, cholesterol 10 mg and 20 mg/l. and thiamine hydrochloride (2 ml of 1000 ppm solution per l.) were additionally admixed with the culture media. Potato tubers and papaya fruits were also used as baits for isolation of the pathogen. The agar media used for culturing were host tissue extract, potato-dextrose, oat meal, papaya extract and bean extract incorporated with 50 ppm Penicillin and Streptomycin, 30 ppm Rose Bengal and 100 to 500 ppm gallic acid.

PLATE 3

PLATE 3

Fig. 1 Coconut palm which succumbed to had rot infection. At the top is the remnant of the eaten crown, surrounded by a few leaves and old leaf bases having water soaked lesions of infection at the point of attachment to the crown. The central tissues are rotten due to the disease.

Fig. 2 Leaf base showing shallow lesions of infection in the middle portion which attached to crown. Discolouration near the base is due to the lesions formed inside an old petiole base.

Fig. 3 Longitudinal section of the damaged crown showing the spread of



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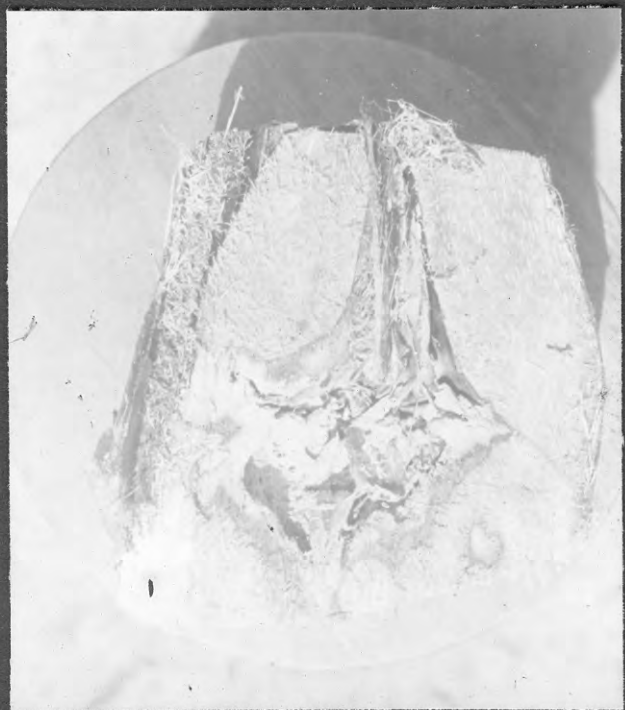
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PLATE 4

- Fig. 1 Coconut palm which succumbed to bud rot infection. At the top is the remains of the rotten crown surrounded by a few leaves and bunches.
- Fig. 2 Top view of the damaged crown. The central tissues are rotten due to the disease.
- Fig. 3 Lateral view of the damaged crown. Discolouration near the bottom is due to the lesions formed inside an old petiole base.
- Fig. 4 Longitudinal section of the damaged crown showing the spread of rot inside.



3



4

(b) SOIL

PROCEDURE.

Selective media of Flowers and Hendrix (1969) and oatmeal agar with Pimaricin (10 mg/l), nutrient agar and soil extract agar were used to isolate the pathogen from soil collected from the base of infected trees.

RESULTS.

(a) INFECTED MATERIALS

During the first two years (1968 and 1969) all attempts to isolate *Phytophthora* sp. from coconut failed although the same fungus could be isolated from 'mahali' affected *Areca triandra* and leaf fall of rubber. Glucose nitrate agar with thiamine HCl and Mycostatin, bean extract agar and potato dextrose agar + thiamine proved useful for this purpose. Similarly samples from bud rot affected oil palm (Kerala), palmyra (Andhra Pradesh) and cardamom (Kerala) yielded *Phytophthora* sp.

Infected samples from coconut were collected for isolation from the older leaf bases showing lesions of infection since the crown area was badly rotten and invariably yielded saprophytic fungi and bacteria (Plate 3). The lesions in such cases are generally dark and appear to be old. The failure to isolate the causal agent from such samples may be due to the fast growing secondary invaders which are generally species of *Fusarium* and bacteria. To avoid the contaminants, in 1970 out of the seven samples collected from coconut two were taken deeper down from the crown which was free from wet rot and showed only slight browning. *Phytophthora* sp. was isolated from one of these samples. Later successful isolations were made easily from samples collected from the basal part of the affected crown which was free from wet rot and hence of secondary organisms. In 1972 season, crowns of seven affected palms were cut at the base of the crown and were incubated at 24°C. Fluffy growth of *Phytophthora* sp. developed profusely from the cut end while the upper part of the crown affected by wet rot further rotted and yielded only bacteria and *Fusarium* sp. (Plate 4).

Incubation of the infected material suspended in tap water at 22°C. for 24 hr. before plating favoured isolation of pathogen by reducing contaminating organisms. Of the media used Glucose nitrate agar, amended with mycostatin (10000 units/l) and Pimaricin (10 ppm) and potato dextrose agar with 2 ppm thiamine HCl were found to be superior. The isolated cultures on inoculation on tender coconut leaflets yielded pure cultures of the pathogen.

Details of the isolation are shown in Table 9. The different isolates were tentatively identified as *P. palmivora* and designated as PA1 (Arecanut), PR1

(Rubber), PPl (Palmyra), POPl (Oil palm), PC1 to PC8 (Coconut) and PCd1 (Cardamom.) (Plate 5).

(b) ISOLATION FROM SOIL

Twelve soil samples collected from the base of bud rot affected coconut palms were screened for *Phytophthora* sp. by the dilution plate method using the selective medium of Flowers and Hendrix (1969), soil extract agar, oat meal agar and nutrient agar containing 10 ppm Pimaricin. None yielded the fungus.

II BIOLOGY

(a) CULTURAL STUDIES

PROCEDURE.

- i. Radial growth of two isolates of *P. palmivora* (PA1 and PR1) was studied when grown in glucose nitrate agar, coconut leaf extract agar and bean extract agar at 22°C, 24°C and 28-30°C (room temperature). One centimeter discs from 5 day old culture in glucose nitrate agar was used as inoculum. Average value of radial growth on 2 planes at 90° were recorded for 7 days from each of the four replicates.
- ii. Sporulation of the isolates PA1 and PPl grown in glucose nitrate agar and coconut leaf extract agar maintained at 18°C, 24°C and 27°C of temperature was also studied. Smears of media on microscope slides were inoculated with sporangial suspension having approximately 75 sporangia per drop and incubated. Newly developed sporangia were counted at the end of 3rd, 5th and 7th days. For each treatment average of counts from 5 low power microscope fields replicated 4 times were recorded.

RESULTS.

- i. Temperature of 28-30°C favoured best initial radial growth of the isolates PA1 and PR1 in glucose nitrate agar. They covered 9.5 cm growth within 5 days in coconut leaf extract agar. PR1 showed the same characteristic in bean extract agar (Table 10).
- ii. Studies on sporulation showed that PA1 preferred 24°C over 18°C in both glucose nitrate and coconut leaf extract agar media. Isolate PPl sporulated best at 18°C in glucose nitrate agar (Table 11).

The studies were not repeated and further work with other isolates were not carried out due to lack of facilities.

(b) INFECTION CYCLE

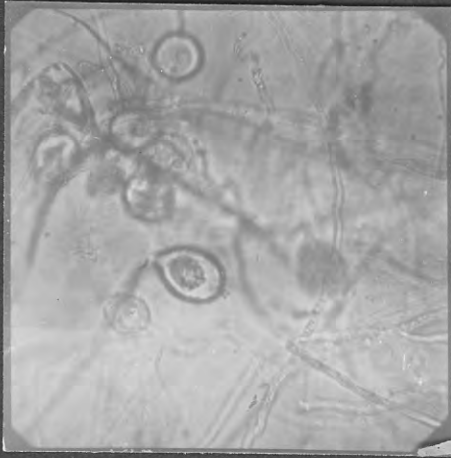
At 22-24°C the sporangia in water suspension germinated within 3 hours. On tender coconut leaves PC1 and POPl inoculated as sporangial suspension and incubated at 22-24°C and 100% relative humidity, established infection within 5 to 7 hours. Infection cycle of PC1 on cut bits of coconut petiole incubated similarly, was found to be 72 hours for primary infection, 48 hours for development of secondary sporangia and another 24 hours for secondary infection. Oospores appeared on these within 4 weeks.

PLATE 5

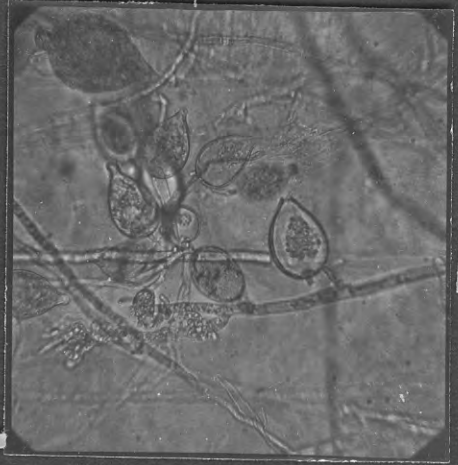
PLATE 6

- Fig. 1 Sporangia of arecanut isolate of *P. palmivora* (PA1) x 450.
- Fig. 2 Sporangia of rubber isolate of *P. palmivora* (PR1) x 450.
- Fig. 3 Sporangia of palmira isolate of *P. palmivora* (PA1) x 450.
- Fig. 4 Sporangia of oil palm isolate of *P. palmivora* (POP1) x 450.
- Fig. 5 Sporangia of coconut isolate of *P. palmivora* (PC1) x 450.
- Fig. 6 Sporangia of cardamom isolate of *P. palmivora* (PCd1) x 450.

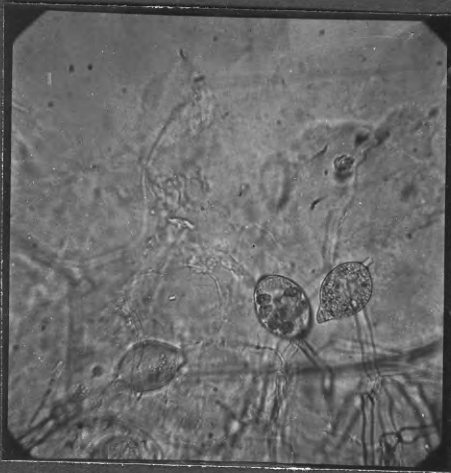
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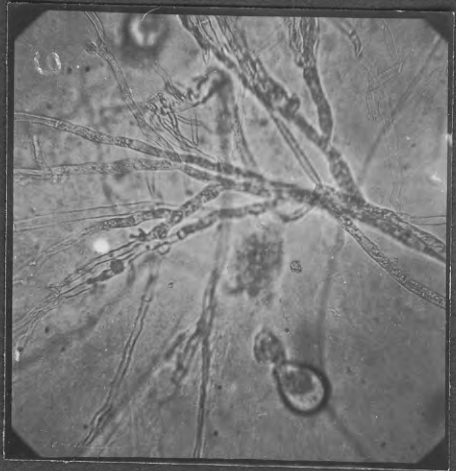
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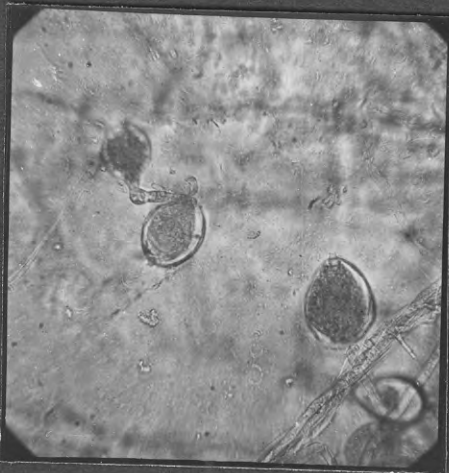
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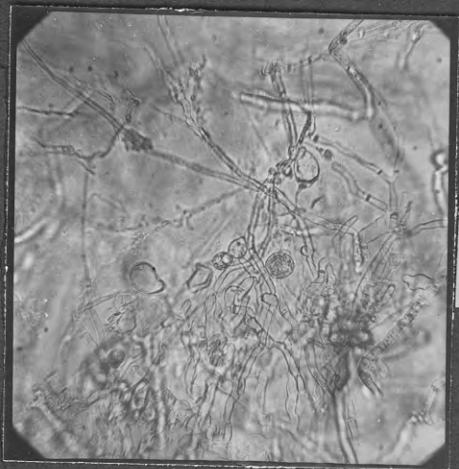
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9 - SURVIVAL

The pathogen was isolated from basal part of the crown from naturally infected coconut palms 3 months after the disease occurred in the field. The pathogen was reisolated from one coconut seedling inoculated with PA1 under field conditions after 6 weeks' weathering under field conditions.

Survival of *Phytophthora palmivora* (PC1) as mycelia in steamed soil and in infected soil was studied in soil was studied for a period of 8 weeks. Periodical samples of the soil and tissue revealed that the sporangia remained viable

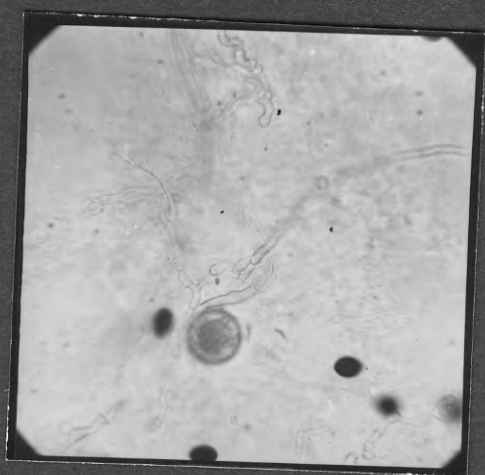
Survival of oospores was also observed in culture medium (glucose nitrate) after infected coconut tissue was plated and wherein *Thielaviopsis* was observed as a contaminant (Plate 6).

PLATE 6 PATHOGENICITY OF *Phytophthora palmivora*

Twigs of old seedlings of palm trees, coconuts, mangoes, etc. were tested under field condition and in the field 5 to 10 year old palms were used as test materials of *Thielaviopsis* sp., *Arctocarpus* spp., *Mungifera indica*, *Bougainvillea rosasinensis* were the test materials used as twigs and individual cut bits of coconut petioles and tender leaves were also used. Sporangia suspension from 3 to 4 day old culture or cork-borer discs from 7 day old cultures in agar medium and 20 day old culture in soil oats were used as inocula for *Phytophthora* sp.

3 to 4 day old *Phytophthora* sp. isolated from rubber was used as inoculum and when cultures were available isolates from other crops were used. Isolates of bacteria from the crown of bud rot affected coconuts were used. The inocula were introduced into the crown through the 3rd leaf axil in the palms in the field and through 1st or 2nd leaf axil in the laboratory.

Observations in the field were done during the South West monsoon. Observations were carried out in airconditioned rooms where the temperature was maintained between 22-24°C. Humidity was provided by spraying the plants with sterile water and covering them by alkathene bags, inside of which was sprayed with water. Twigs of rubber, mango etc. were kept in glass jars containing water and covered with alkathene bags while cut petioles and leaves were maintained in humid chambers.



(c) SURVIVAL

The pathogen was isolated from basal part of the crown from naturally infected coconut palm over 5 months after the disease occurred in the field. The pathogen was re-isolated from one coconut seedling inoculated with PA1 under controlled conditions after 8 weeks' weathering under field conditions.

Survival of *P. palmivora* (PC1) as mycelia in steamed soil and in infected petiole of coconut buried in soil was studied for a period of 8 weeks. Periodical examination of the soil and tissue revealed that the sporangia remained viable during this period.

Development of oospores was also observed in culture medium (glucose nitrate agar) 2 months after infected coconut tissue was plated and wherein *Thielaviopsis* sp. occurred as a contaminant (Plate 6).

III PATHOGENICITY

(a) PATHOGENICITY OF *Phytophthora palmivora*

PROCEDURE.

One to two year old seedlings of coconut, arecanut and oil palm were tested under controlled condition and in the field 5 to 10 year old palms were used as test plants. *Hevea* sp., *Artocarpus* spp., *Mangifera indica*, *Bougainvillea* sp. and *Hibiscus rosasinensis* were the test materials used as twigs and individual leaves. Similarly cut bits of coconut petioles and tender leaves were also used. Sporangial suspension from 3 to 4 day old culture or cork-borer discs from the gowing ends of cultures in agar medium and 20 day old culture in soil oats were the source of inocula for *Phytophthora* sp.

To begin with *Phytophthora* sp. isolated from rubber was used as inoculum. Later, as and when cultures were available isolates from other crops were made use of. Two isolates of bacteria from the crown of bud rot affected coconut were used. The inocula were introduced into the crown through the 3rd, 4th and 5th leaf axils in the palms in the field and through 1st or 2nd leaf axil in young seedlings.

The inoculations in the field were done during the South West monsoon. Other inoculations were carried out in airconditioned rooms where the temperature was maintained between 22-24°C. Humidity was provided by spraying the test material with sterile water and covering them by alkathene bags, inside of which was also sprayed with water. Twigs of rubber, mango etc. were kept in conical flasks containing water and covered with alkathene bags while cut petioles and leaves were maintained in humid chambers.

FIELD EXPERIMENTS

Pathogenicity tests were conducted in the field during S.W. monsoon in 1969, 1970, 1971 and 1972.

In 1969, 20 numbers of 5 year old coconut palms were inoculated with PPI grown in soil oats for 21 days. 95-97 per cent relative humidity and 25°C. of temperature prevailed at the time of inoculation. However during the post inoculation period relative humidity ranged from 82-89 per cent and temperature from 25.8 to 28.3°C. The seedlings neither produced lesions of infection nor symptoms of disease.

During 1970, one week after the onset of S.W. monsoon 5 year old coconut seedlings, 5 each per isolate were inoculated with PA1 and PR1 using sporangial suspension when relative humidity ranged from 95-96% and temperature from 23.4 to 26.7°C. Another set of 12 palms were inoculated with the infected material from the crown of affected palms. During the week following inoculation relative humidity ranged from 63 to 96 per cent and temperature 22.8 to 32.9°C.

Seedlings inoculated with PA1 and PR1 developed lesions on petioles but failed to produce disease symptoms excepting one which showed slight paling of the middle whorl of leaves. Palms inoculated with the infected material failed to produce infection. There was only a single instance of natural infection in the farm during this period.

Similar inoculation with the 5 isolates of *Phytophthora* sp. was carried out on 3 to 5 year old W. C. T. coconut seedlings during S.W. monsoon of 1971. No symptoms of infection/disease developed on the seedlings. The inoculations were done when the relative humidity was 97 per cent and temperature 23.3°C. which remained in the same range for two days after inoculation. Later the relative humidity ranged from 59 to 95 per cent and temperature 23.5 to 30.6°C while rainfall was meagre. A single case of natural infection occurred during the S.W. monsoon of this year at the farm.

Five year old hybrid coconut (Tall x dwarf) seedlings grown in pots were inoculated with coconut petioles artificially inoculated with PCI and naturally infected tissues, 12 seedlings each during the S.W. monsoon in 1972. Although relative humidity ranged from 97 to 98 per cent at the time of inoculation it varied from 66 to 96% during the subsequent period with rainfall less than 10 mm on most of the days. Temperature ranged from 22.3 to 32.0°C. None of the inoculated palms took infection. Disease incidence was also nil in the farm. However in the backwater area, nearly 5 KM away from the farm 5 to 10 years old palms growing in low lying gardens along the coast were found to be affected 5 weeks after the onset of S.W. monsoon.

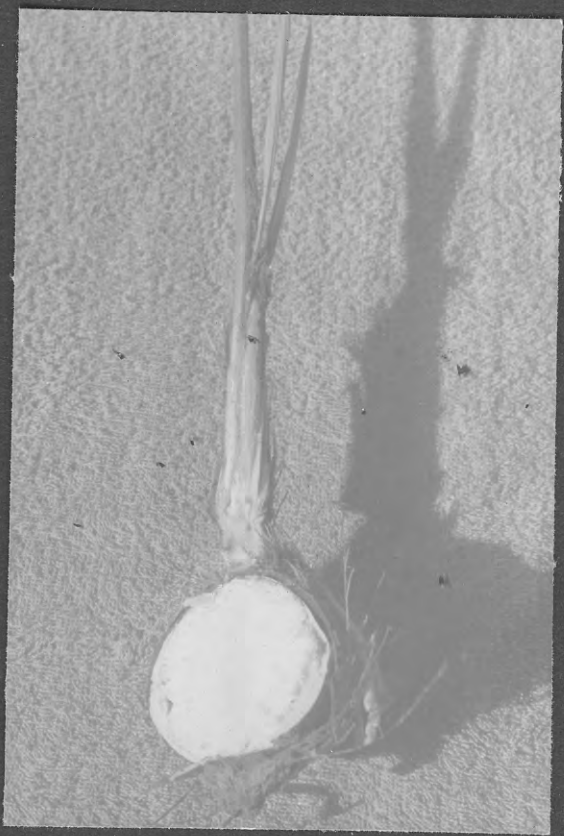
PLATE 7

- Fig. 1 Potted W.C.T. seedling showing brownish lesions on the spear leaf and petiole. The seedling was artificially inoculated with sporangial suspension of *P. palmivora*.
- Fig. 2 Longitudinal section of the seedling which succumbed to artificial inoculation with *P. palmivora*. Note the brownish lesions distributed among the tissues of the tiny crown.



1

2



UNDER CONTROLLED CONDITIONS.

i. ON COCONUT (WCT)

Two year old coconut seedlings growing in pots were inoculated with sporangial suspension of PA1, PR1 and *P. meadii*-2 seedlings each and were maintained at 22-24°C (air conditioned room, Rubber Research Institute of India, Kottayam). Two seedlings were also inoculated with bacteria isolated from bud rot affected tissue.

Of the three *Phytophthora* cultures *P. meadii* had larger number of sporangia than PA1 and PR1 in unit volume, the minimum being in PA1. One of the seedlings inoculated with *P. meadii* developed water soaked lesions typical of *Phytophthora* infection after 3 days' incubation. Two weeks later the seedling was split open to check up the progress of infection. The lesions had turned to brown spots but rotting had not reached the bud. One of the seedlings inoculated with PR1 also developed similar symptoms ten days after inoculation.

Further trials were carried out in an air-conditioned room at Kayangulam on batches of 20 or 12 seedlings. Results are summarised below:-

One year old coconut seedlings growing in pots were inoculated with PA1, PR1, PPI, PCI and POPI seven to 10 seedlings per isolate. All the isolates established infection and produced lesions on the spear leaf petiole and leaf sheath within two weeks after inoculation (Plate 7). Majority of the seedlings (Table 2) succumbed to the infection. Incubation period varied from 10 weeks to 36 weeks except in one instance where the seedling died in 5 weeks after inoculation. Pathogens were reisolated from all the infected seedlings.

ii. INFECTION TRIALS ON COCONUT VARIETIES AND HYBRIDS

One year old coconut seedlings, Chowghat dwarf and Malayan dwarf and hybrids from Tall, Dwarf and Gangabondam were inoculated with PCI. Chowghat dwarf plant 5 each tested twice withered within a month without producing any symptoms of infection. Malayan dwarf and the hybrids produced the symptoms as in West Cost Tall, the Malayan dwarf being less susceptible than the others. Incubation period was considerably shorter in all except in the hybrids where the female parent was West Cost Tall (Table 13).

iii. ON ARECANUT AND OIL PALM

All the five isolates of *Phytophthora* infected one year old arecanut (west cost ordinary) and oil palm (malayan seedlings). The arecanut plants withered before the infection penetrated to the bud region while a few of the oil palm seedlings succumbed to the infection (Table 14).

iv. OTHER HOSTS

Mangifera indica, *Artocarpus hirsuta*, *A. integrifolia*, *Hibiscus rosasinensis*, *Bougainvillea* sp. and *Havea brassiliensis* were inoculated at the leaf axils on twigs with all the five isolates. Infection was established on all hosts other than *H. rosasinensis* within 48 hr. after inoculation and the leaves withered and dropped in a weeks time.

(b) PATHOGENICITY OF BACTERIA

In the light of the suggestions of Dr. D. N. Srivastava, Asst. Director-General. ICAR the possible role of bacteria in the incidence of bud rot was tested.

i. Two unidentified cultures of bacteria isolated from bud rot affected palm were tested in the same manner as *P. palmivora* for their capacity to infect coconut seedlings under controlled conditions of temperature and humidity. Two sets of 2 and 3 seedlings were tested for each bacterial culture. No symptoms of infection were observed.

ii. Two sets of tender coconut petiole pieces were inoculated with species of *Pseudomonas*, *Xanthomonas* and *Erwinia* isolated from coconut, one healthy tissue and another artificially infected with PCI and were maintained in the air conditioned room. The bacterial cultures failed to infect healthy coconut tissue but hastened rotting and secondary infection when initially infected with PCI (Plat 8).

iii. Under field conditions twelve 5 year old coconut seedlings growing in pots inoculated with *P. palmivora* (PCI) during the S. W. monsoon in 1972 and which failed to produce disease symptoms were later inoculated with the two cultures of bacteria. No symptoms of bud rot disease developed even after three months.

FUNGICIDAL TRIALS

EXPERIMENT 1.

PROCEDURE:

Ten to fifteen year old palms were inoculated with soil oats inoculum of PR1 100 g. each during the pre-monsoon period (May) in the 3rd, 4th and 5th leaf axils. Two weeks later five palms each were treated with Bordeaux paste, Phenyl mercury acetate and chloride and Dithane M-45 (0.3%). The fungicides were applied in the inoculated leaf axils. Five palms were maintained as inoculated untreated control.

PLATE 8

- 1 Bits of tender petiole of coconut inoculated with three isolates of bacteria and *P. palmivora*. (L to R) First two bits in each of the three groups are inoculated with *Pseudomonas* sp., *Xanthomonas* sp. and *Erwinia* sp. respectively. The 3rd bit in each group was first inoculated with the sporangial suspension of *P. palmivora* followed by the respective bacterial isolate. The small spots seen on the bits inoculated with the bacterial isolates alone are only deposits from the inoculum. Lesions are formed on bit 2, which was primarily infected by *P. palmivora* and the lesions hastened further rotting.
- 2 Bits inoculated with *P. palmivora* for 24 hours. The other petioles below the bit are uninoculated.



of the palms including control developed any symptoms of disease and the effect of fungicidal treatment could not be assessed.

EXPERIMENT 2.

These fungicidal treatments were given to five year old palms during the early

Fungicidal treatments:-

- | | |
|-----------------------------|------------------------|
| (a) Phenyl mercury acetate | --0.3% |
| (b) Phenyl mercury chloride | --0.3% |
| (c) Dithiazon M.45 | --0.3% |
| (d) Aureofungin | 9--4 : 4 : 4 |
| (e) Demosan | --10 ppm. |
| (f) Control | --inoculated untreated |

Palms infected by leaf spot showing lesions on the main stem and were inoculated with the initial site of infection and (PPI) and the other two were inoculated with (PPI) and (PPI). Ten ml of sporangial suspension of the pathogens were applied to the petiole base of a palm which has been treated with PPI after 2 weeks. The results showed that the site of infection in the control was the oldest petiole and the first 2 the oldest petiole below the main stem. Lesions of *Phytophthora* infection were observed on the younger and two older ones besides the inoculated, and the pathogen was not isolated in spite of the fungicidal treatment (Plate 9). However the pathogen was not reisolated.

The inoculated controls developed any symptom.

The effect of the fungicides could not be assessed due to the failure of the development of the disease further experiments were carried out in the laboratory using detached palm leaves. The pathogens tested were PAI, PCI, PPI, POPI and (PPI). Twenty cork borer discs from the periphery of the leaflets were used for inoculation. The leaflets were kept at 95 per cent humidity. Phenyl mercury acetate, Phenyl mercury chloride and Dithiazon all at 0.3 per cent concentration, Aureofungin at 10 ppm, Demosan at 1.0 per cent and Dithiazon at 1200 ppm were applied to the leaflets prior to inoculation.



1



2

RESULT:

None of the palms including control developed any symptoms of disease and hence efficacy of fungicidal treatment could not be assessed.

EXPERIMENT 2.

PROCEDURE:

The following treatments were given to five year old palms during the early morning period.

1. Fungicidal treatments:-

- (a) Phenyl mercury acetate —0.3%
- (b) Phenylmercury chloride —0.3%
- (c) Dithane M-45 —0.3%
- (d) Bordeaux paste —4 : 4 : 4
- (e) Aureofungin — 10 ppm.

2. Control

—inoculated untreated

The treatments were in two series, in the first fungicidal treatments were given two weeks prior to fungal inoculation with PA1 and PR1 and the other two weeks after inoculation. Ten ml of sporangial suspension of the pathogens were injected into the 3rd, 4th and 5th leaf axils. Palms were reinoculated after 4 months. Four months after inoculation yellowing of the young leaves was observed in one palm inoculated with PR1 and treated with Phenyl mercury acetate. On opening the crown lesions of *Phytophthora* infection were observed on 7 leaf bases two younger and two older ones besides the inoculated, indicating the spread of the pathogen in spite of the fungicidal treatment given eight days after inoculation. (Plate.9). However the pathogen was not reisolated.

None of the untreated inoculated controls developed any symptom.

EXPERIMENT 3

In the former two trials the effect of the fungicides could not be assessed in the experiments due to the failure of the development of the disease further to screen the fungicides were carried out in the laboratory using detached leaflets and petioles. The pathogens tested were PA1, PC1, PP1, PO1 and ~~isolated from cardamom~~). Twenty cork borer discs from the periphery of the colonies of each isolate were used for inoculation. The leaflets were incubated at 24°C and 100 per cent humidity. Phenyl mercury acetate, Phenylmercury chloride, Thiram and Ziram all at 0.3 per cent concentration, Aureofungin at 100 and 1000 ppm, Difolatan at 1.0 per cent and Demosan at 1200 ppm were sprayed on the leaflets prior to inoculation.

RESULTS.

Observations on infection as indicated by the development of lesions were recorded after 72 hr (Table 15). Phenyl mercury chloride and acetate induced phytotoxic effects at the level tested.

EXPERIMENT 4

In view of the efficacy of Demosan observed in the previous trial it was further tested using coconut petioles as test material. Demosan at 1500 ppm completely inhibited infection of PCI.

EXPERIMENT 5

Efficacy of Demosan, Thiram, Ziride and Difolatan in checking infection by cardamom isolate of *P. palmivora* was next tested. Again Demosan at 1500 ppm proved to be the best treatment, followed by Thiram, Demosan at 1200 ppm, Difolatan and Ziride.

RESIDUAL EFFECT OF FUNGICIDES AND AN ANTIBIOTIC

Efficacy of Bordeaux paste, Demosan 1200 ppm, Thiram 1200 ppm, Ziride 1200 ppm and Aureofungin 1000 ppm in preventing infection of *P. palmivora* was tested on 5 year old seedlings 4 each per treatment. The fungicides were applied into the three inner leaf axils at the rate of 150 ml. One week and three weeks after the treatment the treated petioles from one seedling each were removed and infectivity of PCI was tested on these. The petioles were cut into 4 pieces from the bottom upwards and inoculated at 4 sites on each piece. They were then incubated at 22-24°C. Infection was established in the two basal cut pieces of all the petioles and also in some others.

As suggested by Dr. Webb, USDA, who visited us twice during the course of the project in 1970 and 1972 efficacy of Benlate as a protectant was tested when the material became available. Since two years neither natural incidence nor development of the disease on artificial inoculation occurred in the field at the Research Station farm the following method was adopted for the trial. Benlate at concentrations of 100 ppm, 500 ppm and 1500 ppm were poured into the three inner leaf axils of 3 seedlings (5 years old) each at the rate of 150 ml. 8 days after the treatment the petioles which received the fungicide were removed, cut into 4 pieces from the base upwards and inoculated with sporangial suspension (10 drops containing 25 to 30 sporangia) of PCI at 4 sites. The inoculated materials were incubated at 24°C. Infection was established in all the cases and profuse growth of the pathogen was observed from the lesion.

DISCUSSION

The controversial problem of bud rot disease of palms was discussed and the identity of the causal agent as *Phytophthora palmivora* (Butl.) was accepted at international level in 1924. In India following the report of Butler in 1906 on the bud rot disease of palms caused by *P. palmivora* Shaw and Sundararaman (1914), McRae (1923) and Sundararaman (1924) proved the pathogenicity of the same on coconut, arecanut and palmyra palms. Yet the unsuccessful efforts in many instances to isolate the pathogen from coconut and the appearance of disease syndrome at the detectable stage as a wet rot still cast doubt on the role of *P. palmivora* as the causal agent. The possibility of pathogens other than *P. palmivora* particularly bacteria having a positive role in the incidence of disease was also suggested.

Observations in early stages of infection on artificially inoculated tissues and of samples collected from the basal part of naturally infected palms revealed that the rotting is dry and not wet. Isolation made from the latter material yielded *P. palmivora* in all instances whereas the tissues affected by wet rot in the later stages harboured mainly bacteria and *Fusarium* sp. It is likely that infection of *P. palmivora* causes only dry rot in coconut as in 'mahali' of areca and leaf fall of rubber and that the wet rot observed in the bud rot disease of coconut is probably due to the secondary invasion of bacteria attracted by the high carbohydrate content of the crown. Reinking (1919 and 1923) reported from Philippines that *P. faberi* (Maubl.) is the primary cause of bud rot of coconut with bacteria occurring as a secondary infection in the injured and weakened tissue. Results of our inoculation trials with three bacterial cultures also suggest a similar situation. The fact that the rotting of coconut petioles initiated by *P. palmivora* is aggravated by the inoculation of bacteria probably indicate that the secondary invaders contribute towards the final collapse of the host.

The occurrence of *Phytophthora* disease have been reported to be highly influenced by weather factors, the classical example being that of *P. infestans* on potato. Bud rot disease is season bound and occurs during the monsoon months and may be classified among the rain favoured diseases, the out breaks of which are well correlated with periods of rain according to Keitt and Jones (1926) Butler (1910) and Menon and Pandalai (1958) reported that bud rot disease of coconut is favoured by high atmospheric humidity. They also observed that the disease is mainly confined to young palms below 25 years. Our observations carried out in the different coconut growing States in India confirm this (Table I) The importance of climatic conditions favourable to the pathogen and the susceptible age of palms in the occurrence of bud rot disease are evident from the non-prevalence of the disease in the adult plantations of Goa, Maharashtra and

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in young plantations in Saurashtra. Weather conditions congenial to the incidence of disease occur in Goa and Maharashtra while in Saurashtra they are non-existent.

Similarly the occurrence of the disease at Kayangulam and Kasaragod reveals close correlation with weather factors. Occurrence of the disease in backwater areas - lowlying and waterlogged - about 5 KM away from the Kayamkulam farm during the periods when disease is absent in the farm may perhaps be attributed to the atmospheric conditions, particularly that of low temperature and high humidity prevailing there due to environmental conditions. Crosier (1934) has shown that field inoculations with swarm spores of *P. infestans* "in the morning following heavy deposition of dew resulted in infection on only those few occasions when the foliage remained wet for a period of 3 to 4 hr after the plants had been inoculated". Krause and Massie (1973) have reported similar situation in the case of potato blight - that microclimatological conditions at stations separated by less than six miles are drastically different for blight forecast.

Data collected on the microclimate of palms of different ages indicate that it is directly related to weather factors particularly that of rainfall. Humidity in the range of 97-100% and temperature 21.0°C and the duration of such conditions - referred to as "favourable days" - appear to influence the development of the disease (Table 3). These observations are in line with that of Crosier (1934) and Kyron (1967) in the case of potato blight and of Hicks (1967) for pod rot of cocoa caused by *Phytophthora* spp. The critical studies of Crosier (1934) reveal that humidity and temperature are the two cardinal factors that determine sporulation, germination and infection of *P. infestans*. The factors favouring successive development of sporangia and infection of the pathogen are closely inter-related and these determine the onset of the disease. Laboratory trials showed that *P. palmivora* requires nearly a week from the time of inoculation as sporangial suspension to complete one cycle i.e. germination, infection and production of secondary sporangia. A series of such cycles are required to enable the pathogen to penetrate the thickly set whorles of leaves and reach the bud region from the foci of infection for the onset of the disease. Field observations have revealed the occurrence of the disease in a minimum period of 5 weeks after inoculation under controlled conditions and in the field after the onset of S.W. monsoon (Table 5 a and b).

All the isolates of *P. palmivora* tested established infection when inoculated on the central spindle of one and two year old seedlings grown in pots, under controlled conditions of temperature (24°C) and humidity (98-100%). The infection spread to the growing point and ultimately caused death of the plant (Plate 6). Under field conditions inoculation of 5-7 year old palms also caused infection, which penetrated to the upper and lower parts of the crown from the point of inoculation (Plate 9), however, failed to cause the disease. It is to be pointed

out that during this period (S.W. monsoon of 1970, '71 and '72) the natural incidence of the disease in the farm where the experiment was carried out was almost nil, the reason for which is sought in the lack of favourable microclimatic conditions.

Positive results of cross inoculations with the different isolates is in conformity with that of earlier workers (Reinking, 1923; Ramakrishnan and Seethalexmi, 1956).

Formation of oospores in single culture in synthetic media when contaminated with *Thielaviopsis* sp. has its parallel in the report of Brasier (1971, '72) on the inducement of oospore production by *P. palmivora* by *Trichoderma viride*. Oospore development was also observed in artificially inoculated and infected plant tissue. Here again the involvement of secondary invaders and probable chemical stimulation as suggested by Brasier of bisexual nature of the organism is possible, which is yet to be worked out. Whatever be the source of inducement for the sexual stage the oospore can serve as potential inoculum during the favourable period provided they are germinable.

The importance of prophylactic method of control or preventive fungicidal treatment, although well aware of, was not possible to work out due to various reasons. The limited trials conducted under laboratory conditions indicate that organic formulations like Demosan may prove effective but are to be tested out in the field.

CONCLUSIONS

Occurrence of bud rot disease of coconut is directly related to the microclimate of the palms, relative humidity and temperature in the leaf axils. The microclimate of young palms, 5 to 20 years old is more favourable to the incidence of disease.

Phytophthora palmivora (Butl.) causing bud rot of coconut, palmyra and oil palm, fruit rot of arecanut and leaf fall of rubber are plastic in pathogenicity. An infection cycle on coconut tissue is completed in 6 days under favourable conditions of temperature (22-24°C) and relative humidity (98-100%). The pathogen survives in infected tissue for over 5 months. Oospore production takes place in 4 to 6 weeks both in culture medium and infected material. *P. palmivora* causes dry rot of coconut crown. The wet rot observed in the late stages of the disease is probably due to the activity of secondary invaders like species of *Pseudomonas*, *Xanthomonas* and *Erwinia*.

NEED FOR ADDITIONAL RESEARCH

The scattered occurrence of the disease and its mode of spread need further investigation.

Since the glass house proposed under the project for the conduct of pathogenicity trials was not available the work was restricted to young seedlings in an air conditioned room.

TABLE 1. Disease incidence in Kerala and Tamil Nadu.

State	District	No. of village/plots surveyed	No. of palms observed	Percentage of disease incidence in palms		
				Below 10 yrs	10-30 years	Above 30 years
Kerala	Cannanore	57/252	15857	0.77	1.94	0.36
	Kozhikode	42/151	10170	1.66	1.32	0.26
	Malappuram	41/205	27205	1.10	0.10
	Palghat	41/205	20250	2.10	0.10
	Trichur	46/230	22528	2.10	0.20
	Ernakulam	46/240	23627	2.00	0.10
	Kottayam	27/104	5177	0.53	1.71
	Alleppey	42/214	31903	6.50	4.40
	Quilon	38/158	19647	5.02	5.66
	Trivandrum	57/294	35052	4.50	3.80
Tamil Nadu	Kanyakumari	41/134	33193	1.60	2.90 2.5
	Thirunelveli	18/27	2379	6.70	4.20 11.1
	Trichinopoly	4/6	868	5.60	0.40 6
	Ramanathapuram	11/28	4050	1.80	1.10 2.9
	Thanjavur	52/115	11164	5.90	1.50 7.4
	South Arcot	18/42	2919	4.50	2.10 6.6

TABLE 2. Rainfall and disease incidence during 1968 and 1969 at Kayangulam, Kasaragod and Ratnagiri recorded from May to August.

	Kayangulam		Kasaragod		Ratnagiri	
	1968	1969	1968	1969	1968	1969
a) Rainfall in mm	1954.9	1118.3	3560.9	2052.5	2100.00	3400.00
b) No. of rainy days	68	55	72	61	—	—
c) * No. of favourable days	84	66	41	33	—	—
d) Disease %	1.8	0.5	1.24	0.1	0.4	1.0

* Favourable days with Relative Humidity between 95-100% in Stevenson screen.

TABLE 3. Meteorological factors and microclimate. May to September.—Kayangulam.

	1968	1969	1970	1971	1972
Rainfall in mm	2234.0	1502.1	1379.2	1619.3	1021.7
No. of rainy days	89	81	77	63	56
No. of favourable days	13.20@	3.6@	3.4@	7.13	**
Disease incidence	1.6-5.4	0.3-1.2	Trace	nil	nil

@ Leaf axil at 9.00 hrs.

** R. H. (Maximum 95-97% except on one day (99%).

TABLE 4. Micro-climate (RH) and disease incidence in relation to age of palms at Kayamkulam—1968-'69.
June-August.

Age of palms	1968		1969	
	No. of favourable days	% disease	No. of favourable days	% disease
Below 5 years	13	0	3	0
6-10 years	16	1.6	6	1.0
11-15 years	20	3.9	3	1.2
16-20 years	18	5.4	4	0.3
21 years & above	—	—	4	0

TABLE 5a Micro-climate and disease incidence in relation to age of palms 1968.

Period in weeks	Rain fall in mm	Age of palms								
		Below 5 yrs.		5-10 yrs.		11-15 yrs.		16-20 yrs.		
		a	b	a	b	a	b	a	b	
Pre-monsoon										
May-4th week	33.5	0	0	0	0	0	0	0	0	
June	1	81.7	1	0	1	0	1	0	0	0
	2	307.6	1	0	3	0	3	0	3	0
	3	75.4	2	0	3	0	2	0	3	0
	4	229.8	1	0	0	0	3	0	2	0
Monsoon July	1	338.9	2	0	2	0	3	1.4	2	0
	2	28.6	0	0	0	0	0	0	0	2.7
	3	328.4	3	0	3	0.8	4	1.5	4	0
	4	86.6	2	0	2	0	2	0	2	2.3
August	1	51.4	0	0	1	0.4	1	1.0	0	1.0
	2	18.3	1	0	1	0.3	1	0	2	1.0
Sept.	1	5.2	0	0	0	0	0	0	0	0
	2	86.8	0	0	0	0	0	0	0	0

a — Favourable days — RH. 95-100

b — Percent disease incidence

TABLE 5 b. Details of the microclimate during the critical period — 1969.

Period	Age of palms									
	Below 5 yrs		5-10 yrs		11-15 yrs		16-20 yrs		21 yrs and above	
	a	b	a	b	a	b	a	b	a	b
14-5-1969	100	25.1	100	25.0	100	25.0	100	25.0	97	25.5
15-5-1969	97	25.5	100	25.0	97	25.5	93	26.5	93	28.0
31-5-1969	95	25.0	93	26.0	93	26.0	94	26.0	94	26.1
*2-6-1969	93	28.0	99	27.2	95	28.0	92	28.2	93	28.5
23-7-1969	93	25.0	97	25.0	93	25.0	97	24.5	95	24.2
24-7-1969	93	25.0	97	24.5	97	25.5	97	24.5	97	24.0

a - R. H.

b - Temperature

* Disease incidence was observed during the first week of June.

Table 6 a. Weather factors and microclimate with disease incidence at the two centres in Kerala — 1970-71.

	Kayangulam	Kasaragod
Rainfall in mm.		
(S.W. monsoon)	1379.2	3567.7
No. of rainy days	77	89
Minimum temp. 0°C	22.5-24.8	21.1-21.6
Maximum temp. 0°C	31.3-33.3	29.6-33.6
Favourable days *	4	15
No. of palms diseased	1	24

* days of high humidity (95-98%) & low temperature (23.0 - 26.0°C) in microclimate at the time of observation (9.30 hr)

TABLE 6 b. Disease incidence and microclimate in relation to age of palms. 1970-71.

Age of palms	Kayangulam		Kasaragod	
	Favourable days	Disease incidence No. of palms	favourable days	Disease incidence No. of palms
3-5 yr.	3	0	15	0
6-10 yr.	4	1	13	13
11-15 yr.	4	0	15	2
16-20 yr.	3	0	13	4
above 20 yr.	4	0	11	5

TABLE 7 a. Analysis of variance - Av. Dry bulb °c

Age group	Kasaragod		Kayangulam	
	Leaf axil	Open air	Leaf axil	Open air
A - 1-5 yr.	25.30	25.29	25.52	25.51
B - 6-10 yr.	25.28	25.27	25.63	25.61
C - 11-15 yr.	25.35	25.32	25.67	25.64
D - 16-20 yr.	25.37	25.37	25.81	25.78
E - Above 20 yr.	25.42	25.45	25.77	25.72
Mean	25.34	25.34	25.68	25.65
S.E.	0.021	0.027	0.022	0.020
C.D.	0.06	0.08	0.06	0.06
Significance	Yes at 1%		Yes at 1%	
Conclusion	<u>E D C A B</u>	<u>E D C A B</u>	<u>D E, C, B A</u>	<u>D E, C B, A</u>

TABLE 7 b. Analysis of variance - Av. Humidity %

Age group	Kasaragod		Kayangulam	
	Leaf axil	Open air	Leaf axil	Open air
A - 1-5 yr.	94.67	94.56	90.84	90.60
B - 6-10 yr.	94.40	94.35	90.82	90.54
C - 11-15 yr.	94.09	93.94	90.66	90.59
D - 16-20 yr.	93.63	93.45	90.86	90.36
E - Above 20 yr.	93.55	93.44	90.63	90.25
Mean	94.07	93.95	90.76	90.47
S.E.	0.125	0.138	0.118	0.120
C.D.	0.35	0.38	—	—
Significance	Yes at 1%	Yes at 1%	No	No
Conclusion	<u>A B C</u> , <u>D E</u>	<u>A B</u> , <u>C</u> , <u>D E</u>		

TABLE 8. Microclimatic observation at Kayangulam-1972-Analysis of variance.

Age group	Humidity percentage		Dry blue °C	
	Leaf axil	Open air	Leaf axil	Open air
A - 1-5 yr.	90.23	90.10	26.44	26.47
B - 6-10 yr.	90.23	90.03	26.37	26.35
C - 11-15 yr.	90.32	90.00	26.29	26.31
D - 16-20 yr.	90.19	89.84	26.34	26.37
E - Above 20 yr.	90.00	89.45	26.39	26.43
Mean	90.19	89.88	26.37	26.39
S. E./Mean	0.109	0.098	0.019	0.020
C D	—	0.27	0.06	0.06
Significance	N. S.	H. S.	H. S.	H. S.
Conclusion		<u>A B C D</u> , <u>E</u>	<u>A E B D C</u>	<u>A E D B C</u>

Year	Source of material used	No. of isolations made	No. of successful isolations	Culture medium used	Antibiotic and promoters used																																																																											
1968-69	Coconut - natural incidence	23	Nil	Oats agar, coconut cabbage extract agar, papaya fruit extract agar, bean agar, soil oats agar, PDA + thiamine HCl 2 ppm.	Pimaricin 50 ppm, Polymixin 100 & Mycostatin 100 ppm, Neomycin 100 ppm, Gallic acid 0.001 M.																																																																											
	Rubber - secondary leaf fall	15	Nil			1969-70	Coconut - natural incidence	9	Nil	Glucose nitrate, soil oats agar, Bean agar, PDA + thiamine HCl 2 ppm.	Mycostatin 1000 ppm, Neomycin 100 ppm, Polymixin 200 ppm, Pimaricin 1000 ppm, Cholestrol 25 mg/dish.	Arecanut - nutfall	5	1	Rubber - secondary leaf fall	7	Nil	<i>Artocarpus hirsuta</i> - leaf spot	10	2	<i>A. integrifolia</i> - leaf spot	6	2	<i>Hibiscus rosa sinensis</i> - leaf spot	7	Nil	<i>Bougainvillea</i> sp. - leaf spot	12	2	<i>Mangifera indica</i> - leaf spot	8	2	1970-71	Coconut - natural incidence	14	1	Glucose nitrate, Nutrient agar + 10 ppm Pimaricin, Oat meal agar + 10 ppm Pimaricin.	— do —	Coconut - artificially infected	13	10	Oil palm - natural incidence	1	1	Rubber - Secondary leaf fall	5	1	Areca seedling - natural incidence	1	Nil	Palmyra - natural incidence	1	1	1971-72	Coconut - natural incidence	2	Nil	Glucose nitrate.	— do —	Coconut - artificially infected	8	7	Rubber - Secondary leaf fall	9	1	Arecanut - nut fall	4	1	Cardamom - azhukal	3	1	Arecanut - artificially infected	20	11	Oil palm - artificially infected	2	2	1972-73	Coconut - naturally infected	7
1969-70	Coconut - natural incidence	9	Nil	Glucose nitrate, soil oats agar, Bean agar, PDA + thiamine HCl 2 ppm.	Mycostatin 1000 ppm, Neomycin 100 ppm, Polymixin 200 ppm, Pimaricin 1000 ppm, Cholestrol 25 mg/dish.																																																																											
	Arecanut - nutfall	5	1																																																																													
	Rubber - secondary leaf fall	7	Nil																																																																													
	<i>Artocarpus hirsuta</i> - leaf spot	10	2																																																																													
	<i>A. integrifolia</i> - leaf spot	6	2																																																																													
	<i>Hibiscus rosa sinensis</i> - leaf spot	7	Nil																																																																													
	<i>Bougainvillea</i> sp. - leaf spot	12	2																																																																													
	<i>Mangifera indica</i> - leaf spot	8	2																																																																													
1970-71	Coconut - natural incidence	14	1	Glucose nitrate, Nutrient agar + 10 ppm Pimaricin, Oat meal agar + 10 ppm Pimaricin.	— do —																																																																											
	Coconut - artificially infected	13	10																																																																													
	Oil palm - natural incidence	1	1																																																																													
	Rubber - Secondary leaf fall	5	1																																																																													
	Areca seedling - natural incidence	1	Nil																																																																													
	Palmyra - natural incidence	1	1																																																																													
1971-72	Coconut - natural incidence	2	Nil	Glucose nitrate.	— do —																																																																											
	Coconut - artificially infected	8	7																																																																													
	Rubber - Secondary leaf fall	9	1																																																																													
	Arecanut - nut fall	4	1																																																																													
	Cardamom - azhukal	3	1																																																																													
	Arecanut - artificially infected	20	11																																																																													
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1972-73	Coconut - naturally infected	7	7	— do —	— do —																																																																											

TABLE 10. Effect of culture medium and temperature on radial growth of *P. palmivora* isolates PA1 and PR1. Average in centimeters recorded at the end of 3rd, 5th and 7th days.

Isolate of <i>Phytophthora</i> used	Tempera- ture in °C-	Glucose nitrate medium			Coconut leaf extract medium			Bean agar medium		
		3rd day	5th day	7th day	3rd day	5th day	7th day	3rd day	5th day	7th day
PA1	22	4.03	6.12	9.50	5.22	6.62	8.02	4.00	6.38	8.18
PR1	22	3.15	5.75	7.05	4.80	6.51	7.50	4.71	7.48	*
PA1	24	4.25	6.75	9.50	5.25	7.40	*	3.60	5.80	6.73
PR1	24	4.14	6.00	9.50	5.17	7.21	8.25	4.41	6.63	8.31
PA1	28-30	5.00	8.04	9.50	6.20	9.50	—	4.45	6.38	8.19
PR1	28-30	4.51	6.00	9.50	6.13	9.50	—	5.56	9.50	—

* Completed growth of 9.50 cm on the 6th day.

TABLE 11. Effect of culture medium and temperature on sporulation of *Phytophthora palmivora* isolates PA1 and PP1.

Isolate of <i>Phytophthora</i> used.	Culture medium	Tempera- ture in °C.	Average number of sporangia present per low-power field microscope at the end of		
			3rd day	5th day	7th day
PA1,	Glucose nitrate	18	Nil	10	10
PP1	—do—	-do-	5	13	51
PA1	Coconut leaf extract	-do-	1	8	3
PP1	—do—	-do-	5	16	24
PA1	Glucose nitrate	24	6	25	50
PP1	—do—	-do-	1	9	33
PA1	Coconut leaf extract	-do-	Nil	1	48
PP1	—do—	-do-	Nil	1	14
PA1	Glucose nitrate	27	10	15	22
PP1	—do—	-do-	3	19	15
PA1	Coconut leaf extract	-do-	Nil	11	17
PP1	—do—	-do-	Nil	2	3

TABLE 12. Results of pathogenicity test with *Phytophthora* isolates on coconut seedlings - West Coast Tall.

Isolate	No. of seedlings		Incubation period in weeks	Reisolation of pathogen + / -	Nature of symptom
	inoculated	infected			
PA1	7	6	5-24	+	Lesion on spear leaf, initially and later on petiole and leafsheath - infection penetrates to the bud causing rotting of the tissues and results in death of the plant.
PR1	6	4	24-26	+	
POP1	10	6	12-28	+	
PPI	10	8	10-23	+	
PCI	10	6	15-36	+	

TABLE 13. Results of pathogenicity tests on coconut hybrids.

Isolate	Test plant	No. of plants inoculated	No. Infected	Incubation period in weeks	Reisolation of pathogen + / -
PCI	Tall x Dwarf	5	3	21-37	+
„	Dwarf x Tall	15	10	7-15	+
„	Tall x Ganga-bondam	14	7	7-19	+
„	Dwarf x Ganga-bondam	5	4	6-18	+
„	Malayan dwarf	10	3	12-15	+

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