

phytophthora diseases

OF TROPICAL CULTIVATED PLANTS



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CENTRAL PLANTATION CROPS RESEARCH INSTITUTE
KASARAGOD 670 124, Kerala, India.

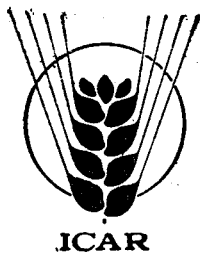
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**PROCEEDINGS OF THE
WORKSHOP ON
PHYTOPHTHORA DISEASES
OF TROPICAL CULTIVATED PLANTS
19-23 SEPTEMBER, 1980**

EDITED BY:

K. K. N. NAMBIAR

Head, Division of Plant Pathology
CPCRI, Kasaragod



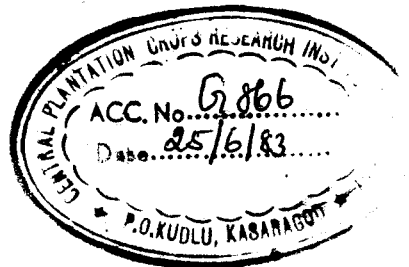
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FOREWORD

The genus *Phytophthora* established in 1876 by de Bary is known to infect an array of plants—trees, shrubs and herbs, both annuals and perennials, wild and cultivated, belonging to many families of flowering plants and conifers. Several of them are economically important in the tropics, subtropics and temperate regions. In India, many plantation crops are affected by this deadly pathogen. The foot rot of black pepper, *Koleroga* of arecanut, leaf fall of rubber, bud rot of coconut etc. are some of the major diseases of plantation crops limiting their production. Though research on *Phytophthora* was initiated about a century ago, not much information is available on many of the *Phytophthora* diseases, particularly in the tropics, on their infection processes, pattern of spread, epidemiology, nature of resistance, disease management, etc.

The objective of the “Workshop on Phytophthora Diseases of Tropical cultivated plants” (WOPD) was to bring together *Phytophthora* scientists working on tropical and sub-tropical crop plants with those working on temperate *Phytophthora* diseases so that there would be interaction of the experiences of the two groups, particularly the younger scientists. With this end in view participation in the Workshop was limited to persons engaged on *Phytophthora* research at present and in the past.

The Workshop was organised by the Indian Council of Agricultural Research at the Central Plantation Crops Research Institute. This Institute is responsible for conducting and coordinating research on coconut, arecanut, cashew, cacao, oil palm, and spices like cardamom, black pepper, ginger, turmeric and tree spices (cinnamon, nutmeg, clove and allspice). Many of these are affected by *Phytophthora* resulting in serious crop losses.

The Workshop was held in two parts, the first part at Kasaragod during 19-21 September 1980, and second part at Calicut during 22-23 September, 1980. The first part comprised presentation of status papers on *Phytophthora* diseases of important plantation crops and also presentation of lead and contributed research papers on different aspects of *Phytophthora* such as epidemiology, taxonomy, physiologic specialisation, resistance, screening methods and control. At Calicut there was a day long field visit during which the delegates had an opportunity to study the *Phytophthora* diseases of cardamom, pepper, arecanut, cocoa and rubber under field conditions. This was followed on the next day by presentation of papers and discussion on black pepper *Phytophthora*, the causal agent of the deadly ‘quick wilt’.

I take this opportunity to congratulate Dr. N. M. Nayar, former Director of this Institute for mooted the idea of holding a “Workshop on *Phytophthora* Diseases of Tropical cultivated plants” and for holding the same at this Institute. The efforts put in by him along with Dr. K.K. N. Nambiar, General Convenor of the Workshop are greatly appreciated. The several distinguished scientists who acted as chairmen and moderated the Sessions deserve special mention and my thanks are due to them and also to the

Sessional rapporteurs. The wholehearted cooperation and active help given by Drs. Y.R. Sarma, and T. Premkumar in putting up a scientific exhibition at Kasaragod is gratefully acknowledged.

I would also like to acknowledge the contribution of Commonwealth Foundation, UK towards the travel grants of four overseas delegates. The financial assistance received from M/s. Travancore Chemicals & Manufacturing Co. Ltd., Alwaye, Sandoz (India) Ltd., Bombay, and May and Baker (India) Limited, Bombay is gratefully acknowledged. Mr. M. C. Pothan, Managing Director, Amalgamated Estates, Pudupady, Mr. Eapen George, Bayer (India) Limited, Calicut, and a few enterprising managers of plantations like Vayitri Plantations Limited, Wynad, Meenakshi Vilas Estates, Kalladi, Wynad, and M/s. A. V. Thomas Group Companies Limited, Chulikara, Wynad, helped us in organising the study tour to Wynad areas and I place on record my heartfelt thanks to them.

A special word of thanks to Dr. B. R. Boccas, who was kind enough to show the delegates a scientific film on *Phytophthora palmivora*. After the Workshop, Prof. Newhook sent us his impressions and suggestions on the Workshop. This is included in the proceedings at the end. We are very much thankful to him for his thought-provoking suggestions.

Lastly, I wish to place on record the tremendous efforts made by the Editor, Dr. K. K. N. Nambiar, Head, Division of Plant Pathology of this Institute, for bringing out this publication.

CPCRI, Kasaragod
2 October, 1982.

K. V. AHAMED BAVAPPA
Director

PREFACE

This volume consists of the proceedings of the first workshop organised on *Phytophthora* in India. The success of Cocoa Phytophthora Workshop held at Rothamsted Experimental Station, UK during 24-26 May, 1976 was a fillip in conducting a workshop of this nature.

It was the desire of the organisers to print the proceedings so as to fall in line with the series of proceedings connected with symposia/workshop on plantation crops held at this Institute earlier.

This volume contains seven chapters corresponding to the technical sessions (including the first chapter on Inaugural Session) at the Workshop. The Second chapter contains only Status papers on *Phytophthora* diseases of plantation crops. In each of the following four chapters, lead papers are given followed by contributed research papers. Abstracts are given only for the contributed papers. At the end of each paper, the discussions held after its presentation are given. A section has been added in the end after the Plenary Session to include abstracts of papers accepted for presentation but not presented as the contributors of these papers were absent.

As Editor, I had a relatively easy task, largely because of the cooperation by the authors who showed great willingness to stick to the schedule and also to the instructions given for the preparation of the manuscript. I am very much grateful to them for this cooperation. The help rendered by Dr. Y.R.Sarma, Dr. (Mrs) Rohini Iyer and Dr.P.K.Das by way of suggestions for improving the get up and style of the proceedings deserves special appreciation. I gratefully acknowledge the invaluable help rendered by Dr. (Mrs) Rohini Iyer who helped me throughout in reading the proofs. I wish to apologise for any errors that may have remained unnoticed in spite of the best efforts to avoid them. The editor takes full responsibility for such errors.

Thanks are also due to Dr. C. R. Ramesh for taking the microphotograph used for the frontispiece and also to Shri M. Narayanan, Institute Photographer for the photographs on bud rot of coconut and *Koleroga* of arecanut. I would also like to thank M/s. Sharada Press, Mangalore for their co-operation in bringing out this volume.

CPCRI, Kasaragod,
2 October, 1982.

K. K. N. NAMBIAR
Editor



A view of the inaugural session



Some of the exhibits

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Friday, 19 September, 1980
11.00 A.M. - 12.00 Noon

INAUGURAL SESSION

Inauguration : Dr. G. RANGASWAMI

Chairman : Dr. N. M. NAYAR

Rapporteur : Dr. K. K. N. NAMBIAR

PHYTOPATHOGENIC PHYTOPHTHORAS

G. RANGASWAMI

Adviser (Agriculture), Planning Commission, New Delhi - 110 001

One of the earliest plant pathogens identified about a century ago was *Phytophthora infestans* (Mont.) de Bary causing 'potato blight' in Europe. Since then about seventy species and their varieties of the genus *Phytophthora* have been reported on hundreds of host plants, with world-wide distribution. There were several species of the fungus causing recurrent devastating damages to economic crop plants in many countries. Characteristically, the disease seems to appear overnight; before one could recognise its presence, the sweeping damage is already done. Some of the members of *Phytophthora* appear to be more specific to perennial plants than to annuals. They affect various plants including roots, stem, foliage and fruits, causing severe rots and cankers. The onslaught of these diseases are so severe, there seems to be no effort on the part of the host plant to develop and resistance - crop varieties resistant to *Phytophthora* infections are relatively unknown (Rangaswami 1961, 1972).

2. Pythiaceae fungi are among the most important plant pathogens, of which the genera *Pythium* and *Phytophthora* are well known, not only to the plant pathologists but also to the farmers in general and orchardists in particular. The fungus family Pythiaceae was created by Schroeter in 1897 to include *Pythium* and *Nematosporangium*. Later in 1923, Fitzpatrick brought *Phytophthora*, among a few other genera, under the family Pythiaceae. While several other genera were included under this family, numbering up to 14, some of them have been considered synonyms of the others. But most commonly the genera, *Pythium*, *Phytophthora*, *Pythiogeton* and *Trachysphaera* are included under this family (Vasudeva, 1962; Rangaswami *et al.*, 1970). Since extensive studies have been carried out on *Pythium* as well as *Phytophthora*, voluminous literature have poured in and continue to accumulate on these two genera.

3. Pythiaceae fungi are generally dependent on high moisture for their growth and reproduction. Asexual reproduction is often by means of ciliated zoospores which require a film of water for their mobility. Therefore, severe epiphytotic occur during rainy seasons when the asexual multiplication and spread of the propagules are made easy for the pathogen. Another important characteristic which favours the fungi is that they are capable of surviving over long periods of months and years as facultative saprophytes in soil and on organic tissues. Their sexual reproductive phase also helps in evolving new varieties, strains and races with improved pathogenic efficiency. These qualities have helped this group of pathogens to remain unconquered by the most eminent plant pathologists and plant breeders.

4. The genus *Phytophthora* founded by de Bary in 1876 is differentiated from *Pythium*, its twin sister genus, on the basis of formation and germination of sporangia.

Though several mycologists consider that since there are no sharp lines of demarcation, the two should be merged into one. Others consider that the antheridial and sporangio-phore characteristics are distinct enough and genetically stable ones to give them their identity. Classification of *Phytophthora* into species has also undergone many changes over the years. While Tucker (1931) recognised only twenty species and Leonian (1934) twenty-two species, the literature continues to entertain many more species. This has already created confusion in the minds of researchers and there seems to be no attempt to overcome these difficulties.

5. While *Phytophthora* species are known to attack equally virulently both annual and perennial plants, certain species are more common in perennial orchards than the rest. *Phytophthora citrophthora* (Sm. & Sm.) Leonian takes a heavy toll year after year in the orchards throughout the world causing gummosis and brown rot. *P. palmivora* Butl. causes severe 'leaf fall' and 'fruit rot' of mandarine oranges in submontane regions of peninsular India. Different varieties of strains of the same species also cause 'bud rot' of various palms in many tropical regions of the world. Of the nearly 200 cultivated plant species inflicted by the fungus, *P. infestans* on potato and tomato, *P. palmivora* on arecanut, coconut, palmyrah, cocoa, rubber, citrus, papaya and tobacco, *P. citrophthora* on citrus and strawberry, *P. cactorum* (L & C.) Schr. and *P. cinnamomi* Rands. on pines are considered among the more serious. While various groups of cultivated plants have been infected by *Phytophthoras*, curiously enough the cereals are free from their damage.

6. While *P. infestans*, *P. cactorum*, *P. citrophthora* and *P. cinnamomi* are prevalent in sub-tropical and temperate regions, the various varieties and strains of *P. palmivora* are tropical in their habitat (Butler and Jones, 1949). Since most of them are associated with such host plants and in such agro-climatic regions of the world which derive benefit from monsoonic rains, the damage caused is not only seasonal, but also restricted to certain agricultural tracts. High atmospheric temperature and near-saturative humidity, with limited variations between day and night temperatures seem to favour the pathogen. While the diseases occur in the plains and on the hill slopes, they are relatively unknown at higher elevations above one thousand meters.

7. The various diseases caused by *P. palmivora* are well documented in respect of their symptoms, causal agents, control measures, etc. In India *P. palmivora* causes extensive damage in the southern States, especially in Kerala, Karnataka, Tamil Nadu and Andhra Pradesh. While the disease on areca, named as 'Mahali', 'Koleroga' or 'Fruit rot' was earlier known to be caused by *P. arecae* (Colem.) Pethybridge, the same is now considered synonym of *P. palmivora*. 'Bud rot' of palms caused by *P. palmivora* is common in India, Sri Lanka, Fiji Islands, East Africa, Mauritius, Mexico, Philippines, West Indies, etc. (Rangaswami, 1961)

8. The plants affected by *P. palmivora* are of great economic value to India. Coconut which is affected by 'bud rot' is one of the most important oilseed plants and also, there are very many economic uses for various parts of the plant, such as coir from the fibre, roof thatch from the foliage, timber from the trunk, activated carbon from the nut-shell, etc. Areca palm is also a much valued tree with very many uses, besides the nut

being a foreign exchange earner. Among the *Citrus* species, the mandarines are among the most delicious of fruits and they are recurrently affected by *P. palmivora* causing 'leaf fall' and 'fruit rot' resulting in severe damage to the crop in South India. Tobacco, an important commercial crop grown in Andhra, Karnataka, and Tamil Nadu is severely affected by *P. palmivora* (Syn. *P. parasitica* var. *nicotianae* (Breda de Haan) Tucker) in certain years when climatic conditions are favourable.

9. In spite of the fact that such economic crops as mentioned above are severely damaged resulting in losses of several millions of rupees annually, adequate research is not being carried out. Studies to assess the monetary losses due to the diseases occurring on different crops in different areas would help in devoting proper attention to the research needs. The etiology of these diseases are not well understood. The pathogen seems to have specialised in affecting its various hosts, though there are some reports of cross-infectivity of its varieties and strains among some palm hosts. The occurrence of different strains of the fungus is known in other countries, but in India very limited work on this has been carried out, since the reports made in 1948 by Thomas and Ramakrishnan. Detailed studies are required to establish the nomenclature and host-specificity of *P. palmivora* and other related species.

10. Another area which needs immediate attention is the study of life cycle of the fungus. More specifically, the means of 'over-summering' of the fungus under different ecological and agro-climatic conditions are to be better understood. The sexual reproduction of the fungus and genetic basis for occurrence of new strains and physiologic races, if any, should be critically examined. The possibilities of hybridizing different species and varieties of the genus to examine the genetic basis of evolution of new species and varieties, when explored, could help in proper classification and nomenclature of the fungus.

11. Since the fungus is known to be more exacting in its requirements of certain environmental factors such as high humidity and critical maximum and minimum atmospheric temperatures for rapid asexual multiplication, detailed studies of these and other meteorological factors would help in understanding the disease onset and spread leading to forecasting of the same, so as to help farmers to take adequate preventive measures to check the onslaught. In this context it should be recognized that Phytophthoras are rich farmers' enemies, whether they own orchards or irrigated crop fields. Over the past three decades the area under irrigation in the country has more than doubled and is expected to cover nearly half of the cultivated area by the end of the present century. This would mean more moisture in the soil and more intensive cropping system with two to three crops in the same field. Such an activity would favour plant pathogens, especially the moisture-loving ones like *Phytophthora* spp. It would be necessary on our part to work out a cropping system that would discourage this group of pathogens. For example, crops which are susceptible to *Phytophthora* infection in a particular season when the atmospheric temperature and humidity are conducive should be avoided, whereas the same could be grown in the 'off-season' or summer season.

12. Not all fungicides are known to be as effective as copper fungicides such

as Bordeaux mixture in controlling the *Phytophthora* diseases of palms and citrus, especially in heavy rainfall areas of the West Coast and Malnad. A critical evaluation of the host-pathogen-fungicide interactions under the prevalent agro-climatic conditions is required to understand to formulate effective control measures. Since we know that the pathogen occurs on such crops like *Citrus* in a particular season, i.e. soon after the outbreak of S. W. Monsoon is there a better way of preparing the plants than at present, to meet the challenges of the pathogen? We have also not taken adequate measures to minimise the loss after harvest of the fruits, by the *Phytophthoras* carried from the field. Detailed studies are required to be carried out on these aspects.

13. While on one hand we observe that there is little or no resistance to infection by *P. palmivora*, on the part of the host plants, there seems to exist a high degree of specificity on the part of the pathogenic varieties of the same fungal species. While attempts to develop disease-resistant varieties of crops against *Phytophthora* have not yielded any fruitful results, the pathogen on its part seems to have evolved into more efficient varieties, races and strains capable of causing more damage to the crops and the commodities. Realising this unhappy situation, we should take more effective steps than in the past to combat the virulence of the pathogen.

14. Certain basic studies when carried out would help in formulating better means of countering the attack by the *Phytophthoras*. Why is it that the cereals do not succumb to the attack by the *Phytophthoras*, whereas members of most other families become more readily susceptible? The genetic and biochemical bases for this phenomenon if understood would go a long way in formulating biochemical means of controlling the diseases. Another area of basic research would be to identify the specific physiological requirements to induce sexual phase and also germination of oospores to give rise to asexual life-cycle of the pathogen. The factors which protect the fungus against adverse climatic conditions enabling its survival as a facultative saprophyte and/or in dormant form need to be understood. These studies on the fundamental aspects of genetics and physiology of the *Phytophthoras* would open new areas for us to more effectively tackle their pathogenic qualities.

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Friday, 19 September, 1980
2.00 P.M. – 5.00 P.M.

SESSION 1

PHYTOPHTHORA DISEASES OF CROPS

Chairman : **Dr. S. Y. PADMANABHAN**

Rapporteur : **Dr. (Mrs.) ROHINI IYER**

BUD ROT OF COCONUT

K. RADHA AND THOMAS JOSEPI*

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Irinjalakuda 680 121, Kerala, India.

INTRODUCTION

Among the diseases of coconut, bud rot is the most common and perhaps the oldest. The earliest occurrence of bud rot was in Grand Cayman in 1834 (Tucker, 1926). Since then, there had been several cases reported from various countries. These included even instances where bud rot was secondary or incidental like the West End Bud rot of Jamaica (Ashby, 1920; Martyn, 1948), the bud rot of Malaya (Sharples, 1925), and that of West Africa (Dowson, 1925). Consequent on the discussions at the Imperial Conference held in London in 1924, Butler concluded in 1925 that in true bud rot, rotting of the bud was primary and the pathogen involved was *Phytophthora palmivora*. In India, the disease occurred as an epidemic in palmyra and coconut palms (Bulter, 1906). Radha and Joseph (1974) observed the disease to be generally sporadic in all the coconut growing states; however, exceptional cases where 30 or 40 affected palms in a single garden were also noticed.

Symptoms

Paling of the leaves in the inner whorl (3rd or 4th leaf) is the earliest visible symptom to be followed by the collapse of the spear leaf. Lesions of fungal infection on leaf sheaths, fleshy leaf bases, and inflorescences in the respective leaf axils from the middle whorl of leaves (7th/8th leaves) upwards occur. The lesions appear as tiny and water soaked to begin with, later become dark brown and sunken. In advanced stages of disease the tender part of the crown including the bud completely rots (FIG. 1.1) into a slimy mass and emits foul smell while older leaves with bunches remain healthy.

The Pathogen

The causal agent of bud rot was first suspected to be *Pestalotia palmarum* by Busck (1902). Later *Bacillus coli* (*Escherichia mingula*) was considered to be responsible for the disease (Earle, 1903; Petch, 1906; Stockdale, 1907; Fredholm, 1909; Johnston, 1910 and 1911). Subsequent investigations revealed that true bud rot of coconut was fungal in origin (Shaw and Sundararaman, 1914; McRae, 1923; Tucker, 1926). Although Butler reported the causal organism as *Pythium palmivorum* in 1906, he renamed it as *Phytophthora palmivora* in 1925. That *P. palmivora* and *P. faberi* were biological variants capable of attacking coconut was recognised by Gadd (1927). Further, Ashby (1929) suggested that *P. palmivora* Butl. was the same botanical species as *P. faberi* Maubl. and *P. theobromae* Colem. According to Briton-Jones (1940) it may exist as a number of distinguishable

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morphological and pathological strains. Thomas *et al.*, (1947) showed that 25 hyphal tip isolates of *Phytophthora* from various hosts including palms fell into 'plus' and 'minus' strains which when paired produced oospores. The capability of *P. palmivora* to infect various crops causing different types of damages had been reported (Reinking, 1923; Ramakrishnan and Seethalexmi, 1956; Hickman, 1958; Chee, 1969).

Association of bacteria

Reinking (1919 and 1923) reported from the Philippines that *P. faberi* (Maubl.) was the primary cause of bud rot of coconut with bacteria occurring as secondary infection in the injured and weakened tissue. In view of the wet rot of the coconut grown in bud rot affected palms contrary to the dry rot usually caused by *Phytophthoras* the role of bacteria was investigated recently by Joseph and Radha (1975). They observed that *P. palmivora* initiated dry rot in young tissues far in advance of the wet rot. While *P. palmivora* was isolated from the dry rot affected tissue, organisms associated with wet rot were species of *Xanthomonas*, *Pseudomonas* and *Erwinia*. Infection trials on coconut petioles showed that the primary infection by *P. palmivora* when followed by bacteria aggravated the damage due to the wet rot. The bacteria were unable to establish infection on injured coconut petioles.

Isolation of causal agent

Phytophthora being a shy fungus several workers had adopted different methods involving surface sterilization, selective media, and incorporation of antibiotics in the substrate for its isolation. Joseph and Radha (1975) tried various methods for isolation using selective media suggested by Hansen (1960), Eckert and Tsao (1962) and Hendrix (1965). They found that the selection of the tissue for isolation was important. Infected parts of the crown having no wet rot readily yielded *Phytophthora* on culturing while wet rot affected material yielded only bacteria and Fusaria. Incorporation of antibiotics was not essential, but addition of 0.2 per cent Thiamine hydrochloride (1000 ppm) improved the growth of the fungus in the Glucose-nitrate medium of Hendrix.

Influence of environment

Butler (1910), and Menon and Pandalai (1958) reported that the disease was related to humid atmospheric conditions. Radha and Joseph (1976) observed that micro-climatic parameters of temperature and humidity at leaf axil level of palms exerted considerable influence in the incidence of disease. Temperature range of 21-24°C and relative humidity of 98-100% were found to be optimum. Duration or recurrence of such "favourable days" determined disease development. The "favourable days" occurred more frequently in young palms, 5-10 years old than in younger or older palms. This explains the observation of Menon and Pandalai (1958) that the disease was common in young palms.

The two overriding factors of humidity and temperature determine sporulation, germination and infection of *Phytophthora* (Crosier, 1934). Radha and Joseph (1976) recorded that *P. palmivora* required nearly a week from the time of inoculation with sporangial suspension to complete one cycle of germination, infection and production of another crop of sporangia on cut coconut petiole. A minimum period of 5 weeks was required for manifestation of disease symptoms in young palms.

Survival

The possibility of *P. palmivora* surviving in the leaf axils during dry months was suggested by Menon and Pandalai (1958). Radha and Joseph (1974) noticed survival of the fungus in infected coconut crown for over five months. They also observed production of oospores in the infected crown as well as *in vitro* when the culture was contaminated with *Thielaviopsis* sp. Brasier (1971) also observed inducement of oospore formation in *P. palmivora* due to *Trichoderma viride* and explained it (Brasier, 1972) as the effect of chemical stimulation. These observations are suggestive of the possibility of survival of the fungus as oospores under unfavourable conditions.

Experience of Pieris (1962) in Sri Lanka on the efficacy of Phenyl mercury urea compounds as powder or pellets applied between young leaves in preventing bud rot indirectly indicate the survival of the pathogen in leaf axils.

CONCLUSION

The present status of knowledge on bud rot disease and the pathogen indicates two important gaps, namely on the mode of dispersal of the pathogen and sufficient information on microclimate to develop a disease forecasting system as in the case of *P. infestans* (Beaumont, 1947; Krausie and Massie, 1953).

The requirement of frequent "favourable days" for the build up of the inoculum and the repeated infection cycles to reach the bud could be taken advantage of for prophylaxis. Application of antibiotics or chemicals having bactericidal properties along with fungicides to avoid secondary infection is to be considered.

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DISCUSSIONS

H. S. Sohi: The speaker has emphasised that the infection first develops on the leaf bases followed by leaf petioles and the buds. At what stage does bud rot occur, since the disease has been named 'bud rot'?

ANSWER: Bud rot stage appears later. The fungus complete a few cycles of growth to reach the bud which once infected collapses and rots.

D. N. Srivastava: There are reports that bud rot of coconut and other palms are incited by soft rot bacteria. Have attempts been made to check up if one or more forms of bacteria associated with bud rot of coconut palms are capable of inciting the disease without the involvement of *Phytophthora*?

ANSWER: Yes. None of the bacterial cultures isolated from bud rot affected coconut palms was capable of inciting the disease nor establishing infection without the prior infection of *Phytophthora*. Inoculation of the bacterial cultures was done on tender leaf petioles with and without *Phytophthora*. Only in the former series where *Phytophthora* was inoculated prior to bacteria did wet-rot set in.

S. Y. Padmanabhan: Pin-prick inoculation was done?

ANSWER: No.

C. S. Venkataram: Are talls more resistant? How does the fungus survive or over winter?

ANSWER: No. Perhaps as oospores; the fungus has been observed to produce oospores in infected crown.

T. N. Sreenivasan: Is *Phytophthora* soil borne? Whether the higher susceptibility of small palm is due to the proximity of inoculum in soil?

ANSWER: *P. palmivora* is soil-borne. The higher susceptibility of young palms is considered to be due to the micro-climate at leaf axil level being more favourable to the pathogen.

R. S. Mehrotra: Were the species of the bacteria identified? Were all the different combinations of organisms tried?

ANSWER : Yes.

D. N. Srivastava: Amongst the three bacteria reported to be associated, one was named as *Xanthomonas* sp. Members of this genus are all known to be plant pathogens. This identification, therefore, needs reconsideration and confirmation to avoid any confusion and misunderstanding in the future.

ANSWER : The suggestion will be taken care off.

D. H. Lapwood: The inoculations perhaps did not simulate the natural conditions?

ANSWER : Yes, it did when done in the field on young palms under ideal weather conditions.

M. J. Albuquerque: Were insects tried ?

ANSWER : No.

ABNORMAL LEAF FALL DISEASE OF RUBBER CAUSED BY PHYTOPHTHORA SPP.

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INTRODUCTION

Hevea brasiliensis the para rubber, is a sturdy quick growing, tall deciduous tree thriving well in moderately good deep loamy acidic soils. A warm humid equable climate and a fairly distributed annual rainfall of 2,000 mm are necessary for the satisfactory growth and optimum yield of rubber plants. Extended period of drought and continuous heavy rainfall and prevalence of extreme winter are not congenial climatic conditions for growing rubber. In general, the climatic conditions in most of the rubber growing regions in India, except that of Kanyakumari District, are not optimum or ideal for the crop. The annual rainfall in the rubber growing areas, except Kanyakumari District is always high, varying from 3000–6000 mm. About two-thirds of this rainfall is normally received during the South West monsoon months of June to September. Because of the very high rainfall rubber plants become susceptible to the ravages of several fungal diseases. During the South West monsoon period rubber plants are affected by a serious leaf disease called “abnormal leaf fall” caused by the fungus *Phytophthora*.

History and Distribution

The earliest record of abnormal leaf fall disease caused by *Phytophthora* in India was in 1910 from some estates in Palapilly area near Pudukad, Trichur District, Kerala State (McRae, 1919). The attention of growers was drawn to this disease by the fact that healthy seeds could not be obtained for planting owing to the prevalence of pod rot which was followed by defoliation. As years passed on, the disease had spread to all other rubber growing areas and at present it is the most destructive disease of rubber in India.

This disease was first investigated in India by McRae during the year 1914. Subsequently it was reported from Sri Lanka and Burma (Petch, 1921). In Java, Sumatra and Borneo pod rot has been recorded but damage caused by leaf fall is not high. Pod rot and leaf fall caused by *Phytophthora* have also been recorded from Cambodia, Vietnam, Liberia, Ghana, Nigeria, Cameroon, Congo, Brazil, Peru, Nicaragua, Costa Rica and Venezuela. In Malaysia serious abnormal leaf fall disease was reported during 1966 from Langkawi island and Kedah on the main land (Chee, 1969; Chee *et al.*, 1967). This disease was also observed on the East Coast in 1967, infecting plants during North East monsoon. Incidence of pod rot and leaf fall due to *Phytophthora* attack was reported from Thailand also (Chee and Greenwood, 1968). Even though this disease is recorded in several countries severe disease incidence necessitating adoption of control measures every year is observed only in South India.

Symptoms

Initially, the green pods are affected resulting in pod rot. Water soaked lesions which are pale green in colour are observed on the green pods. A few drops of latex ooze out from the lesions which coagulate and form dark spots on the infected pods. The fungal mycelia ramify inside the pericarp and also penetrate into the endosperm of the seed. The pericarp rots very soon and a white fluffy growth of the fungus appears on the surface of the fruits. The pathogen produces large crops of sporangia appearing as a thick white cheesy coating on the surface of the rotting pods under continued wet weather. Most of the infected fruits remain undehisced and attached to the branches. In some years when severe disease incidence is noticed all the fruits are destroyed making it difficult to obtain viable seeds.

On the leaves, petioles are the main targets of infection, though lesions are often observed on the stalk of leaflets and lamina as well. On the petioles dark brown water soaked lesions appear with a drop of latex coagulated in the middle. Under highly favourable weather conditions, the affected leaves often fall off with the leaflets intact and while they are still green in colour. The infected leaves sometimes become yellowish red or light purple in colour before falling off. Lesions formed on the leaflets are also water-soaked and dull green in colour. Sometimes lesions are also observed on either side of the mid-rib or on the lamina. In course of time the colour of lesions changes from dull green to various shades of black. Defoliation due to the incidence of abnormal leaf fall disease is often sudden and is completed within a fortnight under favourable conditions. After defoliation, the pathogen invades the green twigs and causes extensive die-back. Because of this, refoliation in the severely affected plants will be very sparse and hence they present a denuded appearance till the general refoliation after subsequent wintering.

Loss caused by disease incidence

Loss caused by this disease is reflected in several ways. On young immature rubber upto 3 years, leaf fall and shoot rot occur causing extensive die-back. In such cases the growth is retarded resulting in extended period of immaturity. Undesirable branching habit is also observed to develop in shoot rot affected plants. In mature plantations extensive defoliation results in considerable loss of crop. Field trials indicate that depending on the planting material and the intensity of disease infection yield loss from 30 to 50% occurs due to this disease. Extensive die-back of leaf bearing twigs also results in loss of vigour of the affected plants (Ramakrishnan and Pillai, 1961b). Artificial defoliation of mature Gl. I plants by clipping off leaves also indicated yield drop of 30% when defoliation was 75% even though die-back and other harmful effects of a parasitic disease were not involved in this case. This suggests that defoliation due to the disease may cause more yield loss (Pillai *et al.* 1974).

Pathogen causing the disease

McRae (1919) after investigating this disease for the first time identified the pathogen as *Phytophthora meadii*. Petch (1921) reported that this disease was caused by *P. faberi* in Sri Lanka. According to Ramakrishnan and Pillai (1967a), Thomson found that *P. heveae* caused bark rot and pod rot disease of rubber in Malaysia. Wellman (1954) reported that *P. palmivora* caused leaf fall and pod rot disease of rubber in Nicaragua

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(Ramakrishnan and Pillai, 1961a). Several *Phytophthora* spp. are reported to be causing pod rot, bark rot, patch canker and leaf fall disease of rubber in different countries. After detailed investigation Tucker (1931) concluded that there was no valid difference between the different species causing abnormal leaf fall disease and he preferred *P. palmivora* as the causative organism of this disease. Subsequently the pathogen causing this disease in India was identified as *P. palmivora* and *P. meadii* have been isolated from abnormal leaf fall disease infected specimens (Peries, 1966; Satchuthananthavale, 1963). In Malaysia *P. botryosa* was reported to be causing leaf fall and pod rot disease of rubber (Chee, 1969b). In India recently *P. meadii*, *P. nicotianae* var. *parasitica* and *P. botryosa* were also isolated from infected specimens (Thankamma *et al.*, 1968; Thomson and George, 1973, and Thomson and George, personal communication).

Hyphae of the fungus are intercellular or intracellular and found in all tissues of the infected part including the xylem vessels. Sporangia are found externally emerging through the stomata of the host plant. The shape and size of sporangia vary according to the species. Each sporangium is borne at the tip of the stalk which is attached to the broader base.

During favourable climatic conditions the pathogen resorts to profuse asexual reproduction by producing crops of sporangia which aid in the quick dispersal and rapid spread of the disease. The sporangia liberate reniform biciliate zoospores. They germinate to produce germ tubes, which can establish fresh infections on the host. Sporangia may also germinate directly producing germ tubes which could also cause fresh infections. It has been revealed that entry of the pathogen is through the stomata only in the case of abnormal leaf fall disease of rubber (Thankamma *et al.*, 1975).

The pathogen produces terminal or intercalary chlamydospores which are spherical in shape and hyaline. These spores are observed to germinate readily producing germ tubes. Sexual reproduction involving antheridia and oogonia resulting in the formation of thick walled oospores was noticed in culture as well as infected plant parts under field conditions. Oospores of *Phytophthora* produced in culture, on infected plant parts and those present in the soil can germinate and establish new colonies (George and Thomson, 1975; Thankamma, 1969). During off season *Phytophthora* could be isolated from soil collected from infected areas by using special medium. It was also observed that healthy rubber pods blown down by strong pre-monsoon wind often become infected on the ground before the commencement of the favourable season. All these indicate that the pathogen can overwinter in the soil with the help of thick walled oospores (George and Thomson, 1975).

Phytophthora is observed to infect very large number of host plants. *Areca catechu*, *Anacardium occidentale*, *Artocarpus integrifolia*, *Artocarpus incisa*, *Artocarpus hirsuta*, *Borassus flabellifer*, *Bougainvillia*, *Cocos nucifera*, *Colocasia* spp., *Citrus* spp., *Elettaria cardamomum*, *Jatropha curcas*, *Loranthus* spp., *Punica granatum*, *Piper nigrum*, *Spondias mangifera*, and *Theobroma cacao* are a few economically important and commonly occurring plants affected by this pathogen. When favourable climatic conditions prevail the pathogen becomes active and the disease incidence commences by the arrival of the primary

inoculum. Origin of primary inoculum could be from the thick walled oospores present in the infected and dried up material of the previous season, any other alternative hosts or from the soil itself. In the case of *Hevea*, infection is first noticed on green pods on which unlimited quantity of inoculum is built up for further spread and intensification of the disease. Disease spread is achieved by water splash and rain water running down carrying the oospores and the sporangia from the infected and rotting pods. Disease spread through the sporangia in small water particles blown by wind is also possible. This was demonstrated by isolating the pathogen from sporangia, in small water particles collected on vaseline coated slides exposed in specially designed spore traps kept at varying distances of 60 to 120 meters from mature rubber plants. Under field conditions insects like cockroaches, ants, vinegar flies and beetles which were observed to visit infected pods carried viable sporangia on their body parts. Artificial inoculation carried out using body washings of these insects under laboratory conditions produced typical infection lesions on detached leaves and fruits. Live specimens of those insects when incubated with surface sterilised green pods in moist chambers produced typical infections, indicating that those agencies could also be responsible for the spread of the disease (Thomson and Pillai, 1976).

Climatic conditions favouring disease incidence

Rainfall is the most important factor influencing the incidence of this disease. In Kanyakumari District the total annual rain fall is only about 2000 mm and during South West and North East monsoons the rain fall is normally below 300 mm in any month and hence this disease is not normally present in that District (Ramakrishnan and Pillai, 1961a). However, during 1961 and 1967 when the monthly rainfall was over 300 mm during the month of July and August, mild disease incidence was observed there also. During 1974 the South West monsoon was very heavy and severe disease incidence was noticed in most of the estates in this District leading to almost complete defoliation in highly susceptible clone like PB 86 and Tjir-1.

Continuous spell of rains for 7-10 days with 250-300 mm of rain fall without intermittent sunshine will mark the commencement of the disease. During this period the minimum temperature will be in the range of 22°C – 25°C and maximum 26°C – 30°C, with a relative humidity of about 98%. When high rainfall and relative humidity with low temperature prevail continuously the disease spreads rapidly and assumes epiphytotic proportions. Under South Indian conditions leaf fall commences a week after the onset of South West monsoon. Normally, the disease incidence is noticed by the second week of June and reaches its peak by the middle of July. However, during the years when the monsoon is late very heavy disease incidence is noticed during the middle of July to middle of August. Incidence of the disease during the month of May is also noticed after the premonsoon showers.

Clonal susceptibility

In general, all high yielding clones and clonal seedlings are susceptible to abnormal leaf fall disease under Indian conditions. Clones like PB 86, RRIM 600, Tjir 1, Tjir 16 and PR 107 are all observed to be highly susceptible to the disease. But clones like RRII 105, GI. 1, GT 1 and BD 10 are observed to retain more leaves than the susceptible

clones with similar prophylactic spraying (Ramakrishnan and Pillai, 1961b). However, without spraying, these clones are also observed to be affected severely. Reaction of seedling rubber to this disease is observed to vary and the leaf fall may not be severe and uniform as observed in the case of susceptible buddings. Successful artificial inoculation method for screening planting materials to ascertain their susceptibility to this disease by inoculating the petioles on cut twigs has been developed (Pillai and Chee, 1968).

Control measures

McRae after investigating the disease, recommended removal of all infected and dried up branches, fruits and fruit stalks of the previous season from the tree, and to destroy the potential sources of primary inoculum. He also suggested defruiting or deblossoming to prevent pod infection and further large scale multiplication of the inoculum. However, these methods were not only uneconomic but ineffective, and hence were not adopted by the growers. In later years, investigations were carried out by Ashplant (1928) at the Mycology Station, Mundakayam, Kerala State. He found that premonsoon prophylactic spraying of plants with 0.75% Bordeaux mixture was effective for the control of this disease (Ashplant, 1928). This method of protection became popular with the planters.

Systematic investigations on various diseases and pests affecting rubber and their control measures were commenced in India from 1955 onwards. To begin with recommendations of defruiting and deblossoming were experimented and found to be uneconomic and ineffective. Field experiments revealed that 1% Bordeaux mixture was more effective in controlling the disease than 0.75% Bordeaux mixture recommended by Ashplant (1928).

Though very effective, many difficulties are experienced in using Bordeaux mixture for spraying extensive areas. This fungicide is to be applied as a high volume spray necessitating the use of about 4500 to 5000 litres per hectare for satisfactory coverage. Enormous quantity of water is required for spraying large areas especially during the summer months of April-May, when water scarcity used to be experienced in most of the estates. The preparation of Bordeaux mixture is cumbersome and the keeping quality of the mixture is very poor and hence the mixture prepared may have to be used on the same day itself. Above all high volume spraying using Bordeaux mixture is a very slow, costly and highly labour intensive operation. To obviate these difficulties several ready-to-use copper fungicides were tried against this disease. It was observed that these fungicides as a single premonsoon spray were not effective in controlling this disease. Several fungicides other than copper were found ineffective for the control of this disease under field conditions. Dusting of plants with different fungicide formulations using power dusters was also subjected to field trials. But that method of protection was found to be ineffective.

Field trials with copper oxychloride dispersed in agricultural spray oil, sprayed through low volume applicators, proved effective for the control of this disease. As a result low volume spraying using micron sprayers from the ground or aerial application through helicopters are also being recommended for the control of this disease (Pillai

and George, 1973). For aerial application 6.2 litres of oil based copper oxychloride 40% or 8 kg of copper oxychloride 56% dispersed in 37 to 42 litres of spray oil is being used per hectare (Pillai, 1977). Thermal fogging using oil based copper fungicides is being experimented for the control of this disease. The total area receiving prophylactic spraying is on the increase every year indicating that the planters are becoming increasingly conscious about the adverse effect and loss of yield caused by this disease.

Chemical control of this disease though effective and practicable is expensive and will have to be repeated every year. A permanent solution will be to evolve clones resistant to this disease. For achieving this objective, a breeding programme has been initiated. From existing seedling population in estates plants showing tolerance to this disease were selected and clones were established. Such clones were again screened in the laboratory as well as under field condition. Clones imported from other countries were also screened and those showing resistance to this disease were selected. The clones selected were RR11 33, F 4542 and FX 516. But the yield of these clones was found to be poor. These clones were used for crown budding on high yielding susceptible clones like RRIM 600, 628 and GT 1. Such experimental plantings were left unsprayed and it was observed that leaf retention during the last several years even after the plants were brought under tapping was very good. The crown budded areas come into tapping about a year later

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DISCUSSIONS

T. N. Sreenivasan: Whether *P. botryosa* was isolated at Kottayam or in Andamans? Is it advisable to bring the diseased materials from isolated islands to the mainland? Are you not risking the introduction of the pathogen?

ANSWER: *P. botryosa* was isolated at Kottayam from infected specimens brought from Andaman Islands. The specimens were brought in sealed envelopes and all the materials and cultures were handled with utmost care to prevent the release of the species in the main land. This species is reported to cause abnormal leaf fall disease in Malaysia.

S. Y. Padmanabhan: Have you tried spraying following the removal of inflorescence?

ANSWER: It has been tried. But it is not economical.

C. S. Venkataram: Have you noticed any strainal variation among the isolates?

ANSWER: No. Pathogenicity difference between different regional isolates was not noticed in artificial inoculation studies.

C. Mohan: What method of tree injection was followed?

ANSWER: A locally fabricated equipment in combination with a pressurised knapsack sprayer was used for injection.

'AZHUKAL' DISEASE OF CARDAMOM

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Cardamom (*Elettaria cardamomum*), the "queen of spices" is cultivated in Karnataka, and Tamil Nadu states of India. Nearly two-thirds of the area under cultivation of this crop is in Kerala contributing to 75 per cent of the total production in this country. It is estimated that the production of cardamom during 1978-79 was about 4000 tonnes and has enabled to earn foreign exchange to the tune of Rs. 60 crores. The major constraints in the production of cardamom are the incidence of pests and diseases. "Azhukal" disease is now considered to be the most important disease of this crop, next only to 'Katte' or mosaic disease. A loss up to 30 per cent has been estimated by Nambiar and Saïma (1976). This is comparatively a new disease of cardamom, characterised by the rotting of leaves, panicles and capsules. The disease was first reported by Menon *et al.* (1972) and has been observed to be widely prevalent in the Cardamom Hills of Idukki District in Kerala. The disease is locally known as "Azhukal", meaning rotting. The incidence is observed with the onset of South West monsoon, becomes severe during August-September and continues to prevail up to December.

Symptomatology

The symptomatology of the disease was studied in detail by Nair (1979). The symptoms appear first on the exposed portions of immature unopened leaves as dark-green, water-soaked lesions. These lesions enlarge and the affected areas decay. The decayed portion becomes translucent, necrotic and the affected leaves fail to unfurl. Lesions may appear on the tips or margins of leaves. Under favourable conditions these lesions turn light yellow in colour and spread rapidly to cover the entire leaf lamina. The affected areas turn necrotic and the leaves shrivel and gradually shred. Finally, the leaf breaks at the base of the petiole, but remains hanging to it.

The leaf sheaths covering the pseudostem show brownish discolouration with water-soaked margins at the soil levels. A characteristic reddish pink colour can also be seen on the inner sheath even before the rotting. The discoloured areas develop into greyish patches of irregular shape and size with brown margins. Gradually, the basal portions of the pseudostems decay and break at the collar region by the slightest disturbance. The infected young shoots can be removed by a gentle pull. The basal portion of the affected pseudostem appears hollow from which a foul smell is emitted.

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The initial infection of rhizome is discernible as discolouration of the internal tissues. The outer surface of the rhizome becomes dark brown and watery and slimy due to the rotting. New buds are affected and they fail to develop into new shoots. All the shoots developed from the infected mother rhizome may not be affected during the same season. However, they get infected gradually. Infection of roots is evident as the discolouration of the root tips, followed by necrosis of cortical cells which spreads towards the base of the roots. The epidermis and cortex disintegrate exposing the vascular tissues. Almost all the roots are infected and decayed.

The panicles and capsules of all stages of maturity are also infected. The infection starts as small water-soaked spots on the capsules which enlarge and cover the entire surface. The infected capsules show a dull green discolouration and present a water-soaked appearance. Usually young immature capsules are infected first which shed within 3 to 5 days, leaving the stalk bare. Severely infected capsules get shrivelled and the epidermis peeled off exposing the longitudinal fibrous tissue beneath. Mature capsules, when infected, become shrivelled on drying and are inferior in quality. In infected immature capsules the seeds fail to develop which will become slimy and decay. The infection usually spreads from the capsules to the panicle. The symptoms appear on any region of the panicle as water-soaked lesions which turn yellow and later become brown in colour. The infected areas become necrotic, shrivel and dry, usually from the tip to the base. Capsules on infected panicles fail to develop to maturity but dry up. The infected panicles persist with the dried capsules even after the shoots have fallen. The disease severity is uniform in the major cultivars of cardamom, viz., the Mysore, the Vazhukka (Hybrid) and the Malabar (Nair, 1979).

Causal Organism

The causal organism is reported as *Phytophthora* sp. by Menon *et al.*, (1972). Thankamma and Pillai (1973) reported that the disease was caused by *Phytophthora nicotianae* var. *nicotianae*. Studies with the isolate of *Phytophthora* from cardamom and *P. palmivora* from coconut and rubber revealed that the isolates could cross infect the hosts. The results of the cross inoculation studies indicate that capsule rot of cardamom (Azhukal disease) may also be caused by *P. palmivora* or by its strain or variety (Radha and Joseph, 1974). Nambiar and Shirma (1976) reported *Pythium vexans* as another important pathogen of capsule rot of cardamom in addition to *Phytophthora* sp. A species of *Phytophthora* isolated from coconut also induced rotting of cardamom capsules (Nambiar and Sarma, 1976). They also observed that *Fusarium* sp. isolated from the affected capsules was non pathogenic, but enhanced the decaying process of capsule infected by *P. vexans*. They further reported that *Pythium* sp. isolated from cardamom seedlings affected with damping-off was also pathogenic to capsules both under laboratory and field conditions and indicated the important role of this fungus in the epidemiology of the disease. However, Nair (1979) reported isolation of *Phytophthora* sp. invariably from all infected parts of cardamom which has been identified as *P. nicotianae* var. *nicotianae*. Further, the studies conducted earlier at the Plant Pathology Laboratory, College of Agriculture, Vellayani, Kerala State have shown that pure culture of *Phytophthora* sp. can be isolated from seeds of infected capsules of cardamom. The slimy coating of seeds contained abundant sporangia

of *Phytophthora* sp. without any contamination from other microorganisms. This indicated that *Phytophthora* sp. is the major causative agent of Azhukal disease (unpublished).

The morphological characters of the pathogen have been described by Thankamma and Pillai (1973) and Nair (1979). Thankamma and Pillai (1973) reported that oospores were not formed in single culture, but were produced readily when paired with a rubber isolate of *Phytophthora meadii*. However, Nair (1979) observed oospore formation in culture when the soil isolate of the fungus was grown on oatmeal agar medium for 25 days at 25°C.

Studies on Isolation of *Phytophthora*

Nair (1979) carried out extensive trials on isolation of *Phytophthora* from 'Azhukal' affected plants. The organism was isolated on oat-meal agar medium from different parts of the infected plant, viz., leaf, stem, rhizome, root, panicle, capsule and seed. In all cases, *P. nicotianae* var. *nicotianae*, was obtained in culture. In addition, *Pythium vexans* Fitz. and two species of *Fusaria* were also isolated from infected rhizomes (Nair, 1979). The species of *Fusaria* were identified as *Fusarium equiseti* (Corda) Sacc. and *F. solani* (Mart.) Sacc.

The details of the isolation of *Phytophthora* carried out by Nair (1979) are given in Table 1.1. The yield of *Phytophthora* sp. was 38.3 per cent from infected leaves while it was 46.7 from stem and 10 per cent from roots. The per cent yield of the organism from panicle was only 6.7 whereas 36.7 and 56.7 per cent isolations yielded the fungus from capsule and seed respectively. The fungus could be isolated in a pure form from the seeds. There was profuse growth of mycelium and sporangia on seeds.

Table 1.1. Isolation of *Phytophthora* from different infected plant parts

Source	No. of bits plated	No. of bits yielded <i>Phytophthora</i>	Percent yield
Leaf	60	23	38.3
Stem	60	28	46.7
Rhizome	30	14	46.7
Root	60	6	10.0
Panicle	60	4	6.7
Capsule	60	22	36.7
Seed	60	34	56.7

Selective isolation of *Phytophthora* from soil

The comparative studies conducted by Nair (1979) revealed that BNPR + HMI medium (Masago *et al.*, 1977) was the most effective for maximum yield of *Phytophthora* from soil. The medium was not suitable for the growth of *Pythium* spp. However, *Pythium* spp. was more common on the other two media tested (Table 1.2). On the

Table 1.2. Comparative efficacy of selective medium on the direct isolation of *Phytophthora* from soil

Medium	Mean number of colonies (20 mg soil/plate)		
	<i>Phytophthora</i>	<i>Pythium</i>	Bacterium
PVP	24	14	3
F & H	20	11	2
BNPRA + HMI	26	2	4

selective medium, *Phytophthora* produced dense white colonies after 48 hours of incubation. The medium facilitated 8.3 per cent increase in yield of propagules of the fungus per gram of soil as compared to PVP medium of Tsao and Ocana (1969). On PVP medium, the number of colonies of species of *Pythium* was more. The F & H medium (Flowers and Hendrix, 1969) was the least effective in the selective isolation of *Phytophthora*. The BNPRA + HMI medium helped in the quantitative estimation of *Phytophthora* propagules in the soil and to isolate the pathogen in pure form. However, the average bacterial colonies were slightly more on this selective medium.

Isolation of the organism from the root zone of azhukal affected plants

Nair (1979) determined the density of population of *Phytophthora* propagules in relation to the distance from the root zone and depth of soil. The results showed that the surface layer of soil adhering to the rhizome of infected plants recorded the maximum count of *Phytophthora*. The number of propagules reduced with increase in distance from the base and depth from the soil surface (Table 1.3). There was a reduction of 70

Table 1.3. Percentage reduction in the number of propagules of *Phytophthora* in relation to the distance from the food base and depth of soil (Figures in parenthesis represent the actual mean values)

Depth (cm)	Distance from the food base (cm)			
	0	5	10	15
0	0 (1200)	70.0 (360)	95.0 (60)	100.0 (0)
5	55.0 (540)	87.5 (150)	97.5 (30)	100.0 (0)
10	85.0 (180)	92.5 (90)	100.0 (0)	100.0 (0)
15	97.5 (30)	100.0 (0)	100.0 (0)	100.0 (0)

per cent in the number of propagules at a distance of 5 cm away from the food base and the soil surface. The pathogen could not be recovered 15 cm away from the food base even from the surface soil. The observations indicated that the fungus is typically a root inhabitant having less saprophytic ability.

Effect of soil amendments on the population of *Phytophthora* and microflora of the soil

Effect of organic amendments on the population of *Phytophthora* and other microflora of soil was studied by Nair (1979). The results showed an increase on the total fungal population in amended soil (Table 1.4). Neem cake and saw dust significantly increased the fungal population by 24.6 per cent and 22.8 per cent respectively while in control, it was only 8.5 per cent. In neem cake amended soil, species of *Aspergillus* and *Trichoderma* were the predominant fungi. In soil amended with cardamom crop residue there was an increase of 24.3 per cent in the total bacterial population while a reduced count was recorded in control and saw dust amended soils (Table 1.4). Maximum increase in total actinomycetes population was observed in neem cake amended soil (59 per cent) followed by rubber cake (22.2 per cent) and cardamom crop residue (15.7 per cent). The population of actinomycetes in control and saw dust amended soil was comparatively low.

Table 1.4. Effect of organic amendments on the population of soil microflora ($\times 10^4$)

Treatment	Fungi			Bacteria			Actinomycetes			Phytophthora		
	I	II	Deviation(%)	I	II	Deviation(%)	I	II	Deviation(%)	I	II	Deviation(%)
Control	14.0	15.2	8.5	910	750	-17.5	80	65	-18.75	7.0	4.0	-42.8
Saw dust	13.5	16.6	22.9	1040	950	- 8.5	65	55	-15.3	5.0	3.0	-40.0
Cardamom crop residue	13.5	16.0	18.5	820	1020	24.3	95	110	15.7	5.0	3.5	-30.0
Neem cake	13.0	16.2	24.6	980	1100	12.2	110	175	59.0	6.5	2.0	-69.2
Rubber cake	14.0	16.4	17.1	1060	1180	11.3	90	110	22.2	7.0	5.0	-28.5

I Population before the addition of amendments

II Population 90 days after the addition of amendments.

Among the amendments tried, neem cake gave increased suppression of *Phytophthora* population in soil than control; the reduction was 69.2 and 42.8 per cent in the amended and control soils respectively. The survival period of chlamydospores of the pathogen was increased with the addition of saw dust or cardamom crop residue or rubber cake to the soil. In general, addition of neem cake as soil amendment affected maximum increase in the total fungal and actinomycetes population which resulted in the suppression of *Phytophthora* propagules in the soil.

Collateral hosts of the pathogen

Studies on the host range of the pathogen causing Azhukal disease of cardamom have shown that the cardamom isolate can infect rubber and coconut on artificial inoculation. Similarly, the isolates of *Phytophthora palmivora* from rubber and coconut infected cardamom also (Radha and Joseph, 1974). *P. nicotianae* var. *nicotianae* has been reported on *Anacardium occidentale* which caused shoot rot and leaf fall disease (Thankamma, 1974). On a survey of the natural hosts of the fungus in the cardamom plantations, it was observed that wild *Colocasia* plants growing as weed in the vicinity of the plantations can serve as a collateral host of the pathogen (Nair, 1979). He also

reported that isolates of *Phytophthora* sp. from fruits of rubber and black pepper vines could not infect cardamom plants.

Epidemiology

Nair (1979) conducted studies to correlate the incidence of Azhukal disease with the climatic and environmental conditions. He observed maximum disease incidence (11.03%) during the month of August when the atmospheric humidity was the highest (90.64%) coupled with heavy rainfall (400.4 mm). During the period, the number of propagules of *Phytophthora* was very high in the soil (786.6/g.soil) which is related to high soil moisture content, 37.0%. The details are presented in Table 1.5.

Table 1.5. Effect of climatic and environmental factors on the incidence of Azhukal disease

	Disease incidence	Soil moisture (percent)	Soil temperature °C	No. of propagules/g soil	Atmospheric temperature °C	Relative humidity (per cent)	Rain fall (mm)	No. of rainy days
January	0.25	18.6	21.2	70.2	23	43.7	6.7	0.7
February	0.07	16.8	22.0	40.0	25	46.4	24.7	2.7
March	0	16.1	24.4	27.1	26	49.5	49.3	3.0
April	0	16.5	25.6	11.2	28	44.2	99.6	8.7
May	0.40	24.2	25.4	87.5	26	66.8	105.0	12.0
June	2.68	34.3	21.1	203.5	21	82.8	321.3	26.3
July	7.53	36.5	21.3	543.4	22	84.7	396.1	27.7
August	11.03	37.6	20.4	786.6	21	90.6	400.4	28.0
September	4.76	26.3	21.2	336.0	23	75.7	66.4	13.0
October	6.67	28.7	21.8	452.1	23	67.7	254.9	15.0
November	5.24	29.4	20.9	359.3	23	62.6	264.9	10.0
December	4.77	27.0	19.4	313.2	23	76.0	33.5	6.0

The disease incidence was practically absent during the months of March and April when the soil and atmospheric temperatures were maximum and soil moisture, relative humidity and number of propagules of the pathogen were minimum. However, a gradual and steady increase in the number of the pathogen in soil was observed with the onset of rains. The number of propagules was only 87.5/g soil during the month of May, but it gradually multiplied and recorded 203.5, 543.4 and 786.6 during June, July and August respectively.

The results have clearly revealed that Azhukal disease of cardamom is directly related to soil moisture, atmospheric humidity and number of propagules in the soil which are influenced by rainfall and number of rainy days during the period coupled with low soil and atmospheric temperatures.

Phytophthora population in soil and the incidence of Azhukal disease were high in gardens of high soil pH. As the soil pH reduced, the number of propagules in the soil

also reduced. It was observed that soil pH between 6 and 7 was favourable for the survival of the pathogen which in turn increased the incidence of the disease (Nair, 1979).

Survival of the pathogen

Nair (1979) conducted detailed studies on the survival of the pathogen in soil. The pathogen was recovered up to a period of 4 weeks from the air dry field soil containing sporangia and hyphae of the fungus which was incubated at 25°C. The population reduced by 72 per cent after incubation for 2 weeks and the pathogen was not recoverable after 4 weeks from air dry soil (Table 1.6). In moist soil, the fungus survived up to six

Table 1.6. Longevity of *Phytophthora* in field soil

Source	Moisture status of soil	Colonies recovered from 20 mg soil week after inoculation									
		0	2	4	6	8	16	24	32	40	48
Sporangia and hyphae	Air dry	47	13	4							
	60% field capacity	56	22	10	2						
Chlamydospores	Air dry	76	69	61	49	30	16	5			
	60% field capacity	80	93	67	55	50	41	37	33	28	22

weeks. The fungus population was reduced to 60 per cent in 2 weeks time and 96 per cent in 6 weeks. The fungus could not be recovered after 6 weeks of incubation. The results also showed that the saprophytic existence of the pathogen in the soil was only for a period of 6 weeks in moist soil.

The fungus could survive in the air/dry field soil up to 24 weeks in the form of chlamydospores. Only 13 per cent reduction in population was observed after 2 weeks of incubation. But, gradually, the population declined. The percentage reduction was 73 and 93 after 16 and 24 weeks of incubation respectively (Table 1.6). The ability of the organism to survive in the form of chlamydospores increased significantly in soil having a moisture level of 60 per cent of the field capacity. The fungus remained viable in the moist soil even after 48 weeks of incubation; the per cent recovery was 27.3. In all the treatments, the reduction in the population of propagules was rapid during the first few weeks. The results showed that the pathogen in the form of chlamydospores can survive in moist field soil for longer periods.

The studies further revealed that the pathogen can remain viable on rhizomes of infected cardamon plants for a period of 32 weeks in air dry soil. In moist soil, the fungus remained viable for more than 48 weeks in infected rhizomes (Table 1.7). Even though most of the tissues decomposed and decayed, the fungus was still recoverable after 48 weeks. The results indicated that the pathogen is capable of surviving in infected rhizomes for considerably long periods of time and may act as source of inoculum for the initial spread of the pathogen.

Table 1.7. Survival of *Phytophthora* in Azhukal infected rhizome

Tissue	Soil moisture status	Recovery of <i>Phytophthora</i> incubation in weeks									
		0	2	4	6	8	16	24	32	40	48
Rhizome	Air dry	+	+	+	+	+	+	+	+	—	—
	60% field capacity	+	+	+	+	+	+	+	+	+	+

+ Positive isolation

— Negative isolation

Control measures

The disease can be effectively controlled by spraying the crop with one per cent Bordeaux mixture (Menon *et al.*, 1973; Nambiar and Sarma, 1974). Menon *et al.*, (1973) recommended three sprayings whereas Nambiar and Sarma (1974) reported that two sprayings with Bordeaux mixture were sufficient to get effective control. Copper oxychloride (0.2%) with a wetting agent was also effective in controlling the disease (Nambiar and Sarma, 1974).

Wilson *et al.* (1974) studied the *in vitro* effect of different organic fungicides and reported that complete inhibition of growth of the fungus could be obtained with Ceresanwet, Difolatan, Dithane C-90, Dithane M-45, Kocide, Miltox and Thiride. Nair (1979) reported that the hyphae and sporangia of the fungus could be completely inhibited by drenching the soil with Agallol 3 (200 ppm). Bordeaux mixture (1%) and Dexon (100 ppm) reduced the population by 86 and 83.5 per cent respectively. The chlamydospores of the fungus were less sensitive and none of the chemicals could eliminate chlamydospores in the soil.

Suggested Lines of Future Work

- i) Determination of the role of *Pythium vexans* in the causation of Azhukal disease and in its epidemiology.
- ii) Systematic studies to determine whether *Phytophthora nicotianae* var. *nicotianae* is genetically related and forms a strain/variant of *P. palmivora*. The host range of the organism has also to be taken up.
- iii) Estimation of crop loss due to Azhukal disease in various cardamom growing areas.
- iv) Determination of the effect of fertilizers and soil amendments on incidence and intensity of the disease.
- v) Studies to evolve suitable parameters to predict epiphytotics.
- vi) Assessment of the efficacy of organomercury fungicides as soil drench (at the onset of the monsoon) in reducing the soil-born inoculum. Studies are also required to ascertain the persistence period of the fungicides in the soil and in the crop.

- vii) Assessment of the efficacy of low volume spray using oil-based copper fungicide in the control of the disease.
- viii) Assessment of newer organic fungicides-*in vitro* and *in vivo* studies. Economics of the measures to be worked out.
- ix) Studies to evolve high yielding and resistant cultivars of cardamom.

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DISCUSSIONS

P. H. Tsao : Which oil cake was used?

ANSWER : The cake obtained from the seeds of *Melia azadirachta* L. after expelling the oil was used.

R S Mehrotra : How did you maintain the pH?

ANSWER : The pH of the soil from different estates were determined and the observations were recorded.

P. H. Tsao : Nitrogenous amendments with low C/N ratio are more effective than those with high C/N ratio

ANSWER : This is an observation with other fungi and is true in this case also.

P. H. Tsao : Besides *P. nicotianae* var. *nicotianae* are there other *Phytophthora* spp. known to attack cardamom in India, For Eg. *P. palmivora*?

ANSWER : Yes. Radha and Joseph (1974) has reported that isolates of *P. palmivora* from coconut on inoculation infected cardamom under laboratory conditions.

Thomas Joseph : Could *P. palmivora* infect cardamom?

ANSWER : In my trial *P. palmivora* isolated from coconut did not infect cardamom.

C. V. Venkataram : Have you noticed any interaction between *Phytophthora* and nematodes?

ANSWER : This aspect was not studied.

Abi Cheeran : Is the symptom a wet rot? Were bacteria present in the lesions?

ANSWER : Yes, wet rot. Bacteria used to appear in culture but *Phytophthora* was predominant in the isolations.

Y. R. Sarma : What is the percentage of clump-rot infections prevalent in the high ranges?

ANSWER : Incidence up to 11% clump rot was observed in certain localities which was associated with the Azhukal symptoms. Incidence varies with the location.

C. S. Venkataram : Liming is a common practice in plantations. What was said now is bit confusing.

ANSWER : Yes. It requires detailed studies.

H. S. Sohi : The seed harbours the fungus, i.e. seedlings can become infected easily since infected seeds can contaminate nursery soil. Therefore, nursery growers should be careful.

D. N. Srivastava : The fact that the seed is infected need not necessarily cause the disease.

ANSWER : The infected capsules are not suitable for collection of seeds and the nursery growers select only healthy plants for seed purposes.

Abi. Cheeran : Have you studied the differences between *P. nicotianae* and *P. palmivora*?

ANSWER : The morphological characters of *P. nicotianae* isolated from cardamom were distinct from that of *P. palmivora*. The identify was confirmed through CMI.

LATE BLIGHT OF POTATO

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INTRODUCTION

Late blight caused by *Phytophthora infestans* (Mont.) de Bary is by far the most serious disease of potatoes. The pathogen is capable of multiplying rapidly and may cause a heavy damage to potato crop in the shortest possible time.

In India, late blight (FIG. 1.2) was first recorded in the Nilgiri hills sometime between 1870 and 1880 (Butler, 1918). Strangely enough it never recurred there till 1961 (Srivastava, 1962) and has since then been followed by annual occurrences. In the Northern parts of the country, the late blight appeared first time during 1883 in Darjeeling from where it spread rapidly to the adjoining hills. The westward movement of the disease continued and was observed in 1897 in Kumaon hills and in 1902 in Simla hills (Butler, 1903). The disease has now become an annual feature of the crop in the Northern Hills. Its distribution and occurrence there were reviewed by Dutt (1964a, 1964b). In the plains the blight was first observed in 1898-1900 in Hugly district of West Bengal and later it was observed to occur in different areas in the northern plains. In the plains of South India, late blight was observed in Karnataka in August, 1935 and has now spread to various districts (Srikantaiya, 1962).

Loss

Late blight has the potential of quickly developing into an epiphytotic form and is often responsible for heavy loss in yield resulting even in complete crop failures. The main loss in yield is due to defoliation and haulm destruction brought about by the disease. The blight-affected tubers rot both in the field and storage, thus adding to the loss. The loss in yield depends upon the potato variety, and time of appearance and severity of the blight. Cox and Large (1960) have reviewed the blight losses which may vary on an average from 10 to 20 per cent in most of the countries. The average tuber rot due to blight may be about 5 to 10 per cent.

In India, the loss in yield caused by the defoliation of the potato crop by late blight has been reviewed by Dutt (1979). At Patna, a loss in yield of 15-75 per cent has been reported (Paharia, 1961), while in Assam a loss of 10-65 per cent was recorded (Majid, 1952). The loss in yield in some of the districts of Uttar Pradesh was estimated to be about 40 per cent during 1971. At Jullundur (Punjab) the average loss during 1962 and 1963 was found to be about 20 per cent in variety 'Upto Date' and 22 per cent in Kufri Red. In the Simla hills the field trials have been conducted annually since 1951 and the loss in yield was observed to range from nil to more than 70 per cent with an average of about 40 per cent (Table. 1.8).

Table 1.8. Loss in yield caused by *Phytophthora* blight in potato variety Upto Date in the Simla hills during 1951 to 1977.

Year	Percent loss in yield	Year	Percent loss in yield
1951	0.0	1965	40.6
1952	52.7	1966	33.6
1953	0.0	1967	50.0
1954	0.0	1968	52.0
1955	21.0	1969	47.9
1956	22.0	1970	56.6
1957	35.1	1971	73.4
1958	35.1	1972	55.0
1959	30.4	1973	72.2
1960	42.0	1974	65.5
1961	41.7	1975	45.6
1962	30.0	1976	34.0
1963	52.3	1977	23.9
1964	47.8		
		Average	39.3

It was observed from a study of the loss due to late blight that on an average for every 72.1 metric tonnes of potatoes produced in Mahasu district (Himachal Pradesh) there was a loss in yield of 27.9 metric tonnes of potatoes. Thus, in this district alone the blight was responsible for an annual loss in yield of potatoes amounting to a loss of 7.5 million rupees during 1963 and 1964.

The tuber rot caused by *P. infestans* was almost nil in blight resistant variety Kufri Jyoti while the loss may be more than 5 per cent in a susceptible variety Upto date. The tuber rot loss is higher in large and medium sized tubers as compared to that in the small tubers. The tuber rot loss is higher (8.5 per cent) in clay soil in the Simla Hills as compared to 2.3 per cent in sandy loam soil. The tuber rot loss is lower in higher parts of fields in the hills as compared to that in the lower parts where the drainage is poor. The tuber rot loss in the Simla Hills was found during 1961-63 to be 6.7 per cent in unsprayed crop, 5.2 per cent in the crop sprayed with organic fungicides and 3.9 per cent in the crop sprayed with copper fungicides. In storage the blight tuber rot is not heavy, being only one per cent.

Recently another species—*P. erythroseptica*—has been recorded to cause rotting of potatoes in the Simla hills. Up to 10 per cent tubers were affected by this disease during 1978 (Rai, 1979).

Physiologic races of *P. infestans*

Several races of the late blight pathogen have been recognised. The new races may originate due to mutation, heterokaryosis and parasexuality (Erwin *et al.*, 1963;

Leach and Rich, 1967). Mutants have been induced by irradiating zoospores of the organism with ultraviolet rays. New races may also arise on resistant varieties due to physiologic adaptation (Mills and Peterson, 1952). This was also observed in the laboratory where new races could be developed by serial passage of a race through the resistant genotype Graham *et al.*, 1961; Dutt, 1965). The genetic recombination plays the most important part in the origin of new races in places where oospores of the organism have been observed in nature (Romero and Erwin, 1969).

To identify a race of *P. infestans* a leaflet of each differential host is inoculated with a filter paper disc (3 mm) dipped in a water suspension of zoospores. The inoculated leaflets are kept on moist moss in a wooden flat covered with a hardborad sheet and incubated at 18°C. These are observed for infection on third and fifth day after inoculation and the races are designated on the basis of an international nomenclature (Black *et al.*, 1953). This system is based on the reaction of genotypes carrying recessive gene and major genes R1, R2, R3 and R4 singly or in different combinations. Later, more major genes were located and now the number has gone up to 11.

Originally 16 physiologic races of *P. infestans* were expected to occur. Later, as more genes were designated it was possible to determine more complex races. Macolmson

Table 1.9. Physiologic races of *Phytophthora infestans* recorded in India.

Locality	Race	Total
Simla Hills	0; 1; 4; 7; 8; 10; 11; 2, 3; 2, 4; 3, 4; 3, 8; 3, 9; 4, 8; 4, 11; 7, 8; 7, 9; 7, 11; 8, 9; 10, 11; 1, 8, 10; 3, 4, 7; 3, 7, 8; 3, 7, 9; 4, 7, 11; 4, 10, 11; 7, 8, 9; 8, 10, 11; 1, 2, 3, 4; 1, 3, 4, 7; 1, 4, 10, 11; 3, 4, 7, 9; 3, 4, 7, 11; 1, 3, 4, 10, 11; 3, 4, 7, 8, 9; 3, 4, 7, 8, 10; 3, 4, 7, 8, 11; 3, 4, 7, 8, 10, 11; 1, 2, 3, 4, 5, 7, 8, 11.	(38)
Mukteshwar	1, 7, 8.	(1)
Shillong	0; 1; 4; 1, 2; 1, 4; 2, 4; 3, 4; 3, 7; 1, 2, 4; 1, 4, 5; 1, 4, 7; 3, 4, 7; 1, 2, 4, 5; 1, 2, 4, 7; 1, 3, 4, 5; 1, 3, 4, 7; 2, 3, 4, 7; 3, 4, 7, 8; 1, 2, 4, 5, 6; 1, 2, 4, 5, 7; 1, 3, 4, 5, 7; 1, 3, 4, 7, 8; 1, 3, 4, 5, 6, 7; 1, 2, 3, 4, 5, 6, 7; 1, 3, 4, 5, 6, 7, 8; 1, 2, 3, 4, 5, 6, 7, 8.	(26)
Darjeeling	0; 4; 1, 4; 2, 4; 1, 3, 4; 3, 4, 7; 4, 10, 11; 1, 3, 4, 7; 2, 3, 4, 7; 3, 4, 7, 8; 3, 7, 10, 11; 1, 2, 3, 4, 5; 1, 2, 4, 10, 11; 2, 3, 4, 7, 8; 2, 3, 4, 7, 11; 3, 4, 7, 10, 11; 1, 2, 3, 4, 7, 8; 1, 2, 3, 5, 10, 11; 1, 2, 4, 8, 10, 11; 2, 3, 4, 7, 10, 11; 1, 2, 3, 4, 5, 6, 7; 1, 2, 3, 4, 5, 7, 8; 1, 2, 4, 5, 8, 10, 11; 1, 2, 3, 4, 5, 6, 7, 8.	(26)
Ootacamund	1; 4.	(2)
Kodaikanal	1, 3, 8.	(1)
Northern plains	0; 4; 11.	(3)

(1969) observed 70 different races in U.K. The number has now increased very much. In India, up to 1965, only the simple races viz., 0, 1 and 4 were prevalent (Dutt, 1965). With the introduction of blight resistant varieties a number of specialized races were found to develop. Up to 1979 a total of 72 different races were recorded (Phadtare *et al.*, 1971; Khanna *et al.*, 1975, 1977, 1978; Bhattacharyya *et al.*, 1976 and Sheo Raj *et al.*, 1978). These are summarised in Table 1.9.

Sources of infection

Late blight infected tubers are mainly responsible for the perpetuation of the disease from crop to crop. In the hills, the temperature in the stores is quite low and the late blight pathogen remains viable in the seed potatoes. In the plains the seed potatoes are kept in the cold stores and the blight organism has been reported to remain viable in such tubers (Pushkarnath and Paharia, 1963). The affected seed tubers may develop shoots which may be infected by the pathogen developed from the mother tuber. Such infected plants serve as primary foci of infection. The sporangia from the affected leaves may be splashed by rain to the nearby plants or these may be disseminated by wind over wide areas and are responsible for the infection of the crop.

Forecasting blight occurrence

Moisture and temperature are the main factors limiting the development of blight and these conditions determine the time of occurrence of the disease. Cool (12°C to 15°C) and humid (above 85% RH) weather conditions with heavy dew or rain alternating with warm (18°C–20°C) moist period favour the rapid development of the disease. On the basis of the weather conditions, different systems have been developed to forecast outbreaks of blight (Malik *et al.*, 1955). In England, Beaumont (1947) suggested two criteria necessary for blight to develop. These are (i) a minimum temperature of 10°C and (ii) a relative humidity not falling below 75 per cent for at least two days.

In India, a beginning has been made to forecast occurrence of blight in the potato crop in the hills. It has been established that (i) if 7-day moving precipitation of 20 mm or more for Simla, 38.5 mm or more for Shillong and 28.9 mm or more for Ootacamund and mean temperatures of 23.9°C or less prevail for at least 7 consecutive days, late blight would appear within 3 weeks and (ii) if the hourly temperature remains in between 10°C to 20°C associated with the relative humidity of 80% or more continuously for 18 hours for at least two consecutive days, the late blight would appear within a week (Bhattacharyya *et al.*, 1978).

Resistance breeding

Potato varieties possess two types of resistance (i) field immunity, and (ii) field resistance to late blight infection. Field immunity is a hypersensitive type of reaction and the host tissues are killed soon after the penetration of the fungus. The pathogen does not develop in the dead tissue and for all practical purposes such a variety is immune to the blight. Field immunity is governed by major dominant genes (designated as R genes) and is specific to the respective races. However, this type of resistance may not help for long as it is likely to collapse with the appearance of new races. The field resistant varieties possess only partial resistance to the disease. Such varieties may show some

blight infection but remain green in the field much longer than the susceptible varieties and thus suffer very little damage. Field resistance is non-specific and is independent of changes in the racial pattern. It is controlled by a number of minor genes or polygenes. A potato variety possessing both major genes and polygenes can effectively reduce the loss caused by late blight. A number of South American wild potatoes have been found to possess resistance to late blight. Of these, *Solanum demissum* has been widely used in breeding programmes as it possesses both major genes and polygenes for resistance to blight. In India, several hybrids have been developed by utilizing genes derived from *S. demissum*.

For development of blight resistant varieties a large number of seedlings are raised with the seed derived from crosses between resistant types. Thirty seedlings are transplanted in seedling boxes and as soon as these are about 5 cm high, these are transferred in a chamber with a temperature from 15-18°C and above 95% R.H. The seedlings are sprayed with a water suspension of swarm spores (2,500 spores/ml). Twenty four hours later the seedling boxes are transferred to a cool shady place and examined for blight reaction four days after inoculation. The susceptible seedlings are discarded and the resistant ones are transplanted in pots. Next year these are planted in the field and besides observing their reaction to blight in the fields, these are also tested in the laboratory for their reaction to different races of the blight pathogen. Later, the tubers are also tested for their reaction to late blight.

A number of blight resistant varieties namely Kufri Jyoti, Kufri Jeevan, Kufri Khasigaro and Kufri Naveen were selected and released for commercial cultivation (Dutt and Bhatia 1973 and Bhatia *et al.*, 1974). Among these, Kufri Jyoti has become the main variety in Himachal Pradesh as well as in the higher hills of Nepal. The variety gives much higher yields over variety Upto Date which was the main commercial variety, but is susceptible to blight. Its performance in the Simla Hills is given in Table 1.10.

Table 1.10. Average yield of blight resistant Kufri Jyoti and susceptible Upto Date varieties in the Simla Hills

Year	Yield in q/ha		Percentage increase over Upto-Date	S. E. ($\pm m$) q/ha	C. D. (0.05) q/ha
	Upto Date	K. Jyoti			
1969	106.0	262.2	147.4	6.8	20.3
1970	98.9	250.0	152.8	12.2	34.0
1971	68.2	175.7	157.6	15.4	44.7
1975	53.9	135.3	251.0	6.7	19.3
1977	79.3	149.7	188.8	3.7	12.0
Average	81.2	194.5	179.5		

Kufri Jyoti besides being resistant to late blight possesses immunity to common race of wart pathogen and is becoming quite popular in West Bengal hills where wart and blight

are serious problems of the potato crop. In Meghalaya hills, Kufri Khasigaro and Kufri Naveen had become quite popular. However, Kufri Naveen has recently been found very susceptible to purple top-roll and is getting degenerated. Kufri Jyoti is also becoming popular in these hills. In the Nilgiri hills, Kufri Muthu has been found to be a promising blight resistant variety. In the North Indian plains Kufri Alankar and Kufri Jyoti are being cultivated for the control of blight. However, Kufri Alankar does not possess any field resistance and a new variety K. Badshah has been released recently.

At present stress is laid on the selection of potato varieties possessing foliage resistance to blight. In the recent studies it has been found that tuber resistance is more important, as a variety possessing resistance in tubers mostly also possesses resistance in foliage while vice-versa may be true. The selection procedures have accordingly to be recognised as the blight-affected tubers are responsible for the perpetuation of the disease. Potato variety Kufri Jyoti possesses a high degree of blight resistance in the tubers.

Control

The late blight control can be achieved to a great extent through the use of fungicides. Even resistant varieties have to be given 1-2 sprayings of a fungicides to avoid development of infection which can lead to appearance of new races of the pathogen. For an effective control of blight a careful selection of a fungicide and timely application is essential. Once the blight has appeared in a crop it becomes very difficult to control the disease. In the Simla Hills, Vasudeva and Azad (1952) found Burgundy mixture to be effective

Table 1.11. Efficacy of different fungicides in the control of potato late blight in the Simla Hills during 1971-73 period

Treatment and rate per hectare	1971		1972		1973	
	A	B	A	B	A	B
Bordeaux mixture-CuSO ₄ (8 kg):						
Lime(8 kg): Water (800 litres)	10.0	22.0	5.1	20.9	11.2	46.1
Difolatan 80w, 2.5 kg	39.0	16.9			39.1	25.2
Difolatan 4F, 2.0 litres	45.0	11.9	35.0	19.1	36.2	21.4
Dithane M-45, 2.0 kg	25.1	11.6	42.8	18.1	50.0	18.6
Brestan 60W, 600 g	47.9	8.9	55.8	15.6	27.0	12.7
Fycol 8E, 3125 ml	33.0	16.8	15.0	19.9	52.2	16.5
Kocide 101, 4 kg	33.0	12.4				
Miltox 4.5 kg	66.0	10.5				
Dithane M-45 2.0 kg + Triton CS - 7			43.9	17.9	42.8	16.7
Dithane C-90.2.0 kg			59.9	14.3		
Dithane Z-78, 2 kg first spraying and Bordeaux mixture in subsequent sprays			15.5	18.5		
Control (Unsprayed)	100.0	5.8	100.0	10.0	100.0	9.8
L. S. D. = 0.05		12.2		2.7		9.4

A = % foliage infection

B = Average yield (metric tonnes/ha)

against blight while Dutt (1962) found Bordeaux mixture to give better results. The results of the trials from 1971 to 1973 are presented in Table 1.11.

A review of the fungicidal control trials carried out by different workers in various potato growing regions of India and the results of these tests has been given by Dutt (1979). The results of the field trials carried out by the author during different years have been presented annually in Fungicide and Nematicide tests published by the American Phytopathological Society.

Spraying the crop with a fungicide also helps in checking the tuber rot loss. Cultural practices like proper earthing up, good drainage and harvesting when the soil is dry also help in reducing the tuber rot.

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DISCUSSIONS

D. H. Lapwood : Were Tin sprays tried? Where do you get more tuber blight? in the hills or plains? Are there no locations in between? When do you get pink rot - in summer?

ANSWER : Duter and Brestan were tried and found quite effective in the control of late blight. Tuber blight is an annual feature in the hills. In the plains whenever blight occurs in a severe form the tuber rot is heavy as all the varieties growing in the plains are susceptible to blight. The major area under potatoes is in the

plains but the crop is grown in the mid-hills as well as in the higher hills. Pink rot is observed in the hills during summer. It has not yet been observed in the plains.

P. H. Tsao : Can you prevent infection if tubers are spared? Can soil serve as a source of inoculum?

ANSWER : Infected tubers serve as source of infection and if there is no tuber infection the disease could be controlled. The pathogen does not remain viable in soil upto the time of raising the next crop.

C. Krishnamurthy : Has the economics of tin sprays been worked out?

ANSWER : Tin compounds have been used only on experimental scale. The experiments at the Central Potato Research Institute, Simla have shown the use of tin compounds to give an increased yield of 58 Q/ha. over control (untreated susceptible variety) resulting in a net gain of Rs. 5,000/- per hectare.

R. S. Mehrotra : Does the mycelium survive in rotten tuber?

ANSWER : The mycelium does not survive in the rotten tuber.

D. N. Srivastava : I would like to have information on :

- 1) The reference giving details of the fact that potatoes in the cold store carry the pathogen and those in the ordinary storage in plains do not.
- 2) Why should the disease cause so much losses when resistant varieties are readily available?
- 3) Do our strains of *Phytophthora infestans* produce chlamydospores?
- 4) Does the fungus get into the stems from infected tubers and remain latent there and then manifest on leaves when conditions become favourable?

ANSWER : 1) Pushkaranth and Paharia, K. D. 1963. Survival of *Phytophthora infestans* in infected tubers in cold storage in the plains of India. *Indian Potato J.* 5 : 48-51

They have reported that the pathogen survived in the tubers. In the country stores, temperatures go beyond 37°C at which temperature the pathogen does not survive. However, this aspect may have to be studied. Our laboratory studies have shown that exposing the blight affected tubers at 45°C for 5½ hr kills the pathogen.

- 2) The area under susceptible varieties is still very large and that is responsible for the loss in yield due to late blight.
- 3) Yes. Chlamydospores are produced in the culture as well as in the affected stems in the hills.
- 4) The fungus from the infected tubers gets into the stem up to the soil level and at the appearance of favourable weather conditions the sporulation may develop on the stem serving as the primary source of infection.

PHYTOPHTHORA DISEASES OF CITRUS

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INTRODUCTION

In India, citrus is cultivated practically in every state, but Maharashtra, Andhra Pradesh, Karnataka, Punjab, and Assam are the leading citrus growing states. It represents the third largest fruit industry and covers about 6 per cent of the total land under fruits. Sweet oranges (*Citrus sinensis*) are grown mostly in Punjab, Maharashtra and Andhra Pradesh; and mandarin oranges (*C. reticulata*) in Assam, Meghalaya, Coorg (Karnataka), Nagpur (Maharashtra) and in Kerala. Acid lime or Kagzi lime (*C. aurantifolia*) is largely cultivated in Maharashtra, Tamil Nadu and Andhra Pradesh. Lemons (*C. limon*), Pummelos (*C. grandis*) and grape fruits (*C. paradisi*) are cultivated in scattered pockets on a limited scale.

Citrus diseases

Die-back or decline is the most important problem at present being faced by the citrus industry. This malady has been in existence since the 18th century but has assumed serious proportions during the last few decades. It has already killed millions of trees in various citrus growing areas of the world including India. Almost all the citrus species and varieties in commercial cultivation are susceptible. Many factors like unsuitable soil and climate, lack of systematic manuring, stock-scion incompatibility, high water table, soil pH, general neglect and the attack of pests, and diseases have been reported to be responsible for this. Amongst the diseases, root-rot, foot-rot, collar rot, gummosis, leaf fall and fruit rot (*Phytophthora* spp.); powdery mildew (*Acrosporium tingitanum*); scab (*Elsinoe fawceti*); canker (*Xanthomonas citri*); greening (MLO), stubborn (MLO); Iristeza (virus), exocortis or scaly bark (virus) and xyloporosis (cachexia) play a very important role in decline in different countries. Most of these diseases are prevalent in India and are responsible for citrus deterioration.

Various species of *Phytophthora*, including *P. nicotianae* var. *parasitica*, *P. palmivora*., *P. citrophthora* and *P. arecae*, have been reported to attack citrus producing a variety of symptoms. The pathogen can attack any part of the plant causing root rot, foot rot, collar rot, gummosis of branches, leaf blight and leaf fall, and fruit rot symptoms. Invariably the same fungus produces a variety of symptoms which mostly depend on the variety involved, age of host and the environmental conditions. *Phytophthora* diseases have been reported to be responsible for the decline of citrus from different citrus growing countries including U.S.A., Australia, Sri Lanka, and India. Devarajan and Aiyappa (1944) and Ramakrishnan (1954) reported leaf fall and fruit rot of mandarin oranges caused by *P. palmivora* Butl. from Coorg, Wynad (Kerala) and some parts of Pulney

Hills (in Tamil Nadu). The disease was noticed by the end of May or June depending on the onset of monsoon rains. It continued up to October-November and the incidence came down subsequently due to lack of rains and rise in temperature. Ramakrishnan (1954) further reported that *P. palmivora* was also capable of causing bark rot, crown rot and girdling of the base of the stem. Narasimhan (1927) recorded *P. arecae* on fruits of *C. medica* and *C. limon* in Karnataka, but did not state clearly whether this fungus caused gummosis or not. Kamat (1927) reported gummosis caused by *P. palmivora* Butl. to be a severe disease. Later, Naik (1949), Ramakrishnan (1954), Govinda Rao (1954) and Anonymous (1953) reported that gummosis was common in the sweet orange plantations all over South India. It was attributed to more than one species of *Phytophthora* but *P. palmivora* was more commonly associated. Chowdhury (1951) recorded that gummosis caused by *P. parasitica* was wide spread in Assam and was often associated with *Fasarium lateritium*. At Regional Fruit Research Station, Abohar (Punjab), foot rot accounted for 14 to 18 per cent of declining in trees under different varieties of sweet orange (Kapoor and Bakshi, 1967). In the surveys conducted by Dr. Fraser during 1966 for finding out the cause of citrus decline in India, root rot was found to be fairly widespread in high rainfall and heavy soil areas of Nagpur and Srirampur in Maharashtra state, and Abohar in Punjab. *P. nicotianae* var. *parasitica* has been consistently isolated by the author from diseased plants showing various types of symptoms in Coorg and Nagpur areas. Similar observations were also recorded by Lele *et al.* (1968) from Nagpur. Fawcett and Klotz (1948) reported *P. citrophthora*, *P. parasitica* and *P. palmivora* to be responsible for brown rot, gummosis and foot rot in U.S.A. A brief description of the symptoms produced by different phases of the disease is as follows.

(i) **Root rot:** This phase of the disease is quite common and widespread on mandarins in Coorg and Wynad areas of South India and North-Eastern Hill states (Assam, Meghalaya and Sikkim). In Coorg, the incidence varied from 1-26 per cent in different orchards during the monsoon season (June to September) in the past few years. The highest incidence was recorded on 8-9-year old Coorg mandarin seedlings interplanted with coffee in heavy soils. Seedling trees were more susceptible than budlings. The root rot phase is frequently overlooked but under conditions of high soil moisture it may be one of the most destructive fungal diseases responsible for deterioration of trees. Much of the damage attributed to waterlogging is in fact due to *Phytophthora* rot. *P. nicotianae* var. *parasitica* was consistently isolated from diseased roots of mandarin from Coorg. This on artificial inoculation produced typical symptoms of root rot. Gummosis, leaf fall and fruit fall symptoms were also observed and these most probably are produced by the same pathogen. Field evaluation of 18 different root stocks for Coorg mandarin at Gonicoppal and Chethalli stations of Indian Institute of Horticultural Research, Bangalore (India) indicated that after 20 years of orchard life, none of the root stocks could offer complete resistance. The results of screening of root stocks indicated that all the trifoliate, citranges and rough lemon were resistant to root infection (Basant Ram, *et al.*, 1978). Hybrid seedlings obtained with trifoliate as one of the male parents were resistant. Root stocks like Kodakithuli, Rough lemon, Rangpur lime and Cleopatra mandarin also imported resistance when used in crossing. Use of trifoliate as a source of resistance to the hybrid progenies has been well documented by Cameron and Frost (1968).

(ii) **Gummosis:** This phase manifests itself in the form of large water soaked patches on the basal portion of the stem near the ground level. Soon these patches turn brown and the bark may split, through which gum-like ooze exudes. The infection may spread in both directions on the stem, reach the main branches and roots. The external symptoms may often be restricted to the basal stem portion, whereas the infection may spread further. Gummings is sometimes noticed on higher branches also. Brown rot, collar rot or foot rot symptoms are also noticed on trees showing gummosis. It is reported from all parts of the country affecting almost all the varieties. The infection is attributed to more than one species of *Phytophthora* but *P. palmivora* is reported to be more commonly associated. *P. citrophthora*, *P. parasitica* and *P. palmivora* have been reported to be responsible for brown rot in U.S.A. (Fawcett and Klotz, 1948).

Environmental factors greatly influence the onset and advance of the disease. Heavy or ill drained soil, excessive irrigation, prolonged contact of the trunk and crown with water or moist soil, low budding, deep planting, piling up of soil around the collar, injuries to crown roots or stem during cultural operations predispose the trees to collar rot or foot rot disease. Govinda Rao (1954) reported more gummosis in black soils and in areas with high water table. Uppal and Kamat (1936) observed higher incidence of gummosis by *P. palmivora* in Maharashtra at 25-28°C. Different species and varieties of citrus react differently to gummosis. Klotz and Fawcett (*vide* Fawcett, 1936) reported that lemon was most susceptible, followed by lime, sweet orange, certain varieties of mandarin, rough lemon, sour orange and Kumquat. According to Klotz (1950) lemon and citranges were highly susceptible. Limes, grape-fruit, rough lemons, sweet oranges, mandarins and some selections of trifoliate oranges were less susceptible, while sour orange and Kumquat (Marumi and Nagami) were resistant. According to Fraser (1949) lemon and sweet lime are susceptible, while sweet orange, rough lemon and grape-fruit are moderately susceptible. Sampson tangelo, Thornton tangelo and sour orange are fairly resistant. Carrizo citrange and *Poncirus trifoliata* are, however, reported to be immune to the disease. Cleopatra mandarin is reported to be intermediate in susceptibility between sour orange and the rough lemon. In India, Uppal and Kamat (1936) reported mosamhi and pummelo as highly susceptible and lime and jambheri resistant. Hayes (1957) indicated that grape fruit and tangelo were very susceptible. Chowdhury (1951) noticed that pummelo, sweet orange, Adajamir (*C. Assamensis*), acid lime and Soh-Sarkar (*C. karna*) were highly susceptible to *P. citrophthora* in Assam, while rough lemon was less susceptible and mandarin and sour orange were immune. Under South Indian conditions, Ramakrishnan (1954) observed that sour oranges were resistant, while Sathgudi, jambheri, grape-fruit, mandarins and acid limes were more susceptible than sour orange. Lemons and citranges were highly susceptible. Govinda Rao (1954) reported that Vadlapudi and Kichili (*C. maderaspatana*) were more susceptible to collar rot. Singh (1962) found that sweet lime, Ka.nakhatta and Italian 76 (*C. jambhiri*) stocks were quite resistant for the Hill variety of mandarin at Saharanpur. Jambheri and Floride rough were intermediate, while Seville orange and Sylhet lime were rarely susceptible stocks. Sweet lime stock had no collar rot disease and was categorised as a resistant stock.

(iii) **Leaf blight, leaf fall and fruit rot:** (*P. nicotianae* var. *parasitica*, *P. palmivora*). This is serious on mandarins in high rainfall areas of South India during rainy weather. There

are reports about its occurrence from Nagpur and North-Eastern states. It results into leaf and fruit shedding. The affected trees are not killed but are exhausted resulting in drying and death of twigs. The defoliated leaves emit a very foul smell. Different mandarin varieties are comparatively more susceptible. High humidity and water stagnation favour disease development.

(iv) **Damping off of seedlings:** (*P. n. var. parasitica*). Pre- and post-emergence damping off of seedlings in the nursery beds is quite common resulting in heavy mortality when it is associated with *Rhizoctonia solani*, *Sclerotium rolfsii* and *Fusarium* sp.

Etiology, cultural characters, host range and spread: *Phytophthora* spp. are soil inhibiting fungi. They produce sporangia and zoospores on infected plant parts especially on defoliated leaves and fruits as well as on soil surface during the rainy season. The zoospores are carried by rain or irrigation water or dew or by wind and cause infection through the bud joints or any other injured part of the host. Immature green fruits borne on branches just touching the soil are infected directly from the inoculum present in the soil. Detailed cultural studies conducted by the author and his associates on *P. n. var. parasitica* revealed that oat meal agar, corn meal agar and okra seed extract agar supported good growth of the fungus. Okra seed extract agar showed maximum sporangial production. A temperature of 25-30°C and pH of 5.0 with a relative humidity of 98-100 per cent were optimum for its growth. It utilized sucrose followed by galactose and asparagin followed by aspartic acid as best sources of carbon and nitrogen respectively. Cultural and pathogenic variation among different isolates of the fungus was observed. In Florida strains of *P. parasitica*, similar variability in virulence was also noticed (Grimm and Whidden, 1962). No correlation between mycelial growth and virulence was observed.

Sporangia were produced throughout day and night but their production was more during morning and evening. Relative humidity at saturation was optimum for their production. Their germination was favoured by free water. A mean temperature of not less than 20°C, with an optimum in between 22-28°C was found suitable for disease development.

A study on host range of the fungus revealed that it could infect a variety of plants including *Passiflora edulis*, *Vitis vinifera*, *Hibiscus rosa-sinensis*, *Lycopersicon esculentum*, *Tagetes minor*, *Euphorbia hypericifolia*, *Physalis minima*, *Mirabilis jalapa*, *Commelina obliqua*, *Ipomea muricata*, *Galinsoga parviflora* and *Cynoglossum wallichii* belonging to different families. The fungus mycelium could remain viable in the soil for more than one year.

The use of contaminated planting material from infected nursery beds generally spreads the disease to new areas. Nursery maintenance is generally poor in India and seedlings are raised in the same location without proper rotation and care. This leads to greater disease incidence. In many areas the commercial nurseries are located amongst older citrus plantations which are infected. Under such situations diseased seedlings are raised.

Control

For control of *Phytophthora* diseases of citrus, a number of preventive and curative treatments are adopted. Selection of proper site with adequate drainage, use of resistant root stocks, high budding (30.4 to 45.7 cm), high planting, avoidance of excessive irrigation and water stagnation in tree basins and painting Bordeaux paste on stems above ground level (up to 60.9 cm) at least once a year are important. Replanting citrus in fields/land where this crop has been previously grown is hazardous and should not be done. Such deterioration of trees is quite evident in Nagpur area. No direct treatment is as yet available for the control of the root rot phase of the disease. Its incidence can be minimised by following various preventive measures given above. Use of resistant root stocks would give permanent solution. For control of gummosis and foot rot, application of Bordeaux paste is quite widely practised. Spraying with difolatan (0.3%) or Bordeaux mixture (4:4:40) was found effective against infection on branches as well as against leaf fall and fruit rot phase. Monthly sprays of these fungicides are regularly used in Coorg during the rainy weather. Preplant treatment of seedling roots in hot water at 35°C for 6-10 minutes (Stolzy, 1960) and treatment of seed beds, nursery and planting sites with Vapam at 0.9 litres/10 cm are advocated in U.S.A. But these have not been tried under Indian conditions. Use of disease free planting material avoids spread to new areas.

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DISCUSSIONS

F. J. Newhook : Leaf blight stage is induced by *P. nicotianae* var. *parasitica*. I am interested to know if the spread within the crown is consistent and comparable to other *Phytophthora* syndromes, viz. potato late blight, *P. cactorum* on apple and pear where occasionally in New Zealand, I have observed the whole crops of fruit becoming infected while hanging on the tree, similar to that seen in the abnormal leaf fall of rubber caused by *P. palmivora* and *P. botryosa*.

ANSWER : Symptom expression begins first on branches near the ground and then progresses towards the crown.

F. J. Newhook : Have you observed caducity of cultures of *P. nicotianae* var. *parasitica* isolated from leaves with blight symptoms that would be consistent with detachment of sporangia on the host and spread by rain-splash as is known to occur in those diseases which I have quoted?

ANSWER : We have not yet carried out such an investigation.

P. H. Tsao: *P. nicotianae* is not caducous.

D. H. Lapwood : Do you get benefit by cutting the drooping branches that touch the ground?

ANSWER : Yes please.

C. S. Venkataram : Are any instances of mycorrhizae affording protection against root infection known?

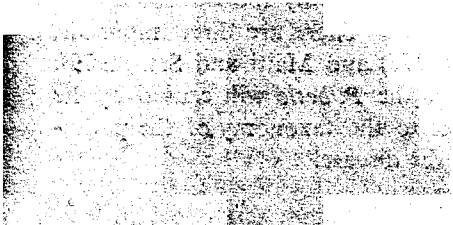
ANSWER : I am not aware in case of citrus.

F. J. Newhook : Do you have instances where scions of different varieties of citrus influence the resistance offered by trifoliolate root stocks? We have such a situation in my country.

ANSWER : We have not critically evaluated this so far.

D. N. Srivastava : Is there any relation between age of the plant and susceptibility?

ANSWER : This needs detailed study. In some varieties more susceptibility is noticed at younger age.



**PIPER BETLE WILT CAUSED BY PHYTOPHTHORA
PARASITICA VAR. PIPERINA DASTUR**

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Piper betle Linn., commonly known as betel vine is a perennial creeper belonging to family Piperaceae and is probably a native of Malaysia. It is widely cultivated in moist, tropical, sub-tropical regions of India, Bangladesh, Burma, parts of south-east Asia and Papua New Guinea for its leaves which are mainly used for chewing. The vines of *P. betle* grow as tall as ten to fifteen feet on a support in about a year's time. Its nodes bear adventitious roots which help it to climb on the support. It has been cultivated in India since the times of *Mahabharata*. The betel leaf which is commonly known as *Pan* occupies a significant place in everyday life of the Indian people. Many people are in the habit of chewing betel leaves with some accessories like lime and catechu.

The betel vine crop, being a delicate one, is attacked by several fungal and bacterial diseases and pests. The most serious of them is the foot rot (wilt) and leaf rot caused by the fungus *Phytophthora parasitica* var. *piperina* Dastur. There are three important reviews on the subject i.e. Mahmud (1952), Saksena (1977) and Maiti and Sen (1979). I shall discuss critically in brief the work done so far on the *P. betle* wilt caused by the fungus *P. parasitica* var. *piperina*, with special reference to the taxonomy of the pathogen, and the recent work done towards the control of the disease.

Symptoms

Wilted vines contrast sharply with the nearby healthy ones by their stark, chlorotic appearance and shrivelled leaves. Initially the leaves develop a slight droop, which affects the whole canopy. As droop becomes more pronounced, the leaves become flaccid and the condition at this stage may be accompanied by a paling of colour to a yellow-green. The more succulent terminal and young lateral branches now become flaccid. Finally, the leaves and shoots turn yellow and wither, eventually drying out to a pale brown colour. The underground parts which are the seats of activity by the pathogenic fungus are more or less completely rotted. If a newly diseased plant is uprooted it is found that the smaller roots are infected first and gradually the rotting spreads through the main roots to the collar region of the plant. As the rotting develops the soft tissues are readily eaten up by the fungus and the fibrous remains give the parts a shredded appearance. The rot of the underground portions is a wet one.

The leaves of the plants are also attacked. Leaves are susceptible to the disease only during the rains when both temperature and humidity are high and favourable. In most of the *P. betle* growing areas of the country, the foot-rot has been reported from June to September. However, according to Venkata Rao *et al.* (1969) wilt due to *Phytophthora* has been observed in Raja Channel area of Salem district of Tamil Nadu to set in during

the month of October and steadily increasing thereafter till January and then declines. According to Venkata Rao *et al.* (1969) rainfall or soil moisture does not appear to be predisposing factor for wilt incidence, but a critical minimum temperature of 23°C and below appears to favour wilt.

The first symptom of leaf rot is the development of a circular brown spot (FIG. 1.3) which later turns blackish and becomes soft and deliquescent in appearance. Under continuous humid conditions the spot rapidly increases in diameter and extends to the major part of the leaf causing a soft rot. The rot may extend to the petiole and in some cases to the stem also. On lower side of the infected leaf in wet conditions a white growth appears at the light coloured margins of the spots. This white growth is due to the sporangiophores of the fungus. If wet conditions are not continuous and if rain lasts only for a day or two with intervening dry warm periods the diseased areas develop concentric zones of development due to alternate favourable and unfavourable periods of growth. Under dry conditions the progress of the disease is checked totally. More than one spot may be found on a leaf and the leaf at any position of the plant is attacked.

Symptoms of foot-rot of betelvine have been described by Dastur (1935), McRae (1934) in India, Thompson (1926) in Malaya and Turner (1968) in Sarawak.

Identity of the causal organism

Hector (1927) and McRae (1934) described foot rot of *Piper betle* from West Bengal. Dastur (1925) reported a serious disease of 'Pan' causing losses of about 80% of the plantations. This was first noticed in the Central Provinces of India during the year 1923-1924 in the wetter months and was seen in subsequent years. In 1927, he published a detailed account of the disease and suggested methods of control. In Malaya, Thompson (1926) observed a similar disease of the betel vine for the first time, stating that the disease was prevalent in the Pahang district during the rainy season of 1924 and was responsible for almost entire destruction of betel vine crop. Subsequently the disease was noticed by Thompson (1928, 1929, 1933, 1939, 1940) and Tempany (1937) in Malaya and by McRae (1928, 1930, 1932, 1933, 1934), Sundararaman (1928, 1934, 1935), Sharangapani (1930), Dastur (1931, 1935), Hector (1931, 1932), Uppal (1931a, 1931b, 1938), Thomas (1939), Asthana and Mahmud (1945), Chowdhury (1944a), Thomas *et al.* (1947), Mahmud (1952), Mehrotra (1961), Tiwari (1968), Chaurasia (1976), Maiti and Sen (1977) from India and Turner (1968) from Sarawak.

The foot rot was claimed to be caused by *Phytophthora nicotianae* var. *parasitica* Dastur (Hector, 1927, McRae, 1934) and the leaf rot caused by *P. parasitica* Dastur (Chowdhury, 1944a, b), *P. parasitica* var. *piperina* (Dastur, 1935) and *Phytophthora palmivora* (Butl.) Butl. (Thomas *et al.*, 1974; Turner, 1969)

There is a considerable amount of confusion regarding the species nomenclature of the pathogen. Dastur (1935) also reported *Pythium piperinum* Dastur to be associated with the disease. *Phytophthora parasitica* was founded by Dastur in the year 1913 on *Ricinus communis*. He found the same fungus on betel vine in the year 1935 and named it as a

new variety, var. *piperina*. This distinction was based upon the larger size of the oogonium and the oospores compared to those of *P. parasitica* Dastur, though he had acknowledged the fact that the size of the oogonium is not constant and is influenced by the medium. In authentic cultures of *P. parasitica*, Tucker (1931) found that the oospores were very variable in size and exhibited a range in diameter from 12–35 μ .

Thompson (1929) reported *P. colocasiae* Racib. as the pathogen responsible for disease on *P. betle*. Thomas and Ramakrishnan (1948) have done detailed work on the morphology, sexuality and relationships of different allied species of *Phytophthora* which are related with *P. parasitica*. They reported that the isolates of *P. parasitica* var. *piperina* formed oospores with opposite plus and minus strains of *P. palmivora*, with *P. parasitica* and with *P. colocasiae*. The degree of oospore formation varied in different combinations. Oospores were formed in greater abundance with isolates recently brought in culture than with older ones. They stated that there was not enough justification to classify *P. parasitica* var. *piperina* as a new variety. The size of the oospores formed in these paired cultures of various combinations fell within the range of *P. palmivora*. Thus according to Thomas *et al.* (1948) all these species (including var. *piperina*) were not different from one another and that there were not enough grounds for distinguishing them separately. It may be emphasized that Thomas *et al.* (1948) laid great emphasis on the production of oospores in paired cultures. It is now well known that interspecific pairing in species of *Phytophthora* leads to oospore formation in many cases and hence much reliance cannot be placed on the oospore production in pairing as a criterion for similarity and dissimilarity in species of *Phytophthora*.

As early as 1929 Thompson in Malaysia identified *P. colocasiae* Racib. on *P. betle*. Turner (1969) after comparing a number of isolates from different geographic locations and hosts concluded *Phytophthora* isolates from *Piper* in South East Asia belonged to the same species and should be referred to as an atypical strain of *P. palmivora*. Similarly Maiti and Sen (1977) made a comparison of two isolates from *P. betle* leaves with those reported for *P. nicotianae* var. *parasitica*, *P. parasitica* var. *piperina* and *P. palmivora* isolated from foot rot and leaf rot of *P. betle* vines and came to the conclusion that those were more or less identical both in shape and dimensions. It was, therefore, suggested by them that all the isolates of *Phytophthora* isolated from foot rot or leaf rot affected betel vine be henceforth called as *P. palmivora*.

It may however be emphasized that in none of the above works on the taxonomy of *Phytophthora* species from *P. betle*, the recently recognized valuable criteria have been used. It has now been clearly demonstrated that the degree of sporangium caducity (expressed as percentage detached sporangia) and sporangium pedicel length are of diagnostic characters in species of *Phytophthora* (Waterhouse, 1974b; Griffin, 1977; Tsao, 1977; Tsao and Tummakate, 1977; Zentmyer *et al.* 1977) and are now emphasized as important taxonomic characters. Proper and correct identification of *Phytophthora* species has long been a difficult task due to the similarities between many of the so called species. Among the most frequently used criteria in existing keys to *Phytophthora* species (e.g. Frezzi, 1950; Leonian, 1934; Tucker, 1931; Waterhouse, 1963) are such sporangium characteristics as size, shape, length and breadth (L : B) ratio, diameter of exit pore, papilla

morphology, and/or apical thickening, many of which are known to be extremely variable (e.g. Brasier, 1969; Hendrix 1967; Leonian, 1925; Zentmyer and Ribeiro, 1977). Amongst the least emphasized sporangium characteristics are conductivity (sporangium falling off at maturity) and length of pedicel, or stalk, of the caducous sporangium. Failure to use sporangium caducity has resulted in occasional misidentifications of some *Phytophthora* isolates. Hence, it is worthwhile to investigate all *P. betle* isolates in India from this point of view. It is also possible that more than one species of *Phytophthora* might be attacking *P. betle* in different regions of the country. Certainly the taxonomy of *Phytophthora* species from *P. betle* needs reinvestigation in the light of the above observation. Table 1.12 gives a comparative study of the sporangial, chlamydospore and oospore measurements given by different workers.

Table 1.12. Relative dimensions of reproductive structures of *Phytophthora* species isolated from *Piper betle* L.

Structure	Dimensions in μ					
	McRae (1934) Leaf & stem	Dastur (1935) stem	Tiwari & Mehrotra (1968)	Turner (1969) stem	Vyas and Chaurasia (1976)	Maiti & Sen (1977) Isolate-I Isolate-II
Sporangium						
Range	19-58 × 15-35	30-65 × 20-41	31-65 × 20-45	32-60 × 18-36		23-66 × 15-41
Mean	35 × 15	41 × 20		45 × 29		40 × 26
Ratio	1.58			1.60		1.53
Chlamydospore diameter						
Range	20-50			21-41		20-40
Mean	33			29		29
Oospore diameter						
Range	14-40	18-35		19-32	28-59.5	17-41
Mean	30			26	38.8	26
Colour	Brown	hyaline to yellow		Amber to brown	Pale yellow to golden	Light Brown

Different workers from time to time worked on the relationship of temperature to growth and sporulation in the pathogen. Thomas (1939) stated that the fungus grew at a temperature range of 15-33°C with an optimum at 30°C. Chowdhury (1944) stated that an isolate he studied from Assam had an optimum of 28°C. It may be mentioned that Husain and Ahmed (1961) found maximum zoospore production and liberation in the fungus at 21-23°C. Tiwari (1973) reported 30°C as the optimum temperature for growth and sporangium production. Lower temperature favoured long period of motility of zoospores but their germination was found over a wide range with an optimum between 20-25°C. pH 7-8 was favourable for growth and sporulation. According to Tiwari (1973) sufficient aeration was also essential for sporulation. Light played an important role in the mode of germination of sporangia (FIG. 1.3a). Sporangia developed in continuous light germinated directly whereas those that developed in complete

darkness germinated indirectly. The motility of zoospores is also reduced considerably in continuous light. Tiwari (1968) correlated the recurrence, perpetuation and dissemination of foot rot and leaf rot diseases to the sporangial production and their germination.

Survival of the pathogen in soil

The pathogenicity of the fungus *Phytophthora parasitica* var. *piperina* was well established by Dastur (1930), Mahmud (1940), Chowdhury (1944) and Asthana (1947). Mahmud (1940) claimed the isolation of the pathogen from the infected *P. betle* orchard soil by Meredith method (Meredith, 1940). Chowdhury (1944) also reported the isolation of the fungus from soil. Mehrotra and Tiwari (1967) tried several other methods also but failed to isolate this pathogen from the infected orchard soils. Even when the soil was artificially infested with *Phytophthora* and kept for a fortnight or so, the efforts to isolate the pathogen were not successful. Negative results were obtained even when the selective medium containing Pimaricin (100 ppm) and Vancomycin (200 ppm) was used (Mehrotra and Tiwari, 1967) for isolating the pathogen from the soil. However, Mehrotra and Tiwari (1967) were able to isolate the pathogen from infected soil by using a selective medium used by Ocana and Tsao (1965, 1966) which contained Pimaricin, Vancomycin and Pentachloronitrobenzene at 2200 and 10 ppm, respectively. The isolation of the pathogen with the help of this modified medium is explained as follows: It has been discovered that spores and mycelia of various *Phytophthora* species have differential sensitivity to Pimaricin. At 10 ppm or less concentration of Pimaricin, the germination of resistant chlamydospores, sporangia, and zoospores are not inhibited, but at higher concentrations, i.e., 100 ppm of Pimaricin, these are inhibited from germination. When 2 ppm of Pimaricin was used, it was demonstrated by Mehrotra and Tiwari (1967) that the pathogen most probably survived as chlamydospores in the soil. It appears that *Phytophthora* even when it is present in dormant perennating structures like oospores and chlamydospores is unable to develop readily and overwhelm it.

Recently Vyas and Chaurasia (1978c) have discovered oospores of the pathogen in infected leaves of *Piper betle* in nature in the orchards. This has added a new dimension to the survival of the pathogen and recurrence of the disease year after year in the orchards. It is thus apparent that the pathogen is capable of surviving in the soil in the form of chlamydospores and oospores.

Mehrotra and Tiwari (1967) demonstrated by Wastie's method (Wastie, 1961) that the pathogen has a very poor competitive saprophytic ability in the soil.

Recently Mehrotra and Narula (unpublished data) have found both A¹ and A² mating types of *P. parasitica* var. *piperina*. Both A¹ and A² compatibility types were recorded from Sagar (M.P.). Crosses were made on CPA with standard A¹ and A² mating types of *P. parasitica* obtained from Prof. G. A. Zentmyer of University of California, Riverside, California, U.S.A. Observations on host tissue inoculation are in progress. Oospores in nature on host plant may be due to dual infection by both mating types or due to the presence of some stimulatory substances and adequate food base in the host tissue for the sexual reproduction of betelvine *Phytophthora*.

Survival of *Phytophthora* in host tissues in soil was examined by Mehrotra and Tiwari (1967). The pathogen could be isolated up to 17 weeks from artificially infested host baits buried in the soil. After 5 weeks and onwards only chlamydospores could be seen in the infected host baits.

Origin of leaf rot

Dastur (1935) observed that the disease in the betel vine orchards developed suddenly during prolonged rainy and cloudy periods which are favourable for the development of zoospores on the infected soil. With the rain splashes, the inoculum was carried over to the lower leaves first then elsewhere on the aerial parts. It was observed by Mehrotra (1961) that irrigation water from the ponds adjoining the orchards can also act as an important source for infection of leaves as the pathogen was isolated from these irrigation ponds. There is a regular practice of sprinkling water on the leaves, the source of water being the contaminated ponds. The growers usually clean the leaves of *P. betle* in these ponds and very often throw away the rot-affected leaf containing *Phytophthora* in and around the ponds. This leads to the contamination of pond water with *Phytophthora* during the rains.

Role of cuttings in spread and recurrence of foot rot and leaf rot.

There are conflicting claims with regard to the role of cuttings in the dissemination and recurrence of foot rot and leaf rot of *P. betle*. It was suggested by Chowdhury (1944), and Asthana and Mahmud (1945) that the disease was primarily carried through the plantation of infected cuttings. According to Asthana and Mahmud (1945) the cuttings taken from an infected orchard carried the disease to newer plantations. The lower portion of the vines carried the pathogen both internally as well as externally while the upper portions of the vines carried the pathogen externally. Saksena and Mehrotra (1970) showed that the infected parts were rotted rapidly and that the mycelium could not be detected in the healthy part to any appreciable distance on either side. They were of the view that the pathogen could not go undetected if cuttings were contaminated or infected with the pathogen. However, Chaurasia, Vyas and Chile (1980) opined that the apparently healthy-looking cuttings of betel vine might contain latent infection of *Phytophthora* and might be responsible for the spread of the disease from one locality to another and also for fresh infection of new orchards in the same locality.

Varietal reaction

Different varieties of *P. betle* have been found to possess different degrees of resistance against betel vine *Phytophthora* (Mehrotra, 1961; Vyas and Chile 1980). Vyas and Chile (1980) tested four varieties of betel vine viz. Madrasi, Sagar benglim, Calcutta bengli, and Kapoori against 3 isolates of betel vine *Phytophthora*. Madrasi variety was the least susceptible while Kapoori variety was the most susceptible. Extracts of Madrasi variety proved fungitoxic to all the isolates.

Interestingly, quantitative analysis of total phenols in healthy leaves of all the four varieties revealed a good deal of positive correlation. The least susceptible 'Madrasi' variety contained maximum amount of phenols (13.75 mg/g dry wt.), while the most susceptible 'Kapoori' variety contained the least amount (8.5 mg/g dry wt). In the leaves of

Calcutta and Sagar varieties phenol content was 11.9 mg/g dry wt and 9.8 mg/g dry wt respectively.

Leaf surface mycoflora in relation to leaf rot

Leaf surface mycoflora in relation to *Phytophthora* leaf rot of *P. betle* was investigated by Vyas and Chaurasia (1978b). A good degree of correlation was found in the pattern of seasonal distribution of the pathogen *P. parasitica* var. *piperina* and number of other fungi with the fluctuation of various environmental factors. The maximum percentage frequency of *P. parasitica* var. *piperina* was observed between August 16 to September 16(1972). Incidentally this was very closely associated with the maximum rainfall, higher percentage of relative humidity and considerable decrease in the number of leaf surface fungi and fall in the temperature during this period. According to Vyas and Chaurasia (1978) heavy rainfall during August and September was probably responsible for washing away the leaf surface fungi including the antagonistic forms; concomitantly low temperature, high humidity, and splashing action during rains became more favourable for the activity of *P. parasitica* var. *piperina* in the *P. betle* orchards.

Rhizosphere and rhizoplane studies

Tiwari and Mehrotra (1968) have studied the rhizosphere and rhizoplane microflora of diseased and healthy plants of two varieties of *P. betle*. A significant observation was that *T. viride* was present in the rhizosphere of the less susceptible Bengla variety and absent in Bilheri variety. Similarly the presence of *T. viride* in the rhizosphere and rhizoplane of healthy plants and its absence in diseased plants was notable.

Oxidative metabolism under Phytophthora leaf rot pathogenesis of Pan.

Vyas and Chaurasia (1979) studied the respiratory metabolism in healthy and *P. parasitica* var. *piperina* infected leaves of *Piper betle*. No direct correlation could be established between respiratory stimulation induced by *Phytophthora* infection and relative polyphenol oxidase and ascorbic acid oxidase activity in the differentially susceptible varieties of *P. betle*.

Control measures

According to Dastur (1935) the foot rot of pan could be controlled by treating the ridges with 2 : 2 : 50 Bordeaux mixture and spraying the vines with the same or with Bousol once every two months. Asthana (1947) recommended that at least a week prior to planting the pan orchard should be irrigated with Bordeaux mixture (4 : 4 : 50) one gallon per 12 linear feet. The cuttings should be immersed in Bordeaux mixture (2 : 2 : 50) for one hour before planting. After two months, the ridges should be irrigated with Bordeaux mixture similarly, once every alternate month till the existence of the garden. Bordeaux mixture applied to the soil and foliage in various doses and intervals resulted in substantial reduction in the foot rot (Dastur, 1927; 1931; Uppal 1931b; Hector, 1931; McRae 1934; Subramanian and Venkata Rao, 1970). Asthana (1947) suggested that only top-most three cuttings, 30-45 cm long should be taken from each vine for seed purposes. The lower cuttings are highly susceptible to foot rot organism as they often carry the disease internally as well as externally. Mathur and Sinha (1952) recommended that the disease could be checked if cultivation was stopped for two consecutive years

and by periodic spraying with 3% 'Perenox', a preparation of cuprous oxide. Subramanian and Venkata Rao (1964) studied the effect of 9 fungicides on foot rot of *Piper betle*. Application of Bordeaux mixture 1% once in every month from the early stage of the crop has given very effective control of the disease.

Chaurasia (1976) studied the effect of quite a large number of organic and inorganic substances including phenolic compounds, metabolic inhibitors, antibiotics and fungicides. He studied the interactions of various saprophytic fungi and/or their metabolites and also some fungicides against betel vine *Phytophthora* by applying several techniques and on the growth by cellophane paper disc method, cylinder plate method and by dry weight measurements.

Vyas and Chaurasia (1978a) recommended successful control of leaf rot and foot rot of pan by dipping the cuttings in a 1% solution of streptomycin sulphate for about 10 minutes before planting and also spraying the betel vine plants with 1% Bordeaux mixture twice a month.

Manurial treatments

Mahmud (1941) showed that if oil cake is replaced by chemical fertilizers so as to give quantities of N, P, K in the same proportion as in the oil cake, there was no appreciable difference in the disease. The earlier workers in Madhya Pradesh and Bombay have suggested restricted use of oil cake as they were found to aggravate the disease (Mahmud 1952). Ploughing the soil with well rotten farm yard manure was recommended as one of the methods of control in foot rot of pan in Bombay (Uppal, 1936). Chowdhury (1944) observed that different manures (which included mustard oil cake, sodium nitrate, a mixture of ammonium sulphate and superphosphate) had no significant effect on the incidence of the disease in Assam. The use of non-nitrogenous manures such as bone meal or superphosphate in the place of oil cakes has also been suggested (Mahmud, 1952). The experience at Pothanur (Subramanian and Venkata Rao, 1964) has revealed that application of farm yard manure to wilt sick soil results in heavy mortality of vines. The mixture of groundnut cake, superphosphate and muriate of potash has also shown increased mortality of vines. Though superphosphate has recorded low disease incidence, the effectiveness of the same has got to be confirmed by repeated trials. Thyagarajan *et al.* (1972) found that farm yard manure in combination with superphosphate considerably reduced the incidence of foot rot of betel vine.

Fumigation of the soil

The effect of three fumigants i.e., carbon disulphide, formalin and ED-CT mixture on the survival and control of *P. parasitica* var. *piperina* was investigated by Tiwari and Mehrotra (1973). Fumigation with carbon disulphide and formalin had definitely a selective effect on the microbial population in the soil. Formalin had phytotoxic effect. Effect of soil fumigation and inoculation of *T. viride* showed that better control was obtained when an antibiotic producing strain of *T. viride* was inoculated after fumigation.

Biological control

Tiwari and Mehrotra (1968) recommended dipping the cuttings in a suspension of *T. viride* spores prior to planting for the control of the disease. Chaurasia (1976) selected

fungi which had good inhibitory activity on *P. parasitica* var. *piperina*. He used *Aspergillus flavus* and *A. oryzae* for their antibiotic ability to control the foot rot disease in the soil. *A. flavus* and *A. oryzae* proved effective even in unsterilized conditions.

Mehrotra and Tiwari (1976) studied the effect of organic amendments and of antagonists growing on organic amendments on foot rot of *P. betle*. Soil amended with corn straw gave best control. Corn straw supplemented with nitrogen (NH_4NO_3) gave still better results. The effect of different antagonists growing on corn straw and til (*Sesamum indicum*) oil cake amendments showed that *T. viride* gave the best control. *Aspergillus terreus* failed to control the disease although it showed great antifungal activity under controlled conditions.

Flood fallowing and sunbaking of the soil

Tiwari and Mehrotra (1977) also recommended the control of foot rot of *Piper betle* by flood fallowing and sunbaking of the soil.

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DISCUSSIONS

P. H. Tsao : Are chlamydospores produced in great numbers? Black pepper MF₄ types do not produce. Your isolate obtained from Dr. Nambiar, is typical MF₄. Did you incubate your cultures under continuous light for sporangia formation?

- R. S. Mehrotra :** We did not incubate the cultures in the light for sporangia formation.
- D. N. Srivastava :** Research on betel vine has been carried out by several eminent workers. Does the literature not conclusively show the latent carriage of the pathogen in apparently healthy cuttings?
- R. H. Mehrotra :** Asthana (1944) proved indirectly by planting cuttings from a heavily infected field. The cuttings were taken from the top portions of the plant as well as from the basal portions. They were surface-sterilized before planting. He inferred that the basal cuttings contained the pathogen both internally as well as externally while the top portions were only externally contaminated. I found that the pathogen was carried only in the necrotic lesions and the pathogen was not found far away from the seat of infection. My contention is that cuttings can carry inoculum and such of those can be distinguished at the time of planting. Recently Chaurasia and Vyas also suggested the possibility of the cuttings carrying latent infection.

PRESENT STATUS OF COCOA PHYTOPHTHORA

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Diseases of cocoa (*Theobroma cacao* L.) caused by members of the genus *Phytophthora* are international in distribution and are the major cause of decrease in yield and increase in the cost of production of cocoa. Our knowledge of this group of pathogens with reference to cocoa was sketchy till recent times. The occurrence of three important events within the span of three years drastically changed our concept of *Phytophthora* associated with cocoa. These events in chronological order are:

- i. Commencement of the International Black Pod Research Project in Nigeria in 1973.
- ii. The publication of the treatise 'Phytophthora Disease of Cocoa' in 1974 (Edited by P. H. Gregory, Longman Group Ltd., London) and
- iii. The Cocoa *Phytophthora* Workshop held at Rothamsted Experimental Station, England in May 1976.

The publication of the book 'Phytophthora Disease of Cocoa' stimulated widespread interest in workers on the various aspects of the Black Pod disease of cocoa. The International Black Pod Research Project initiated the study on the variation in *Phytophthora* from a new angle which led to the eventual identification of the various forms of the fungus. Following a suggestion by Dr. P. De. T. Alvim (CEPLAC, Brasil) a workshop on cocoa *Phytophthora* was held at Rothamsted, England in May, 1976. Several scientists from the different cocoa-growing countries and also from the leading centres of *Phytophthora* research participated in the workshop. A representative collection of *Phytophthora* isolates from cocoa were assembled at Rothamsted prior to the workshop and for the first time in the history of *Phytophthora* research a group of scientists had the opportunity to study first hand the pathogen originating from the different cocoa-growing countries of the world. All the recent findings with regard to *P. palmivora* are the direct results originating from one or other of the afore-mentioned events.

Carruthers (1898) was the first to record a fungus associated with black pod disease of cocoa. He identified the causative fungus as *Peronospora*. Masee (1899) studied the disease on cocoa pods received from Trinidad and identified the fungus as *Phytophthora omnivora* de Bary. Butler (1907) described a fungus causing bud rot of coconut (*Cocos nucifera* L.), palmyra (*Borassus flabellifer* L.) and areca palm (*Areca catechu* L.) in the Godavari River Delta in India. The causal fungus was designated as *Pythium palmivorum*. Faber (1909) gave a description of the fungus associated with cocoa pod disease in Cameroun

and noted the differences of his isolate from *P. omnivora*. He retained only the generic name (*Phytophthora sp.*) for his isolate. The cocoa pathogen was given the binomial nomenclature (*P. faberi*) by Maublanc in 1909. The close relationships among the different '*Phytophthora*' isolated from diseased cocoa, rubber, palmyra, coconut and cotton were established by Butler (1919) and later by Ashby (1929). Butler (1919) renamed the fungus as *Phytophthora palmivora*. Several other related *Phytophthora* species were subsequently found capable of infecting cocoa, (*P. heave*, Thompson, 1929; Chee & Wastie 1970; *P. drechsleri*, Tucker, 1931; Ravise, 1970; *P. meadii*, Baker, 1936; Sreenivasan, 1975; *P. botryosa*, Chee, 1969.)

Compatibility types

P. palmivora is a heterothallic fungus with a few doubtful exceptions. Ashby (1922) paired cultures of *P. palmivora* isolated from cocoa and coconut and reported the production of sexual stages by the fungus. Gadd (1924) classified isolates of *P. palmivora* to the + or 'cocoa group' and the - or 'rubber group' based on the production of sexual stages in paired cultures. Following these reports several studies confirmed the heterothallic nature of *P. palmivora* (Ashby, 1929; Leonian, 1931; Venkatarayan, 1932; Thomas *et al.*, 1948; Spence, 1961).

Morphological differences among various isolates of *P. palmivora* and also the phenomenon of morphologically similar isolates which exhibited different compatibility characteristics were noted by Turner (1960, 1961a, 1961b).

The compatibility types in *P. palmivora* were designated as A1 (= rubber) and A2 (= cocoa) following the studies of Gallegly and Galindo (1958) on mating types and oospores of *P. infestans*. Zentmyer and Mitchell (1970) and Zentmyer *et al.* (1973) reported detailed studies on mating types and their world wide distribution based on the extensive *Phytophthora* culture collection at the University of California, U. S. A.

Waterhouse (1974) stated that *P. palmivora* exists in two distinct Morphological Forms (*P. palmivora* Morphological Form 1 (MF 1: typical form) and *P. palmivora* Morphological Form 2 (MF 2) and also an additional *Piper* form (atypical form). Each of these forms contained the A1 and A2 compatibility types. Further, she expressed the possibility of the occurrence of 'hybrid' strains with characters intermediate between the two forms and anticipated that the cytological studies then in progress (Sansome and Brasier) might yield a clue.

Sansome, Brasier and Griffin (1975) mated two cocoa isolates of *P. palmivora*, one belonging to the A1 and the other to the A2 compatibility types. Cytological examinations revealed the presence of distinctive nuclei in the sex organs. One showed 5-6 large (L) chromosomes and the other 9-12 small (S) chromosomes at metaphase. New isolates from Ghana, Nigeria and Cameroun were also studied and of ten A2 isolates examined nine were of the S and one (Cameroun) of the L chromosome type. This study established the existence of two distinctive chromosome types in *P. palmivora* infesting cocoa in West Africa and the presence of both A1 and A2 compatibility types in the L type.

Cultural studies showed that the L and S types could be distinguished when they were grown in dark on carrot agar medium. Cultures of the S type produced a diagnostic stellate pattern in the centre of the colony with very little aerial mycelial growth while the L type (A1 and A2) produced profuse aerial mycelium without the formation of any definite pattern. The authors were of the opinion that the S and L chromosome types were identical to the 'cocoa' and 'rubber' groups (Tucker, 1931), the 'G' and 'N' forms (Turner, 1960) and Morphological Form 1 and 2 described by Waterhouse (1974a) respectively. However, in a later publication (Brasier and Griffin, 1979), these comparisons were retracted except for Turner's (1960) types 'G' 'N' which were found to correspond with 'S' and 'L' forms respectively. Based on the difference in chromosome size, number and morphology between the 'S' and 'L' types separate species status for these forms were proposed (Sansome, Brasier and Griffin, (1975).

Pedicle length and caducous or persistent sporangia as a taxonomic criterion in *Phytophthora* was suggested by Waterhouse (1974b). Zentmyer (1976) presented evidence to show the importance of sporangial stalk morphology as an important diagnostic tool. He divided the *P. palmivora* isolates into four groups according to sporangial stalk characteristics.

- i. Sporangia with round base, shed with short thick stalk $< 5 \mu\text{m}$ in length, e.g. typical Ghanaian isolates.
- ii. Sporangia with round base, shed with thin stalk of length 5–15 μm , e.g. typical Nigerian isolates.
- iii. More elongated sporangia with 'sloping shoulders' and platform base shed with a long fairly thick stalk $> 15 \mu\text{m}$ in length, e.g. typical Brazilian isolates with a mean stalk length of 50 μm .
- iv. Isolates with persistent sporangia, e.g. a typical isolates.

During the course of the Rothamsted Cocoa *Phytophthora* Workshop (Griffin 1977) a general consensus was arrived at to temporarily group the different forms of *P. palmivora* on cocoa into four morphological forms.

MF 1 Cultures on carrot agar stellate/striate (smooth-combed) with sharp well-defined edge and aerial mycelium usually sparse. Sporangia with rounded base shed with a short broad occluded pedicel. ($< 5 \mu\text{m}$ in length). Cosmopolitan. Predominantly of the A2 compatibility type (A1 in Jamaica). Small chromosomes, $n = 9-12$. An important cocoa pathogen.

MF 2 No cultures were seen at the Workshop but apparently they have the stellate/striate appearance similar to MF 1. Sporangia have a rounded base and are shed with a short occluded pedicel ($< 5 \mu\text{m}$). Geographical distribution not known. Chromosome type not known. Importance as a pathogen of cocoa not known.

- MF 3** Cultures on carrot agar with no distinctive colony pattern, with plentiful cotton-wool like aerial mycelium and with the leading edge less well defined than in MF 1 isolates. Sporangia with a rounded base shed with a thin stalk containing cytoplasm of length $> 5 \mu\text{m}$ but $< 15 \mu\text{m}$. Mainly restricted to the Nigeria/Cameroun/Gabon area. Predominantly of the A1 compatibility type. Large chromosomes, $n = 5-6$. An important cocoa pathogen.
- MF 4** Cultures on carrot agar with a petaloid pattern and with a moderate amount of aerial mycelium. Elongated sporangia with 'sloping shoulders' and a platform across the base are shed with fairly thick long stalks ($> 15 \mu\text{m}$ in length). Predominant in South American countries but not restricted to them. Chromosome type not known. An important pathogen of cocoa.

Brasier and Griffin (1979) published a detailed study involving c 950 *Phytophthora* isolates from cocoa with respect to morphology, physiology, chromosome type and number, cultural characters on carrot agar, cocoa pod lesion characteristics, sporangial morphology including size, pedicel length, chlamydospores and sex organs, compatibility type, growth on synthetic medium, response to *Trichoderma* and temperature relations. The results of this study showed that the majority of isolates of *P. palmivora* from cocoa belonged to one of the three main forms, S(MF1), L (MF3) and MF4. The authors were of the view that all the three forms were separate species on the basis of their cytological, morphological and physiological differences. They proposed that the S(MF1) only should be referred as *P. palmivora* and the term MF1 would be superseded. The L (MF3) type was described as a new species, *P. megakarya*.

None of the cocoa isolates examined by them could be identified as MF2 (Waterhouse, 1974a) and hence it was not retained as an acceptable taxon within *P. palmivora*.

Leonian (1922) reported a stem and fruit blight disease of Pepper (*Capsicum annuum*) and identified the causal agent as *P. capsici*.

Baker (1936) isolated a new *Phytophthora* from cocoa pods and identified it as 'strain C' (*Phytophthora* sp. *arecae-meadii* group). Sreenivasan (1975) reported the isolation of a *Phytophthora* from naturally infected cocoa pods in Trinidad which resembled very closely the isolate described earlier by Baker. This isolate was subsequently assigned to the MF4 group of *P. palmivora* at the Cocoa *Phytophthora* Workshop at Rothamsted.

Hunter *et al.* (1971) described a new *Phytophthora* blight of *Macadamia integrifolia* racemes. Two types of *Phytophthora* were found to be associated with the disease viz. *P. nicotianae* var. *parasitica* and *P. palmivora*. Kunimoto *et al.* (1976) corrected the earlier identification and named the pathogen responsible for the disease of *Macadamia* as *P. capsici*. The photograph of the sporangia published in their paper showed striking similarity to the sporangia found in MF4 of *P. palmivora*.

Zentmyer *et al.* (1977) and Kaosiri *et al.* (1978) presented evidence on stalk length of sporangia and its application as a taxonomic criterion for classifying *P. palmivora* isolates.

They divided eighty isolates of *P. palmivora* from cocoa into three main groups based on the sporangial pedicel length. They also identified a fourth group consisting of atypical isolates with noncaducous (persistent) sporangia. Their three main groups corresponded to the morphological forms 1, 3 and 4 assigned to isolates of *P. palmivora* at the cocoa *Phytophthora* Workshop. Kaosiri *et al.* (1978) also noted the similarities between *P. palmivora* MF4 isolates and *P. capsici*.

Brasier and Medeiros (1978) determined the chromosome type and number of three Brazilian isolates (MF4). These isolates showed 9-12 small type (S) chromosomes.

Zentmyer *et al.* (1979) presented further evidence to show the close similarity between *P. capsici* and MF4 of *P. palmivora*. The authors suggested that the Morphological Form 4 (MF4) of *P. palmivora* is a form of *P. capsici*.

Distribution of the different species of *Phytophthora*

Of the three species of *Phytophthora* on cocoa (*P. Palmivora*, *P. megakarya* and *P. capsici*) only *P. palmivora* is worldwide in distribution. According to Brasier and Griffin (1979) *P. megakarya* is restricted to West Africa (Nigeria, Cameroun, Dahomey, Gabon and Fernando Po); Zentmyer *et al.* (1979) reported *P. megakarya* on Hevea stems from Malaysia and in forest soil from New Guinea. *P. capsici* was initially believed to be present only in the Americas and Caribbean area. Studies by Zentmyer *et al.* (1979) and Bakala (1979) established the presence of *P. capsici* in Cameroun on cocoa. From the available information there is only one country (Cameroun) which has all three *Phytophthora* species recorded on cocoa.

Compatibility types

Even though two (A1 and A2) compatibility types were recorded for each of the three *Phytophthora* species, in nature, one type attains dominance over the other in distribution. Thus in *P. palmivora* the A2 compatibility type is predominant throughout the world except Jamaica, where the A1 type was reported to be prevalent. Contrary to this situation, in *P. megakarya* the A1 compatibility type was reported to be widespread in distribution. There was only one identification of A2 type in *P. megakarya* (Brasier and Griffin, 1979). Both A1 and A2 types were recorded in *P. capsici* on cocoa and the data indicated the predominance of the A1 compatibility type. Zentmyer's (1974) data on variation, genetics and geographical distribution of mating types need to be revised in the light of the new findings on cocoa *Phytophthora* species.

Future Work

The recent advances reported here illustrate the need for similar studies with respect to other *Phytophthora* groups. There are several countries where cocoa was introduced recently. Malaysia and India are two examples of such countries. Representative *Phytophthora* isolates from India were not available for study at Rothamsted. Studies involving large representative samples of pathogens and taxonomic study based on established characteristics of the new samples should be carried out in individual cocoa-growing countries. Several such studies are anticipated in the future. However, a general consensus on methods and standardisation of techniques to be adopted by workers

in this field will go a long way in preventing possible confusion which may arise from future discoveries.

Several of the previous reports of *P. palmivora* on cocoa need to be updated to include recent findings. Reaction of cocoa cultivars to the black pod pathogens should be reassessed. Much of the discrepancies reported by various workers with regard to reaction of cocoa cultivars to the black pod pathogen could now be explained rationally. New strategies have to be developed in devising control measures against *Phytophthora* diseases of cocoa and renewed vigilance should be instituted in quarantine measures to prevent the unnecessary spread of pathogens.

Any stipulation on evolutionary sequence of this group of organisms at this particular period of time may be premature. The new findings will stimulate extensive studies in the genus *Phytophthora*, the results of which will enable us to clarify the status of *Phytophthora* on cocoa satisfactorily.

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DISCUSSIONS

K. V. Chandrasekhara : A1 isolates produce sex organs in presence of *Trichoderma viride*. What is the kind of stimulation it receives? Hormonal? It is interesting to note that *T. viride* is known to produce a kind of antagonism (called hyphal interference) in certain other interactions. Sometimes it acts through viridin, an antibiotic.

ANSWER : A volatile material (still unidentified) is responsible for the effect. The stimulation is effected even when the mycelia are not in contact. *T. viride* is a hyperparasite of several fungi including *Phytophthora*. Probably, the reaction by *Phytophthora* is a 'survival' technique induced by the presence of 'natural enemy'.

KOLEROGA OF ARECANUT

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INTRODUCTION

Arecanut or betelnut palm (*Areca catechu* L.) is a perennial crop the nuts of which are used for chewing purposes besides being used at all religious and social functions in India. They are affected by a number of maladies among which the *kolerga* is important.

Kolerga (*Kole* = rotting, *Roga* = disease) as is called in Karnataka, is otherwise known as *Mahali* (heavy devastation) in Kerala or fruit rot in a number of places. The disease was first recorded in South India by Butler in 1906. It occurs in severe form in heavy rainfall areas (Coleman, 1910; Anstead, 1924; Sundara Raman and Ramakrishnan, 1924; Venkata Rao, 1927; Venkatarayan, 1937; Marudarajan, 1950a; Kamat, 1953; Anonymous, 1954, 1962c, 1967, 1972; Gokale *et al.*, 1955; Dorasami, 1956; Nambiar, 1956; Patel and Nagaraja Rao, 1958; Mundkur, 1967; Seshadri and Rawther, 1968; Singh, 1973).

Loss due to disease

Detailed and regular surveys have not been made to estimate the loss due to this disease. However, an annual loss of 10-75% in parts of Karnataka and Kerala States, or total destruction of crops in individual gardens (Coleman, 1910; Thomas, 1937; Nambiar, 1956; Anonymous, 1960a) have been recorded. Coleman and Venkata Rao (1918) estimated an annual loss of Rs. 4 lakhs in Malnad region (Karnataka) alone.

Symptoms

The fungus makes its active appearance about 15-20 days after the commencement of South West monsoon (May-June) and persists till about October (Marudarajan, 1950a). The first sign of the disease is on the surface of arecanuts where water soaked lesions usually develop towards the base. These lesions gradually spread ultimately covering the entire nut and give dark appearance to fruits. The affected nuts rot and drop down from the bunches (FIG. 1.4.) A felt of white mycelial mass develops on the fallen nuts which soon envelops the entire surface (Butler, 1906; Coleman, 1910; Shaw, 1913; Venkata Rao, 1915, 1919; Marudarajan, 1950 a; Kamat, 1953; Gokhale *et al.*, 1955; Patel and Nagaraja Rao, 1958; Sannegowda, 1961; Mundkur, 1967; Seshadri and Rawther, 1968; Anonymous 1969 a, b, 1972; Nambiar, 1971; Rangaswami, 1972; Singh, 1973). As the disease advances, the fruit stalks and axis of inflorescence are also affected (Sundararaman and Ramakrishnan, 1924; Marudarajan, 1950 a). Affected nuts are lighter in weight and possess large vacuoles and dark brown radial strands internally. Late infections occurring

in August result in drying up of nuts which stick to bunches (Marudarajan, 1950 a; Seshadri and Rawther, 1968). These nuts are often affected by saprophytes like *Gloeosporium* sp and are locally called "dry mahali" in Central Kerala. The decreased susceptibility of nuts to disease with increased maturity (Anonymous, 1962 a, 1963) may be probably due to prevalence of unfavourable weather factors at the time of maturity.

The same fungus also infects the crown region and causes bud rot resulting in the death of the palm. The leaves become yellow, droop down, and drop off one by one leaving the stem bare. Secondary organisms enter the rotting bud and make it into a slimy mass which would emit a fetid odour (Coleman, 1910; Nambiar, 1949, 1956; Anonymous, 1954, 1972; Naidu, 1960; Seshadri and Rawther, 1968).

Fungus

The pathogen was first named as *Phytophthora omnivora* De Bary by Sydow and Butler (1906, 1907). Later, Coleman (1910) called it as *P. omnivora* var *arecae*. Pethybridge (1913) recognized that the fungus was quite different from De Bary's *P. omnivora* and suggested that it might be regarded as *P. arecae*. Finally Butler (1918) considered it as *P. arecae* (Coleman) Pethybridge. Mycelium of the fungus is coenocytic, but sparsely septate in the older stages. It is inter and intracellular, haustoria are finger like, occasionally branched and sparsely produced. The hyphal diameter varies from 8-9 μm (Coleman, 1910; Mundkur, 1967). The fungus grows and sporulates better on steamed cornmeal than on potato dextrose agar and oatmeal agar (Tucker, 1931).

Asexual reproduction is by the production of sporangia with zoospores and by chlamydospores. Sporangia are borne on irregularly branched sporangiophores. Sporangia are papillate, pyriform to elliptical measuring 30-70 μm \times 24-46 μm . (Leonian, 1925, and Herbert, 1929; Gadd, 1927; Sundararaman and Ramakrishnan, 1924; Mundkur, 1967; Singh, 1973). The average dimensions are 34.52 \times 24.67 μm (Tucker, 1931) and 47.92 \times 30.05 μm (Rosenbaun, 1917).

Chlamydospores form one of the perennating structures. Their size varies from 18-40 μm , average being 25.74 μm (Tucker, 1931; Gadd, 1927; Newhook *et al.*, 1978).

The sexual spores, oospores were reported to be absent (Sundararaman and Ramakrishnan, 1924). The failure to observe oospores in nature was thought to be due to the presence of two strains which are localised (Uppal and Desai, 1939). In fact, Uppal (1942) recorded the occurrence of + and - strains. Antheridia are amphigynous and the oogonia range from 28-40 μm (Coleman, 1910) and the oospores from 17.5-24.4 μm (Thomas *et al.*, 1947) to 25-35 μm (Newhook *et al.*, 1978) in diameter. The fungus produces oospores on inoculated arecanuts and on *Cereus formosus* and *Clarkia elegans* (Coleman, 1910), as well as on fresh bean agar (Desai, 1950 a, b). As the homothallic nature of the fungus was observed by some investigators (Narasimhan, 1932; Ramakrishnan, 1954; Ramakrishnan and Seethalakshmi, 1956 a), others (Ashby, 1929; Narasimhan, 1930, 1931 a; Venkatarayan, 1932; Uppal and Desai, 1939; and Marudarajan, 1941) reported that it was heterothallic. Narasimhan (1930, 1931a) reported that the strains from areca and *Loranthus* have male mycelium and those of *Santalum* and *Jatropha*

the female mycelium. Venkatarayan (1932) even observed the formation of oospores when mixed with the heterothallic strain from *Santalum album*. The formation of oospores in the mixed cultures of *P. arecae* with isolates from coconut, palmyrah palms, and *Hevea* but not among themselves was observed by Marudarajan (1941) and Thomas *et al.*, (1947). Ashby (1929) obtained oospores in mixed cultures of *P. arecae* and *P. meadii*, whereas Gallegly (1964) did so in paired cultures of *P. arecae* and *P. infestans*.

Epidemiology

While heavy rainfall with constant high humid conditions (Narasimhan, 1922; Venkata Rao, 1925; Mundkur, 1967;) and an alternation of sunshine and rain (Coleman 1910; Mundkur, 1967) are conducive to disease development, the heavy rain and wind (Coleman, 1910; Venkata Rao, 1925; Nambiar, 1956; Mundkur, 1967) and to certain extent insects and small birds (Coleman, 1910) facilitate its spread. It is pertinent to note that this period is also marked by low temperature (20-23°C). The intensity of koleroga is often very severe in plantations situated in valleys or those surrounded by thick belts of trees (Kamat, 1953) or covered heavily with intercrops resulting in high humid conditions.

With a view to correlating the disease incidence with meteorological data, these were examined for the years 1970-1979 (Table 1.13). The data showed that the annual rainfall

Table 1.13. Meteorological data during South West monsoon season for the years 1970-1979 at CPCRI, Regional Station, Vittal*

Month	Rain fall (mm)	Temperature (°C)		Humidity %	Sunshine (Hrs)
		Max.	Min.		
May	169.1	32.9	24.1	74.1	7.6
June	982.5	29.6	23.0	85.1	3.9
July	1339.7	27.9	22.6	89.2	2.3
August	779.1	28.4	22.7	87.2	3.7
September	294.2	29.9	23.0	81.9	6.1
October	188.0	31.3	22.7	77.5	5.7

*Mean values.

varied from 3255.3 mm (1973) to 5088.6 mm (1978) with an annual average of 3278.8 mm. Much of the rains are received during May-October; i.e. South West monsoon season, heavy rainfall being in June-August. During this period the maximum temperature is between 27.9-32.9°C and minimum 22.6-24.1°C. The humidity varied from 74.1%-89.2%, and sunshine hours 2.28-7.55 hr (Table 1.13) and these facilitate the rapid development of the pathogen (Coleman, 1910; Tucker, 1931). These conditions reach a peak in July.

In the year 1978, the *Koleroga* disease was very rampant and losses ranging from 50-90% were estimated in a number of gardens. When the weather data for this year was compared to the other years from the Station, it was found that in 1978 the favourable conditions for the disease existed for a prolonged period i.e. from May to September. The total rainfall in 1978 was 5088.6 mm, the highest in the last 10 years. The rains started

during the first fortnight of May itself, continued raining almost everyday in June, July and August months, thus leaving no time for the farmers to take up prophylactic spraying operation against the disease. Further the maximum temperature was less than 30°C (June-September) and the humidity was more than 80% throughout the season. The bright sunshine hours was also less in 1978 when compared to other years. These conditions might have favoured the rapid spread of the disease. Though many variable factors are involved in the disease development, and its spread, the weather data and the inoculum potential of the pathogen may be considered to predict the occurrence of the disease in susceptible hosts.

Resting spores and mycelium are the main source of inoculum which are present in dead parts such as dried bare bunches, leaves, diseased nuts and refuse in the garden. During dry season they survive on the upper layers of soil (Coleman, 1910; Nambiar, 1956). Areca palms may also possess latent infection in their crown during non-monsoon periods. Such trees are potent sources of primary infection (Kamat, 1956, Mundkur, 1967; Singh, 1973).

Various bushes and trees, notably *Bryophyllum calycinum* (Venkata Rao, 1925; Narasimhan, 1926, 1927) *Colocasia antiquorum*, *Ficus nitida*, *Jatropha glandulifera*, *Citrus medica*, *C. limonum*, jack, sandalwood, mango, rubber (Narasimhan, 1926, 1927; Ramakrishnan and Seethalakshmi, 1956b; Nambiar, 1956), and coconut (Sundararaman and Ramakrishnan, 1924; McRae, 1924; Gadd, 1927) harbour the fungus. Further, it was known that *P. arecae* on artificial inoculation could infect potato tubers (Rosenbaun, 1914; Tucker, 1931), apple fruits (Tucker, 1931), very young tomato (Coleman, 1910, Dastur, 1913) and brinjal seedlings (Coleman, 1910) as well.

Control

The earlier practice of controlling *Koleroga* comprises providing covers to arecanut bunches, made of either arecanut leaf sheaths called 'Kotte' in Malnad region or a kind of grass in other parts named 'Karada'. These covers, though expected, neither helped in preventing nor in eradicating the disease (Coleman, 1910, 1915; Anonymous 1954, 1956a; Krishnamurthy, 1955; Nagaraja Rao, 1960). Coleman (1910) was the first to recommend spraying of 1% Bordeaux mixture with resin-washing soda as an adhesive to control the disease. Various workers tested the efficacy of different adhesives and spreaders with Bordeaux mixture with good results (Narasimhan, 1923, 1924; Venkata Rao, 1925, 1926, 1927). Porash alum with casein called Martin's Bordeaux mixture (Narasimhan, 1928 a, b, 1931b; Venkata Rao, 1926, 1927, 1929), and vegetable oils, such as groundnut, gingelly, coconut or safflower oil (Narasimhan, 1931 b, 1934, 1935; Thomas, 1938, Thomas and Marudarajan, 1938, 1952; Patel and Nagaraja Rao, 1958; Nagaraja Rao, 1960) added to Bordeaux mixture protected the arecanut palms against *Mahali*. However, it was also shown that plain 1% Bordeaux mixture without any adhesive was equally effective in controlling the disease (Thomas and Marudarajan, 1938; Venkatarayan 1943; Marudarajan, 1950a, b, 1952; Marudarajan and Kalyana Subramanyam, 1948, 1952) and therefore, prophylactic sprayings with Bordeaux mixture alone two-three times a year, has been recommended and is in use ever since, (Sundararaman and Rama Krishnan, 1924; Thomas, 1938; Thomas and Marudarajan,

1938, 1952; Marudarajan, 1950 a, b, 1952; Marudarajan and Kalyana Subramanyam, 1948, 1952; Nambiar, 1956; Patel and Nagaraja Rao, 1958; Anonymous, 1926, 1954, 1956a, 1961, 1967, 1969b, 1972; Nagaraja Rao, 1960; Sannegowda, 1961; Seshadri and Rawther, 1968; Nambiar, 1971). Spraying campaign against *Koleroga* was undertaken in Karnataka on payment basis (Kulkarni, 1924) and an improved sprayer called 'Primus sprayer' was developed for the purpose (Narasimhan, 1938)

A number of other chemicals besides Bordeaux mixture were also tested against the pathogen. Among them, the mercurised copper oxychloride, and blitane inhibited the fungal growth in nutrient media (Rawther, 1969) whereas nickel chloride had no effect (Anonymous, 1963, 1964). Field trials revealed that copper oxychloride checked *Koleroga* (Anonymous, 1956b, 1960a, b, 1962b). However, Thomas (1938), Thomas and Marudarajan (1938, 1952) and Uppal (1942) recorded that they were not effective, and that they even caused copper injury to nuts at 0.5% concentration (Anonymous, 1967). Proprietary copper fungicides such as Fycol 8, Fycol 8E, Oleocop and Lovel sprayed with low volume sprayers too, could not protect the nuts (Ramamurthy, 1962, Rawther, 1969). The reported control of the disease from Adyanadka by spraying mud solution was far from truth (Anonymous, 1959).

Besides attempting to the protective sprays against the disease, it is also necessary to reduce the inoculum potential by adopting phytosanitary measures such as removal and destruction of fallen nuts, diseased bunches, tree tops and other plant parts in the field (Coleman, 1910, 1915; Anonymous, 1926, 1972; Kamat, 1953; Nambiar, 1971; Koti Reddy *et al.*, 1978). Efforts should also be bestowed to eliminate the alternate hosts of *P. arecae* from the vicinity of the gardens, popularisation of the above plant protection measures against *Koleroga* pays rich dividends. Above all, it is worthwhile to develop efficient forecasting systems for different agroclimatic conditions in our attempts to contain the disease.

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*Original not seen.

DISCUSSIONS

H. S. Sohi : The disease attacks the nuts as was pointed out by the speaker. I would like to know whether the foliage is also infected. If so, spraying of the crown is also essential.

Answer : Yes, it is being recommended and practised to prevent bud rot.

Abicheeran : What is the primary source of inoculum?

Answer : The sources of primary inoculum are infected fallen nuts and infected dried inflorescences.

Saturday, 20 September, 1980
10.30 a.m. — 1.30 p.m.

SESSION 2

EPIDEMIOLOGY AND FORECASTING

Chairman: **Dr. D. N. Srivastava**

Rapporteur: **Dr. Y. R. Sarma**

EPIDEMIOLOGY IN THE GENUS *PHYTOPHTHORA*

F. J. NEWHOOK

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It is inevitable that a paper on epidemiology in the genus *Phytophthora* should begin with reference to potato blight caused by *P. infestans*. Not only were the catastrophic outbreaks of the middle of last century the first *Phytophthora* epidemics to be studied, they led to the first conclusive proof that a micro-organism was the cause of a disease. There followed a period when study of pathogens received major emphasis. Later the host and its reaction to pathogens came to the forefront of research, with more and more sophisticated laboratory aids becoming available. Recognition of the role of environmental factors was slow to gain impetus but when it did, the concept of the 'disease triangle' became generally accepted. Investigations remained largely descriptive despite emphasis on measurement of the various parameters—temperature, rainfall, RH, wind, soil, moisture, etc.

However the growing interest in quantitative approach crystallised and was given a crucial boost in the 1960's by van der Plank. As Cowling and Horsfall (1978) say, 'quantitative thinking spread like an epidemic across the field of plant pathology'. Now we have an era of highly mathematical, theoretical plant pathology with computers playing a central role and providing the vehicle for an extension of abstract modelling into the realm of intricate crop/disease simulation.

My paper today is necessarily oriented towards the needs of this particular workshop—to provide the basis for an increased understanding of the topic of epidemiology with particular emphasis on practical aspects. We cannot all become immersed in the theoretical approach. Most of us will, of necessity, be concerned with real 'triangles' most of the time. But we must see our roles in perspective.

Simulation, and modelling in general, depend on:

1. good observation in determining detailed life cycles; pathologists will always be on the lookout for vulnerable stages towards which they can direct better control measures,
2. measuring the factors which are known or thought to influence epidemics at their various stages,
3. translating data into equations which can be handled by computer, and
4. using the results, for example in forecasting, predicting the efficiency of control measures and appraisal of crop losses.

Van der Plank (1960, 1963) pioneered the modern approach to epidemiology with his analysis of the factors which governed the progress of an epidemic with its now-familiar

S-shaped curve—rate of increase at first logarithmic and later slowing as the number of plants available for infection decreases. He stressed the importance of the earliest part of the curve, with incidence <5% or even 0.1%, after which progress is inevitable unless some major change in management or environment occurs. It is helpful to our understanding if we insert on to the classical curve a series of consecutive schematic pathogen life cycles within which each phase is susceptible to different environmental influences. (Kranz, 1974). In fact those single-generation cycles can each be expanded to the form presented by Gaumann in 1951 or further still, into a simulator diagram (Kranz, 1974) showing interactions of elements of the epidemic.

To study the subject further I recommend Whitney (1976), Horsfall and Cowling (1978a), Kranz (1974), Berger (1977), and Scott and Bainbridge (1978).

Meantime, whatever our individual role as plant pathologists, we are concerned to know more about environmental factors as they affect disease. Here, may I make a plea? Make sure that your measurements are as reliable as they can be in the appropriate context. Remember that they may be useful to people other than yourself, and may even play a key role as data for a simulator programme. To this end I commend you to consult Chapter 5 (Pennypacker, 1978) of 'Plant Disease Vol. 2' by Horsfall and Cowling (1978a), dealing with instrumentation for epidemiology. In Chapter 6 of the same textbook Horsfall and Cowling (1978b) deal effectively with measurement of plant disease and the relationship of intensity to loss. In these days of sophisticated instrumentation I cannot help singling out their remarks about the human eye—that it has tremendous precision, that it records logarithmically and that it records diseased tissue below 50% and healthy tissue above that intensity.

The genus *Phytophthora* is an appropriate choice for a discussion of epidemiology since within its range we see above and below ground pathogens, deciduous and non-deciduous sporangia, liberation of zoospores and direct germination, wide host range and degrees of specificity, prolific and sparse production of oospores.

To illustrate the range of epidemiological situations in *Phytophthora* I have chosen diseases associated with four species:

- P. infestans* – late blight of potato
- P. cinnamomi* – root rot of *Pinus radiata*
- P. colocasiae* – taro leaf blight
- P. palmivora* – black pod of cocoa

My research has given me personal acquaintance with the last three. I have chosen the first, late blight, because it is classical and the most intensively studied.

***Phytophthora infestans* – late blight of potato**

When late blight appeared for the first time in Europe, causing the historic and devastating epidemics of 1845 and 1846, *P. infestans* would have been meeting potato varieties that had never been exposed to the pathogen and so had undergone no selection pressure for resistance; the causal nature of the pathogen was unknown; the effective use

of chemical sprays was still in the future; deliberate breeding for resistance was even further away. The scale of the resultant social tragedy provided a dramatic background to the birth and development of our discipline, plant pathology.

One should recognise some landmarks. We know very well the roles of Berkeley and de Bary in establishing the causal nature of the pathogen. The year 1888 saw the first control trial, using Bordeaux mixture. Lutman (1911) was probably the first to demonstrate that weather had a major impact on blight epidemics. During the forties and early fifties Large (1952, 1955) provided a firm base for assessment of disease and epidemic development, and for forecasting. Beaumont (1947) devised a regional forecasting system for blight based on weather factors—minimum periods of high relative humidity (associated with rain) adjusted for various temperatures. Variations on his system are still in use, not only on a regional but also on a farmwise basis. Bourke (1957) preferred to use weather charts to predict arrival of weather patterns known to favour blight. Waggoner (1968) introduced systems analysis (mathematical modelling, simulation) to epidemiology, using late blight of potatoes. Currently, models can be written that take account of control by fungicides as well as weather.

As for the disease itself, the main sources of 'original' inoculum are infected 'seed' tubers (one per square km is sufficient to initiate an epidemic), infected volunteer plants, sporangia in wind-driven rain splash droplets. The life cycle of *P. infestans* is without a sexual phase and needing adaptation to allow for infection by direct germination of sporangia ($<18^{\circ}$) as well as by encysted zoospores ($>18^{\circ}$).

Neither potatoes nor the associated *P. infestans* are normally regarded as part of the tropical agricultural scene. However, the crop is extending its range and recent work in Israel by Rotem and coworkers has extended our knowledge (e.g. Hirst and Stedman 1960) of the importance of microclimatic differences within the crop canopy in relation to late blight. At least partly because of sensitivity of sporangia to heat and desiccation, there are important interactions between *P. infestans*, dew, overnight temperature and rain or overhead irrigation.

There is evidence that *P. infestans* is like some other *Phytophthora* spp., influenced by age of host or host organ. Care must be taken in assessment of this aspect to distinguish the roles of physical changes in the canopy and physiological changes in the plant.

P. cinnamomi* – root rot of *Pinus radiata

In 1953 and 1956, many thousands of mature trees died in groups in pine shelter belts in northern New Zealand. Mortality has been shown to be associated with massive attack by *P. cinnamomi* on feeder rootlets (Newhook, 1959). The disease has been chosen for discussion at this Workshop because it illustrates several epidemiological principles pertinent to the genus.

There is a strong interaction between soil moisture and temperature. The pathogen is present but tolerated in most years, with rootlet regeneration adequately balancing killed rootlets. In epidemic years there has been abnormally high, prolonged summer and

autumn rainfall that has encouraged massive infection of rootlets while soil has been $>15^{\circ}$, a temperature crucial for infection (Chee and Newhook, 1965). After cold wet winter conditions, adverse to rootlet regeneration, trees were unable to meet transpiration requirements, became defoliated and, in most cases, died. Survivors from the 1953 epidemic remained, thin-crowned until the 1956 outbreak which they withstood while vigorous, dense-crowned nearby trees succumbed.

In adjacent forest on infertile clay soils, 35-year-old *P. radiata* showed no reaction to the two epidemics. Trees had extremely sparse crowns analogous to those with little-leaf disease symptoms in southeastern USA (Hepting and Newhook, 1962) and shallow, sparse root systems. Trial plots receiving superphosphate 5 years previously had dense dark green crowns, deeply ramifying roots and prolific mycorrhizae. There had been no mortality in the forest equivalent to that in nearby shelter belts. Rationalisation of the differences between shelter belt trees with 'sudden death', littleleaf trees, and phosphate-treated trees with recovered crowns that are nevertheless greatly restricted by spatial competition, is possible on the grounds of relative transpiration demand with all trees denuded of rootlets. There were similar populations of *P. cinnamomi* in all three situations.

Significantly, soil beneath treated trees seldom reaches saturation because of increased interception of rain and more effective removal of excess soil moisture—thus providing conditions unfavourable for sporulation and zoospore movement of *P. cinnamomi*. We thus see epidemic loss and tolerance in a single host species, depending upon different states of balance in a complex but nevertheless identifiable set of biotic and abiotic factors (Newhook, 1970).

***P. colocasiae* – taro leaf blight**

Taro, *Colocasia esculenta*, in the Solomon Islands, is affected severely by a disease which has similarities to and differences from potato late blight (Gollifer, Jackson and Newhook 1980; Jackson, Gollifer, Newhook, in press). The pathogen affects stored tubers but these rot rapidly and in any case are not used for propagation, a role performed by 'tops' (tuber apices with trimmed petioles). Infection on the latter is short-lived but probably accounts for most transfer to new plantings. Inoculum in soil and on infected detached leaves remains viable for only a few days. Tropical storms with high winds probably account for even more widespread rainsplash dispersal than with potato blight. Obvious blight lesions are confined to leaf laminae which do not become infected until part of a blade becomes sufficiently horizontal (usually third from centre) to retain water despite the waxy cuticle. Allowing for incubation and secondary spread, this means that even at the height of an epidemic each plant cannot have less than effectively 3-4 healthy leaves. This fact inspired a counter-measure to the disease. Regardless of whether they became diseased or were kept healthy by spraying, plants set at closer-than-normal spacings retained 3-4 rather than 6-7 leaves because of lateral competition. Yields per hectare were equivalent to those from sprayed plants at normal spacings.

***P. palmivora* – black pod of cocoa**

In this disease the most prolific and obvious source of inoculum is diseased pods from

which propagules are washed or splashed by rain. Sporangia of *P. palmivora* are not wind-dispersed, dry as well as in water, unlike the situation in *P. infestans* (Hunter and Kunimoto, 1974). Leaf lesions are rare, developing mainly on 'flush shoots' and 'chupons'. Stem cankers are not uncommon, usually associated with flower 'cushions'. Transport by ants and rodents has been demonstrated. The pathogen can perennate on infected pods lying on the ground. In some countries, after a dry spell without disease, *P. palmivora* re-enters the canopy by step-wise progression on pods up the trunk, with initial infections quite obviously derived from the plantation floor. The pathogen can be shown to perennate in soil, although often the ground is so covered with a carpet of fallen cocoa leaves that rain-splash of infected soil would be rare. In Solomon Islands however we have isolated *P. palmivora* readily from debris (including leaflets of the shade tree *Leucaena leucocephala*) retained on cocoa leaves on the plantation floor (Newhook and Jackson, 1977). On the other hand it is rare to find pods low down on trunks and the 'stepping stone' phenomenon is seldom seen. The black pod fungus infects and sporulates on *Leucaena* leaflets caught up on cocoa canopy leaves, providing an additional, unexpected source of inoculum within rain-splash distance of pods (Jackson and Newhook 1978). There are thus sufficient sources to explain why regular, 2-3 day harvesting of infected pods does not halt an epidemic. Local alternative hosts for *P. palmivora* have not been identified in Solomon Islands. Pioneer infections in newly established plantations have been associated with rat damage to pods and with transport of inoculum on pruning tools.

After studying epidemiology in the genus *Phytophthora*, one can justifiably comment that, through its versatility and adaptation to innumerable environmental situations, it has earned its name, translated from the Greek as 'plant destroyer', and its reputation as one of the worlds' most notorious genera of plant pathogens.

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DISCUSSIONS

D. N. Srivastava : Dr. Newhook has given us a clear account of soil borne *Phytophthora* disease and how the root regeneration vis-a-vis root rot has a profound impact on the health of the plant under pathogenesis.

Y. R. Sarma: Has the application of phosphorus suppressed *Phytophthora* populations?

Answer : Supplementing P ensured greater root generation and abundant mycorrhizal development. This enhanced the health of the plant.

FIELD STUDIES ON LATE BLIGHT (*PHYTOPHTHORA INFESTANS*) OF POTATO

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INTRODUCTION

Late blight is perhaps the most important, and certainly the most widely studied of the diseases affecting the potato and is caused by a *Phytophthora* viz. *Phytophthora infestans* (Mont.) de Bary. Given suitable weather the fungus can attack and destroy foliage at any stage of crop growth and because the disease develops from initially discrete lesions the rate at which foliage is destroyed depends largely on how soon every leaf contracts an infection. Premature destruction of foliage results in decreased yields and the infection of progeny tubers during growth or at lifting can further decrease marketable yield. Tubers continue to rot in store and where conditions remain damp this may lead to the bacterial breakdown of surrounding 'healthy' tubers.

Epidemics develop only in wet weather and progress most rapidly when rain is frequent. Unlike Central America e.g. Mexico, where the oospore occurs naturally in soil, epidemics in the rest of the world develop from spread of the asexually produced sporangium. The fungus overwinters in potato tissue as mycelium and spreads as sporangia to new crops mainly from infected seed tubers, from dumps of discarded tubers or from volunteer potato plants. Persisting leaf or tuber wetness is required for successful infection which may occur from the direct germination of the sporangium, when temperatures exceed c. 18°C, but most frequently from the release of zoospores, which swim for a period, encyst and penetrate the host via appressorium and infection peg.

The disease can be successfully controlled by regular fungicide treatment applied to the foliage and much research effort has gone into the timing and frequency of spray applications, fungicides and formulations and the weather conditions favouring spread and epidemic development. In the United Kingdom dithiocarbamate fungicides like mancozeb, zineb and maneb are most commonly used with the organo-tin fungicides based on fentin hydroxide and fentin acetate increasingly used as late-in season sprays to give some control of tuber infection. Most recently chemicals of the acylalanine type have been introduced, reputedly with systemic and possibly curative activity and these are currently marketed combined with a surfacting protective chemical like mancozeb. Unfortunately chemical methods of control are costly in labour machinery and materials and therefore for developing countries the breeding and introduction of resistant cultivars remains their best hope for control. It was in connection with the assessment of varietal resistances and the value of multigenic or 'field' resistance in breeding programmes that work was started at Rothamsted and interest in field epidemics developed. Some of the simple

field techniques adopted for this work are described in the hope that some might prove of use in the study of tropical and sub-tropical *Phytophthora* diseases.

TECHNIQUES

1. Tagging of leaves

Plots of the cultivars to be assessed were set out in Latin Square design and within a plot each of 10 plants had a stem selected and 5 leaves, representing different heights within the crops canopy, tagged with differently coloured plastic covered wire; a particular colour was made to represent a particular leaf position. Frequent visits (depending on the stage of the epidemic but at least once a week) to plants with tagged stems enabled the date and position of first infections to be recorded on 5 of the leaflets making up a potato leaf and their subsequent death. Using this simple method it was possible to show that in the same epidemic cultivars differed in the rate they contracted infection and in the time taken for their destruction after infection. It also showed the importance of lesion position in determining and speed of leaf destruction and how the disease progressed through the crop canopy.

2. Covering soil with plastic film

This technique was adopted in experiments to relate haulm destruction with tuber infection. The tagged leaf method is too slow for routine assessment of haulm (leaf) destruction and so a disease key devised by the British Mycological Society was used weekly to give a quick overall visual estimate. The key has descriptions for 0.1, 1, 5, 25, 50, 75, 95 and 100% disease e.g. 1% is 1 or 2 lesions in a 10 metre radius, 1% about 10 lesions and 5% about 50 lesions, per plant. Each week, plants were dug ('normal' sample) and inspected for small 1mm necrotic lesions which would be evidence of fresh infection. At the same time plants in an adjacent row had the haulms removed and soil of the potato ridge covered with a plastic sheet to prevent further infection (from sporing lesions on leaves or stems) from taking place ('incubated' sample). If 1 mm necrotic lesions were found, when lifted 5 days later on the incubated and not the normal sample then it was assumed that infection must have taken place prior to covering. Using this method it was possible to detect the rain most likely to have been responsible for the tuber infection. The frequency of spread was found similar in different cultivars but the amount of tuber infection following a spread occasion differed. With all cultivars as little as 1-5% blighted haulm could result in tuber infection.

3. Daily disease and field observations and use of glasshouse grown plants

In order to determine more precisely the weather and soil conditions necessary respectively for successful infection of leaves or tubers, various extra parameters need to be recorded, many of them daily.

(a) Leaves. Daily inspection of the crop for presence of 1mm necrotic lesions on leaves and an assessment of their abundance (evidence of fresh spread of disease). The amount of sporulation on lesions approximately 1cm diameter (necrosis) viz. width (mm) of sporing annulus surrounding the necrotic centre and a score for intensity of sporulation, was also recorded.

(b) **Tubers and Soil.** Tubers of plants with diseased haulm were dug daily and were inspected for 1mm necrotic lesions. Sample cores of soil were taken from the ridge surface and from among the tubers (by cutting a face at right angles to the ridge to expose the tubers) and used to assess soil moisture from oven dry weight and fungal activity by spreading a quantity on the cut surface of tuber slices to bait out the fungus. By cutting slices into octants it was possible to give a crude quantitative measure of soil infectivity.

(c) **Potted glasshouse plants.** Freshly produced glasshouse plants were exposed from early morning in a blighted area of the field for 24 hours when they were returned to an isolated glasshouse and incubated under normal glasshouse conditions to see if lesions developed. In this way weather which had proved suitable for infection could be detected. Every evening a set of glasshouse plants were sprayed with inoculum and incubated in a humid chamber overnight before placing out in the field until 1mm lesion were recorded. The probable infection date of lesions on field plants could then be estimated from the date of inoculation of the potted plant showing similar lesion sizes. A similar dating procedure was adopted for tubers which were inoculated by spraying, leaving them in moist conditions overnight before continuing the incubation in the potato ridge.

The results from such detailed observations showed that (i) a source of inoculum on leaves (sporulation records) or in soil (infectivity assessments) was seldom a limiting factor for fungal spread (ii) infection of leaves seemed to occur more frequently of rain and tuber infection to the amount. This is not surprising, for, leaves can become infected during persisting dew, whereas for tuber infection inoculum may have to be transported by rain to and through the soil and then there must be sufficient soil moisture (near saturation) for a water film to persist at the tuber surface long enough to ensure infection.

Using potted plants to detect leaf infection it was possible to compare the weather conditions (temperature, humidity, rainfall etc.) when 'spread' or 'no spread' occurred. At Toluca, for example, 'no spread' days were those with very little or no rain and when humidity did not reach saturation (dew forming on leaves) until after midnight, whereas on 'spread days' some rain had fallen during the day and dew had formed well before midnight.

In order to forecast the amount of tuber infection (assuming no fungicide is used) it is necessary to compare epidemics over a number of years. When this is done it becomes clear that the date and rate of the foliar epidemic can vary greatly from year to year as well as the amount of tuber infection. No single factor seems to determine the amount of tuber blight but the best relationship seems to be with the total amount of rain between 0.1% and 75% defoliation.

4. Other techniques

Water is of fundamental importance for the spread of late blight and therefore various simple techniques were devised to show (i) how water was shed by the crop canopy on to the soil (a set of aluminium gutters set at different positions up the sides of the potato ridge and draining into collecting (conical) flasks (ii) what proportion of the rain ran down stems (a simple aluminium trap was wound around a stem and a plastic tube led from

it to the collecting flask) and (iii) if spores were carried in the water running down stems (the collecting flask replaced by a container (a patent coffee jar) in which a half tuber was suspended in a muslin sling and the plastic tube from the stem trap led through the jar lid to drip water on to the cut tuber surface; after exposure the tuber was incubated in the laboratory to see if sporulation occurred).

Thus it is possible to gain much information from simple and inexpensive pieces of equipment and hopefully some of the methods described might help in the study of other *Phytophthora* diseases.

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DISCUSSIONS

D. N. Srivastava: This is a *Phytophthora* which causes major foliar damage, which is again controlled by the climatic factors in a given locality.

EPIDEMIOLOGY AND FORECASTING OF *PHYTOPHTHORA INFESTANS* INCITING POTATO LATE BLIGHT

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ABSTRACT

Forecasting systems for late blight of potato, caused by *Phytophthora infestans* (Mont.) de Bary, developed by Beaumont (1947), Wallin and Hoyman (1954), Cook (1949), Hyre (1954) and Hyre and Bonde (1955) were tested for their suitability in the Indian Hills (Simla, Shillong and Ootacamund). The success of late blight predictions by the above mentioned methods was limited under our conditions. Therefore, a correlation was worked out between the weather data and the actual date of appearance of the disease under Simla (15 yrs), Shillong (7 yrs) and Ootacamund (5 yrs) conditions. It was established that (i) if the 7-day moving precipitation (30 mm for Simla, 28.9 mm for Ootacamund and 38.5 mm for Shillong which were observed to be the critical rainfall lines) associated with mean temperature of 23.9 C or less continues for 7 consecutive days, the potato late blight would appear within 3 weeks and (ii) if hourly temperature ranges within 10-20°C and associated with high RH (80% or more) for continuous 18 hrs for two consecutive days, the potato late blight would appear within a week. Accordingly a warning service for spraying fungicides on potato crop against late blight has been initiated in the North Indian hills and Nilgiris.

INTRODUCTION

Late blight of potato incited by *Phytophthora infestans* (Mont.) de Bary, appears in a regular epiphytotic form in the Indian hills and sometimes in the northern plains causing 20 to 70% loss in yield (Dutt *et al.*, 1978). Fungicides effective in controlling the disease are solely prophylactic and their efficacy mainly depends upon their judicious and timely application. In order to achieve the best results, development of a disease outbreak forecasting system is very essential.

In many countries successful attempts have been made to evolve reliable forecasting systems for late blight. Based on the environmental factors such as rainfall, dew point, temperature and cloudiness, Van Everdingen (1926) first identified the conditions, known as the "Dutch Rules", for prediction of late blight in Holland. Dutch rules were found usually satisfactory but sometimes the blight appeared even when the Dutch rules were not completely fulfilled. Beaumont and Stainland (1934, 1938) modified the Dutch rules for England and evolved "Beaumont rules" (Beaumont, 1947) based on minimum temperature of 50°F or more and relative humidity of 75% or above for two consecutive days. Subsequently Bourke (1953), Johnnes (1953), Wallin and Hoyman (1954) and many other workers reported correlation of different temperature and relative humidity periods for prediction of late blight in different parts of the world.

Cook (1947, 1949) in the United States used 7-day moving average total rainfall and average mean temperature charts for blight forecast. Hyre (1954, 1955), and Hyre and Bonde (1955) reported 10-day moving graph method to be more accurate than 7-day moving graph (Cook, 1949) which was also supported by Choudhuri and Pal (1959) under Darjeeling hill conditions in India. Thus, criteria for late blight forecasting differ from region to region. Accordingly temperature, rainfall and relative humidity were studied in relation to late blight appearance in three important potato growing hilly regions in India and the findings are summarized and discussed below.

MATERIALS AND METHODS

Disease trap-nurseries, with a susceptible potato cultivar Kufri Chandramukhi were raised from 1974 to 1978 to study the climatological factors for disease development with six replicates of 36 plants each. The nurseries were located around the meteorological observatory at Central Potato Research Institute (CPRI), Simla and Central Potato Research Station (CPRS), Ootacamund. The appearance and build up of the disease were recorded in the trap-nurseries and other fields in the nearby localities also. Records of daily temperature, RH, rainfall and the date of appearance of the disease maintained at CPRI, Simla, CPRS, Shillong and Ootacamund from 1964, 1972 and 1974 onwards respectively, were analysed individually.

Hygro-thermograph records on temperature and RH were taken daily from 15th May to 30th September at Simla and 15th April to 30th November at Ootacamund as late blight was never recorded before 7th June at Simla and 9th May at Ootacamund. The hygro-thermographs were available from 1975 for Simla and 1976 for Ootacamund.

The temperature, rainfall and RH data were correlated with the actual date of appearance of the disease based on the following methods.

1. **Beaumont (1947) method** : This method allows a forecast for an outbreak of late blight within three weeks after the first Beaumont period i.e., 48 or more consecutive hours with a minimum temperature not less than 50°F and RH not below 75%, following a suitable starting or zero date.
2. **Wallin and Hoyman (1954) method** : A late blight favourable period consists of 10 or more consecutive hours of temperature 75°F or less and RH 90% or more and if during a given 7-day interval there was at least one such period with the following days, maximum temperature 90°F or less. The more frequent occurrence of the favourable periods, the greater was the expected blight development.
3. **Cook (1949) and Hyre (1954) method of moving graph** : A favourable period occurs within 15 days when the moving 7-day total rainfall is continuously above the critical line and the 7-day average mean temperature is below 75°F. The critical rainfall line is determined by taking the average rainfall of the total crop season.

4. **Hyre and Bonde (1955) method of moving graph** : This is a modification of Cook's (1949) and Hyre's (1954) method of 7-day moving graph in which a 10-day total rainfall and moving 10-day average mean temperature are considered suitable. In this case also the critical rainfall line is calculated on the basis of total crop season as above.

OBSERVATIONS

Simla : The crop season is from May to September. The average rainfall during the period is 1195 mm and 7-day and 10-day average total rainfall is 53mm and 75.75 mm respectively. The yearly average rainfall is 1560.75 mm and 7-day and 10-day average total rainfall are 30 mm and 42.75 mm respectively.

Shillong : There are two crop seasons. The summer crop season is from March to July. The average rain during this period is 1254.25 mm and the 7-day and 10-day average total rainfall is 57.4 mm and 82.0 mm respectively. The yearly average total rainfall is 2018 mm and 7-day and 10-day average total rainfall is 38.5 mm and 55.0 mm respectively.

Ootacamund : In this area the potato is grown round the year due to equinox condition; yet the two important crop seasons are summer and autumn extending from April to August and August to December. The average rainfall during the summer season is 746.25 mm and during autumn is 683.25 mm. The average 7-day and 10-day total rainfall for summer is 34.2 and 48.8 mm and for autumn 31.4 and 44.8 mm respectively. The average annual rainfall is 1508 mm and the 7-day and 10-day average total rainfall is 28.9 and 41.3 mm respectively.

Comparison of the four methods

1. **Beaumont (1947) method** : When Beaumont's period was taken for forecasting late blight at Simla and Ootacamund it was observed that the conditions were fulfilled in 1977 and 1978 at Simla and for summer crop of 1977 at Ootacamund i.e., only 37.5% success. Out of these successful years it had only two days in 1978 at Simla and 5 days in 1977 at Ootacamund (Table 2.1) left to the farmers to get themselves prepared and spray their crops against late blight. In other years the Beaumont period appeared either a day earlier, on the same day or after the appearance of the disease. Therefore this system was found unsuitable for successful forecasting of the disease.

2. **Wallin and Hoyman (1954) method** : At Simla, this period appeared once in 1975, 4 days earlier, twice in 1976, the first being 31 days earlier and the second one 6 days before; four times in 1977, the first one being 25 days earlier and then 6 to 7 days intervals with the last one being 3 days earlier, and once in 1978, 7 days earlier (Table 2.1). If the first period is taken into account for forecasting, it is not successful for 1976 and 1977 and if the subsequent period is taken into account, no such subsequent period appeared in 1975 and 1978 after the first period before the appearance of the disease. At Ootacamund (Table 2.1), this period appeared once in 1976 both for summer and autumn crops, 8 and 21 days earlier, once for summer crop of 1977, 21 days earlier and two times for the autumn crop of 1977, the first being 21 days and the second 13 days earlier. There-

Table 2.1. Forecasting late blight in Indian hills by different methods

Place	Year	Date of 1st record of late blight	Dates of blight forecast by different methods		
			Beaumont (1947 method)	Wallin and Hoyman (1954) method	18 hr temp. and RH method
Simla	1975	July 14	July 14	July 10	July 11
	1976	July 15	July 14	June 14; July 19	June 18; July 13
	1977	July 2	June 19; July 2	June 7; June 13	June 28
	1978	June 28	June 22	June 23; June 29 June 21	June 26
Ootacamund Summer					
	1976	May 20		May 12	May 12
	1977	May 9	May 4	April 17	May 3
Autumn					
	1976	Oct. 11	Sept. 10	Sept. 18	Sept. 27
	1977	Sept. 1	Sept. 22	Aug. 10; Aug. 20	Aug. 24

fore except for the summer crop of 1976, the conditions were the same as that in Simla and this method also cannot be safely relied upon towards forecasting of the disease.

3. **Cook (1949) and Hyre (1954) method of moving graph :** By this method successful forecasting could have been done for 8 years (1966, 1967, 1968, 1969, 1971, 1973, 1974 and 1975) out of the 15 years at Simla (Table-2.2); 3 years (1973, 1977 and 1978) out of

Table 2.2. Comparison of the 10 and 7-day moving graphs for forecasting late blight in Simla hills taking the critical rainfall line on the basis of crop season and whole year

Year	Date of appearance of late blight	Dates of blight forecast based on moving graphs			
		Hyre & Bonde's 10-day moving graph (critical rainfall based on crop season 75.7 mm)	Cook's/Hyre's 7-day moving graph (critical rainfall based on crop season 53 mm)	10 day moving (critical rainfall based on whole year 42.75 mm)	7 day moving (critical rainfall based on whole year 30 mm)
1964	July 17	July 17	July 16	July 9	July 6
1965	July 16	July 26	July 22	July 20	July 16
1966	June 29	July 1	June 23	June 26	June 21
1967	July 13	July 11	July 4	July 5	July 2
1968	July 6	July 19	June 14	June 20	June 14
1969	July 21	July 10	July 7	July 7	July 3
1970	July 5	July 27	July 23	June 26	June 23
1971	June 7	June 15	May 20	May 23	May 20
1972	July 16	July 16	July 13	June 30	June 27
1973	June 30	June 27	June 21	June 23	June 20
1974	July 15	July 2	June 28	June 29	June 14
1975	July 14	July 23	June 30	July 2	June 27
1976	July 15	July 22	July 17	June 20	June 14
1977	July 2	July 18	July 13	July 16	June 15
1978	June 28	July 5	July 2	July 1	June 19

7 years at Shillong (Table 2.3); and in all the years except the autumn crop of 1977 and summer crop of 1978 at Ootacamund (Table 2.4). Therefore its success was only 61.3% under the hill conditions of Simla and Ootacamund and only 43% at Shillong.

4. **Hyre and Bonde (1955) method of moving graph** : Forecasting was possible for 4 years (1967, 1969, 1973 and 1974) out of 15 dyears at Simla (Table 2.2), for 2 years (1973 and 1977) out of 7 years at Shillong (Table-2.3) and for all years at Ootacamund except for 1976, for autumn crop of 1977 and summer crop of 1978 (Table-2.4), indicating its success to be limited to the tune of 35.5%.

Table 2.3. Comparison of 7- and 10-day moving graphs for forecasting late blight in Shillong hills on the basis of critical rainfall line

Year	Date of appearance	Dates of forecast based on moving graphs			
		Cook's and Hyre's 7-day moving graph (Critical rainfall based on crop season 57.4 mm)	Hyre's and Bonde's 10-day moving graph (Critical rainfall based on crop season 82.0 mm)	7 day moving graph (Av. yearly rainfall as critical 38.5 mm)	10-day moving graph (Av. yearly rainfall as critical 55.0 mm)
1972	May 19	May 27	June 25	May 15	May 30
1973	May 18	May 1	May 7	May 1	May 4
1974	May 10	May 23	May 26	May 6	May 12
1975	May 25	June 15	—	May 20	May 23
1976	May 26	May 28	June 19	May 23	May 27
1977	May 16	May 8	May 12	April 28	May 8
1978	May 15	May 8	May 15	May 8	May 12

Therefore, all the above mentioned methods have not been found suitable for forecasting late blight in the hilly areas in this country. Hence, modifications were made on the method of Cook (1949), Hyre (1954) and Hyre and Bonde (1955). The critical rainfall line was taken on the basis of yearly average instead of crop season. In another method, temperature and relative humidity were examined for their reliability in late blight forecast. The details of these methods and their efficacy are described below:

1. **10-day moving graph with 10 days total rainfall on the yearly average as critical line; modification of Hyre and Bonde method (1955):**

Under this method, successful forecast could have been done for 12 years (1964, 1966 to 1976) out of 15 years at Simla (Table 2.4), 4 years (1973, 1975, 1977 and 1978) out of 7 years at Shillong (Table-2.3) and for the summer and autumn crops of 1974, 1975 and summer crop of 1977 but not for 1976 and summer crop of 1978 at Ootacamund (Table 2.4), indicating 67.7% success which is all so not satisfactory.

2. **7-day moving graph with 7-day total rainfall of the yearly average as critical line: modification of Cook (1949) and Hyre (1954) methods:**

This method was successful for 14 years out of 15 years at Simla (Table 2.2) excepting 1965 wherein these conditions fulfilled on the day of blight appearance. Except for 1974

and 1976, the general forecasting dates ranged from 10-19 days with an average of 14.6 days i.e., 2 weeks.

At Shillong (Table 2.3) the dates of forecasting closely coincided with the actual date of blight appearance for all the years (1972 to 1978) and the days available to the farmers ranged between 4 to 18 days with an average of 9.6 days.

Under Ootacamund conditions, except in 1978, the forecasting could have been made successful for all the years (Table 2.4). In 1978 the favourable conditions appeared on the date of the disease appearance. The forecasting dates ranged from 6 to 34 days earlier to the appearance of the disease with an average of 18.6 days.

Table 2.4. Comparison of 7- and 10-day moving graphs for forecasting late blight at Ootacamund hills on the basis of critical rainfall line of crop season and whole year

Crop season/ year	Date of appearance	Dates of forecast based on moving graphs			
		Cook's or Hyre's 7-day moving graph (critical rainfall based on crop season 34.2 mm)	Hyre and Bonde's 10-day moving graph (critical rainfall based on crop season 46.8 mm)	7-day moving graph (critical rainfall on yearly Av. 28.9 mm)	10-day moving graph (critical rainfall on yearly Av. 41.3 mm)
Summer Crop					
1974	June 7	May 17	May 21	May 6	May 20
1975	June 6	May 12	May 11	May 3	May 10
1976	May 20	May 18	May 21	April 29	May 21
1977	May 9	May 3	May 6	May 3	May 6
1978	June 7	June 21	June 25	June 5	June 25
Autumn Crop		(31.44 mm)	(44.8 mm)		
1974	Oct. 23	Sept. 11	Sept. 14	Sept. 11	Sept. 14
1975	Oct. 10	Sept. 22	Sept. 26	Sept. 22	Sept. 26
1976	Oct. 11	Aug. 31	Oct. 14	Aug. 31	Oct. 14
1977	Sept. 1	July 27	July 30	July 27	July 30

Success of this method was found to be 93.6% and it seems to be the most reliable one with adequate time available for the farmers for taking up timely fungicidal sprays, though sometimes the spraying may be too early and the efficacy of most of the fungicides will be lost after 10 to 15 days of their spraying.

Therefore another method was worked out based on temperature and relative humidity records as described below:

3 18-Hrs method of humidity and temperature :

Under this method an 18 hr continuous temperature between 10-20°C associated with relative humidity of 80% or more for two consecutive days was taken as the favourable

period for blight This period appeared once each in 1975, 1977 and 1978 (3 days earlier) and twice in 1976, 26 and 2 days earlier under Simla conditions (Table-2.1) In 1976, the first blight appearance was recorded on June 24 at Fagu (about 8 aerial km away from Simla) i.e., 6 days after the first favourable period. Then the weather conditions became dry and the disease could not be recorded at Simla and its appearance was observed on July 15th, i.e., 2 days after the second favourable period at Simla.

Similarly under Ootacamund, this period appeared once for all the crop-seasons with 8 days, 14 days, 6 days and 7 days earlier respectively.

Therefore this method was found to be the most suitable as only one period could be observed before the appearance of the disease, despite the fact that the gap between the date of the disease appearance and date of forecasting in some cases was too short.

DISCUSSIONS AND CONCLUSIONS

Under Indian hills conditions, rainfall is the primary factor for generation of conducive conditions for late blight. However, the critical rainfall line as determined by Cook (1949), Hyre (1954) and Hyre and Bonde (1955) taking the rainfall of the crop season does not work successfully. 7-day moving graph derived on the basis of yearly average rainfall, worked better in all the three hill regions under report (Table 2.2 to 2.4).

The 18-hr method of temperature and relative humidity developed was found to be the most reliable, both for Simla and Ootacamund (Table-2.1). This period satisfied the dew point difference of $< 30^{\circ}\text{C}$ for two consecutive nights and was the most congenial for disease development, spread and infection (Post, 1957). Our results also confirmed that plants exposed from 5.00 p.m. to 9.00 a.m. gave the first infection. This period was required for two successive days as observed in the detailed laboratory experiments by de-Weille (1963). Therefore, consecutive 18-hr periods for two days with RH 80% or more was required for the pathogen for sporangial formation, maturation, dispersal and infection. By employing this method, 100% success was achieved in forecasting the late blight both in Simla and Ootacamund conditions though sometimes the gap between the date of forecasting and date of actual appearance of the disease was too short, but was sufficient to take up immediate spray. The period of forecast was sometimes too long to warrant fungicidal sprays but was good enough to advise the farmers to keep the spray machines and chemicals ready. A double warning system is, therefore, proposed in which the first warning for making preparations for spray is given on the basis of 7-day moving graph and the second warning for taking up immediate sprays on the basis of 18-hr temperature and relative humidity period.

Using this system the first forecast to the cultivators was given on 19th June, 1978, 29th June, 1979 and 27th June, 1980 and the final forecast was given on 22nd June, 1978, 13th July, 1979 and 11th July, 1980. The disease was observed on 29th June, 1978, 17th July, 1979 and 16th July, 1980 at Simla and the farmers could protect their crop effectively.

With the success of this system a regular forecasting service has been developed through the local All India Radio in these three hilly regions.

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THE OCCURRENCE OF A¹ MATING TYPE OF *PHYTOPHTHORA COLOCASIAE* RACIB. IN NORTHERN INDIA

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ABSTRACT

Phytophthora leaf blight of taro (*Colocasia antiquorum* var. *esculenta* (L.) Schott) has become a limiting factor in its cultivation. Diseased plant material from each field was thoroughly searched for oospores in nature. The mating type of all isolates of *P. colocasiae* was determined by intraspecific and interspecific pairings with A₁ and A₂ mating types of *P. colocasiae* and *P. palmivora* respectively on chick-pea extract sucrose agar (CPA). In addition, isolates were grown alone and paired among themselves on CPA and French-bean agar. Oospores have not been observed in or on the naturally infected leaves. All the isolates collected from three states of North India failed to form oospores consistently in single cultures and in pairings among themselves on CPA and French-bean agar, but did so when paired intra- and interspecifically with A₂ mating type only on CPA and not with A₁ mating type. Thus the fungus under investigation is a heterothallic species with only A₁ mating type in Northern India and it remains to be seen whether A₂ type occurs elsewhere in other parts of India. Our findings are contradictory to Butler and Kulkarni's (1913) report which recorded abundant oospore production in some of the cultures.

INTRODUCTION

Taro or arvi (*Colocasia antiquorum* var. *esculenta* (Linn.) Schott) an important summer vegetable crop, grown in many parts of India and elsewhere, usually suffers from *Phytophthora* leaf blight during the monsoons. The blight becomes severe during cloudy and humid weather conditions resulting in considerable crop losses in terms of green foliage and corm yield. The disease has become a menace for taro cultivation in tropical and subtropical regions.

Phytophthora colocasiae is a highly specialized foliar pathogen and has a very limited host range. Many, if not all, species of *Phytophthora* are potentially homothallic but functionally heterothallic, as substances from sources other than *Phytophthora* have been shown to stimulate oospore production in heterothallic species (Desai, 1950; Zentmyer, 1952; Mukerjee and Roy, 1962; Brasier, 1975; Suzui *et al.*, 1978; Kamjaipai and Ui, 1978).

This paper reports the mating types in *P. colocasiae* hitherto unrecorded from north India.

MATERIALS AND METHODS

The infected *Colocasia* leaves were collected from different fields in Haryana, Punjab and Uttar Pradesh during July-September, 1979. Diseased material was carefully exa-

mined for oospore formation in nature in the host tissue. Multiple isolations of *P. colocasiae* were made on Potato-dextrose-agar medium. All the isolates were paired among themselves and mated intra- and interspecifically with both mating types of *P. colocasiae* (PCO-17: mating type A¹, PCO-1; mating type A²) and *P. palmivora* (P-549: mating type A¹, P-255: mating type A²) respectively. The isolates were grown on chick-pea extract sucrose agar (CPA) (250 g Chick-pea, 20 g Sucrose, 18 g agaragar and 1000 ml distilled water). In addition, isolates were also cultured individually on CPA and French-bean agar (FBA) (100 g dried French-beans, 18g agar and 1000 ml distilled water). Inoculated petri-dishes were incubated at 23°C ± 1°C for a period of 10-15 days in darkness. They were observed for oospore formation through the reverse side of the plate under low power (10x10) of the microscope.

RESULTS AND DISCUSSIONS

Oospores were not observed in or on the naturally infected plant materials during the course of the present investigation. All isolates of *P. colocasiae* were self-sterile and never produced sex-organs either in single strain cultures or among themselves on CPA and/or FBA. It indicates that the pathogen is not homothallic and host or FBA is not providing any stimulus for oospore production. It is contradictory to Butler and Kul-karni's (1913) report that oospores were produced in considerable numbers in some of the cultures, which all belonged to the same strain i.e. arisen from the same original parent, hitherto only in French-bean agar. They continued to develop in sub-cultures over a period of six months, whether the culture was grown for several generations on this medium or one or more generations on other media such as Corn-meal or Prune juice agar and again transferred to French-bean agar.

Oospores with persistent amphigynous antheridia were formed in abundance at the mycelial juncture of two colonies when any one of the isolates was crossed with complementary A² mating type of *P. colocasiae* and *P. palmivora*, but not when reacted with A¹ mating type. Oospores were most abundant in crosses with A² mating type of *P. colocasiae*. In pairings with A¹ mating types or among themselves the repulsion of mycelium was observed. These behaviours reveal that all isolates of *P. colocasiae* collected from north India are of A¹ compatibility type. It remains to be seen whether A² mating type of this fungus occurs elsewhere in India.

Our experimental results agree with those of Ko (1979) who reported that all 101 isolates of *P. colocasiae* from the island of Hawaii, 8 isolates from the island of Maui and 5 isolates from the island of Kauai were of A¹ mating type. However, all five isolates viz. N 224 from CBS, N315 from CMI (IMI 73480), N470 and N471 from L.B. Throrer University of Hongkong, Hongkong and N472 from O.S. Peries, Rubber Research Institute of Ceylon, Agalawatta, of *P. colocasiae* were of A² mating type (Savage *et al.*, 1968).

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Yu of University of California, Riverside, U. S. A. and Institute of Botany, Academia Sinica, Taipei, Taiwan, respectively, for providing us the authentic A¹ and A² mating types of *P. palmivora* and *P. colocasiae*.

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DISCUSSIONS

- T. N. Sreenivasan : How could you identify the new mating type of *Phytophthora*?
- K. L. Narula : With the standard A¹ and A² mating type cultures received from abroad.
- T. N. Sreenivasan : This importing of new mating types into the country should be avoided because of the possible hazards of development of new races if the cultures are released into the new area while handling them in the lab.
- K. L. Narula : Utmost care is taken to avoid such mistakes and cultures are autoclaved once the work is over.

INCIDENCE OF FOOT ROT AND WILT OF BETEL VINE IN ANDHRA PRADESH

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ABSTRACT

Betelvine (*Piper betle*) is grown in Andhra Pradesh in about 3000ha. The important diseases affecting the crop are foot rot and wilt (*Phytophthora* sp.), Sclerotial wilt (*Sclerotium rolfsii*), anthracnose (*Colletotrichum* sp.) and root rot (*Rhizoctonia* sp.) Of the above, foot rot and wilt was a common disease restricted to Tuni, Payakaraopet and Vizianagaram areas where the gardens are of closed type and sclerotial wilt was the predominant one in Ponnur area of Guntur District where the gardens are of open type.

During January, 1980, outbreak of a "mystery disease" causing extensive wilting and drying of betelvine gardens of Ponnur area of Guntur District was reported in the press. Preliminary investigations revealed that the so called "mystery disease" was only foot rot and wilt due to *Phytophthora* sp. Besides, *Fusarium* sp., *Rhizoctonia* sp., and *S. rolfsii* were also isolated. *Phytophthora* foot rot and wilt was never reported earlier from these areas and the damage was extensive and quite a few gardens have been abandoned.

The disease was more severe in heavy soils and was low to nil in light soils. Enquiries revealed that the disease was also observed in some gardens during 1977 and 1968 to some extent. The absence of any concerted control measures contributed to a general build up of inoculum which eventually flared up as an epidemic in 1980. It is suggested that detailed studies deserve to be taken up on a priority basis not only on etiological side but also on the control of the disease through an economic fungicidal schedule, soil amendments, varietal resistance, etc.

INTRODUCTION

Betel vine (*Piper betle* L.) is one of the important commercial crops of Andhra Pradesh and is grown traditionally in about 3,000 ha, mainly in the districts of Srikakulam, Visakhapatnam, East Godavari, Guntur, Krishna, Nellore, Cuddapah and Chittoor. The gardens in the State are usually raised in sandy loam, sandy clay and to a lesser extent in heavy clay and alluvial soils. In the districts of Guntur, Nellore, Cuddapah and Chittoor, the gardens are of open type while those in Visakhapatnam and Srikakulam are of closed type. The standards used are *Erythrina indica*, *Sesbania grandiflora* and *Moringa pterigosperma*. The life of the gardens is usually three years when they are rotated with rice, sugarcane and ragi.

The important diseases of betel vine inflicting considerable losses annually are foot rot and wilt by *Phytophthora parasitica* var. *nicotianae*, sclerotial wilt by *Sclerotium*

rolfsii, anthracnose by *Colletotrichum* sp. and *Gloeosporium* sp., root rot by *Rhizoctonia* sp. and powdery mildew by *Oidium piperis*. Besides these, several fungal diseases of minor importance were also reported from other parts of India (Maiti and Sen, 1979). Though investigations on pathological aspects were in progress in the State during the last two decades, there was no concerted effort to tackle any of these diseases in detail except for occasional fungicidal trials for the control of sclerotial wilt. The incidence of foot rot and wilt caused by *Phytophthora* sp. is usually restricted to Srikakulam and Vishakhapatnam districts while sclerotial wilt was known as the major pathogen in Guntur and Krishna districts. The other diseases occur to a lesser extent in all the tracts.

However during the end of 1979 a severe incidence of wilt disease occurred in Ponnur area of Guntur district and was reported in the Press as a "mystery disease". A rapid survey of the affected areas and preliminary investigations on the symptomatology and etiology were taken up. The observations made so far in this regard are presented hereunder.

Symptomatology

The disease was first observed in November 1979 mostly in two-year-old gardens. The first perceptible symptom consists of disappearance of lustre of the leaves followed by yellowing and signs of wilting. The leaves droop down, become pale, wither and dry up. Rotting and blackening of stem at the collar region and roots was noted in the wilted plants. Once the disease made its appearance in old gardens, subsequent spread occurred even in younger gardens of six to nine months age. However, the intensity was more severe in older gardens. No leaf spot or rot symptoms were observed in any of the diseased gardens.

Distribution and extent of damage

A rapid survey of the affected gardens in Ponnur area indicated that the disease was widespread affecting over 160 ha out of about 450 ha in the area. Over 25 ha of gardens were completely cut off and abandoned due to 100 per cent incidence of the disease. During the survey, it was learnt that stray incidence of the disease symptoms had been noted since 1977. In view of the low intensity, the disease was ignored. The disease which made its appearance during November, 1979 became severe and widespread by January-February, 1980 and there was a gradual decline by April-May, 1980. The disease incidence was high in heavy soils and very low to absent in lighter soils.

Causal organism

Considerable difficulty was experienced in isolating the organisms involved due to frequent contaminations encountered. This difficulty was overcome by using selective medium (Difco cornmeal agar (CMA) 17g/lit; Pimaricin 10 ppm, Vancomycin HCL-200 ppm; PCNB-100 ppm and pH 6.0) particularly for the isolation of *Phytophthora*. The diseased plant parts yielded *Phytophthora* sp. in most cases while in a few, *Fusarium* sp., *Rhizoctonia* sp. and *Sclerotium rolfsii* were also obtained. This is no surprise as this area was previously known to be endemic for *S. rolfsii*. Based on symptomatology, nature of spread and time of occurrence viz., in cooler months when the temperatures were low (20°C-22°C), which is known to be very congenial for the species of *Phyto-*

phthora, it was generally concluded that *Phytophthora* sp. was responsible for the outbreak of foot rot and wilt in this area. Oospore production was not observed either in the culture or in the infected debris. Detailed studies are underway for establishing the etiology of this disease in Guntur district.

DISCUSSION

Foot rot and wilt due to *Phytophthora* sp. is a well known and a very serious disease of betel vine all over India (Saksena, 1977). In Andhra Pradesh this is fairly a common disease mostly restricted to Srikakulam and Visakhapatnam districts where the gardens are of closed type that provide the optimum conditions for the development, spread and perpetuation of the pathogen. The heavy incidence of this disease in these districts forced the growers to migrate to new areas and set up betel vine gardens as it is their only traditional occupation. However, the incidence of foot rot and wilt in Ponnur area has not been recorded earlier and only sclerotial wilt was the predominant disease in this area so far. This is a soil-borne one and comes in a virulent form and if not controlled in time, it is capable of wiping out extensive areas. This is what exactly happened during 1979-80 outbreak of this disease in Ponnur area, where it is wiped out completely 25 ha besides causing considerable damage to another 160 ha. The disease is most severe during or just after rainy season when temperature, relative humidity and soil moisture level are favourable (Dastur, 1927; McRae, 1930). Similarly, in the present case, the disease became severe after the rains from November to February when night temperatures were around 20°C and thereafter declined with increase in high temperatures. As mentioned earlier, the disease was noted in stray cases in 1977 after the devastating cyclone that hit the coastal districts of Andhra Pradesh but this was ignored. The first serious notice was taken only at the outbreak of the disease in 1979-80. During the two years after the cyclone of 1977 and in the absence of any remedial measures, the pathogen should have built up particularly in the ill-drained heavy soils of Ponnur area and led to the epiphytotic of 1979-80 causing enormous damage. The timely recognition and adoption of remedial measures earlier, could have probably prevented this calamity.

While discussing about the sudden outbreak of *Phytophthora* wilt of betel vine in Ponnur area which is separated by about 320 km from the foot rot and wilt endemic area of Srikakulam district, one will be puzzled as to how and from where the inoculum has come. *Phytophthora* is not adapted for long distance dispersal by air. As such this possibility can be ruled out. Then the source of inoculum must be local or from the neighbourhood. Seedling blight of castor due to *Phytophthora parasitica*, black shank of tobacco due to *Phytophthora parasitica* var. *nicotianae*, bud rot of palmyrah due to *P. palmivora* are of common occurrence in the locality. Further, incidence of bud rot of palmyrah has become an epiphytotic after the devastating cyclone of 1977. The *Phytophthora* species reported to attack betelvine include: *P. nicotianae* var. *parasitica*, *P. parasitica*, *P. parasitica* var. *nicotianae*; *P. parasitica* var. *piperina* and *P. palmivora* (Maiti and Sen, 1979). Thus, there appears to be considerable amount of confusion regarding the species nomenclature of this pathogen. In view of this, detailed morphological, physiological, cross pathogenicity and host range studies of the palmyrah,

castor, betel vine, tobacco and other *Phytophthora* isolates prevalent in the locality are necessary for the specific identification of the *Phytophthora* responsible for the epidemic of foot rot and wilt of betel vine in Ponnur area of Guntur district.

The sudden outbreak of foot rot and wilt by *Phytophthora* spp. in Ponnur area for the first time in 1979-80 undoubtedly surprised the growers, administrators and scientists. It is necessary to initiate an extensive study of the pathogen involved. The association of *S. rolfsii* and other fungi in the wilt complex has to be determined. A study of climatic conditions related to the disease intensity has to be worked to forewarn the growers. Considerable studies have to be taken up on a plant protection schedule that the growers can afford. No information is available at present on any varietal resistance in the State. Screening of existing varieties will have to be taken up. Soil amendments to keep the disease under a threshold level may be the most efficient way and has to be worked out.

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DISCUSSIONS

D. N. Srivastava: Is this outbreak of betel vine wilt in the vicinity of betel vine plantation with a past wilt history?

ANSWER : Not at all. It is far away from such areas and the source of inoculum for this new outbreak is not clear. *Phytophthra* has been noticed on castor in this area. However cross inoculation studies were not carried out.

P. W. F. De Waard : Are the soils heavy where the betel vine is grown?

ANSWER : We have clayey type as well as sandy loam where the betel vine is grown.

P. W. F. De Waard : Heavy soils should be avoided because of the high moisture retention in such soils, which may increase the disease incidence.

NUTRITIONAL AND PATHOLOGICAL STUDIES ON FRUIT ROT OF
BRINJAL CAUSED BY *PHYTOPHTHORA NICOTIANAE* B. de
HAAN VAR. *NICOTIANAE*

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ABSTRACT

Phytophthora nicotianae var. *nicotianae*, causing fruit rot of brinjal grew best on natural media. Temperature 30°C and pH 6.1 were found to be optimum for its growth and sporulation. Amongst the carbon compounds, hexose sugar supported significantly good growth; organic nitrogen sources were found to be superior to inorganic nitrogen sources. This fungus was found to be deficient in thiamine.

The disease developed both in injured as well as uninjured fruits of brinjal at all the stages of their development (young to mature) though faster in younger fruits. Disease development was best at 30°C under high humid conditions.

INTRODUCTION

Brinjal (*Solanum melongena* L.) is an important vegetable crop throughout the world. It was observed to be severely damaged by a fruit rot disease during 1974 in southern parts of Rajasthan. The disease occurs every year causing moderate to heavy losses especially during rainy season. It was found to be caused by *Phytophthora nicotianae* var. *nicotianae* (Jain, 1976). Present paper deals with nutritional requirement of the pathogen and the factors affecting disease development.

MATERIALS AND METHODS

Single spore from pathogenic culture of *Phytophthora nicotianae* var. *nicotianae* was used for all the studies. The fungal growth on solid media, both natural and synthetic, was determined by measuring the radial growth of the colony while the mycelial dry weight was recorded for liquid media. The effect of temperature on the mycelial growth was studied by growing the fungus on the standard PDA subjected to various temperatures (10-37°C).

Modified Mehrotra's liquid medium (Sehgal, 1963) was used as a basal medium for all the physiological studies i.e. effect of carbon, nitrogen and vitamin sources on the growth and sporulation of the fungus. H-ion concentration (3 to 9) was adjusted by adding N/10 HCl or N/10 NaOH to the basal medium. For the carbon and nitrogen require-

ments the respective compounds in basal medium were substituted with the desired compound in required quantity to give equivalent amount of carbon or nitrogen present in the medium. Each flask, containing 20 ml of the medium, was inoculated by a single inoculum disc and incubated at 30°C for 15 days. For vitamin studies the basal medium was made vitamin free by treating it with activated charcoal before adding the required amount of vitamins to the medium.

Various factors affecting the disease development were determined by inoculating fresh brinjal fruits with actively growing 7-day-old fungal culture and incubating them at 30°C in desiccators maintaining the relative humidity with sulphuric acid and distilled water (Buxton and Mellanby, 1934). The fruits were also subjected to various temperatures for determining the optimum temperature for disease development. Intensity of the disease was recorded by measuring the diameter of the lesion after 48 and/or 72 hours of incubation.

RESULTS AND DISCUSSION

Physiological studies :

Media : The best growth of the fungus was recorded on natural media e. g. chickpea, maizemal, oat meal, pea seed, french bean and soyabean meal agar (Table 2.4). Rye seed agar and rape seed agar were found to be poor natural media. Among synthetic media glucose asparagine agar, Mehrotra's agar, Brown's agar and glucose nitrate agar supported moderate growth whereas Asthana and Hawker's agar medium was poor. Sporulation was good on natural media, like pea seed and hemp seed agar media. The growth on natural media was fluffy, cottony and aerial where as it was poor and submerged type or sparse on synthetic media. Tucker (1931), Hendrix and Apple (1964) and Tiwari (1973) also found natural media to be better for growth of *Phytophthora* spp., *P. parasitica* var. *nicotianae* and *P. parasitica* var. *piperina* respectively. Mahendra Pal (1974) observed good growth of *P. drechsleri* var. *cajani* on Mehrotra's medium. Mehrotra's modified medium, in which ammonium nitrate was replaced by asparagine, was found to be good for *P. nicotianae* var. *nicotianae* (brinjal isolate). This medium was also reported to be good for *P. parasitica* var. *sesami* by Sehgal (1963).

Temperature : The growth of brinjal *Phytophthora* increased with the increase of temperature up to 30°C and then showed a decline. Maximum growth was recorded at 30°C which was closely followed by growth at 28°C. No growth was recorded at 10°C and 37°C and was very poor at 36 and 15°C (Table 2.5). Roncadori (1965) found that six species of *Phytophthora* grew best at 20°C while 19 species and one variety had optimum temperature between 25 to 30°C. *P. parasitica* var. *nicotianae* fell in second group. Waterhouse (1963) and Westesteijn (1973) also recorded an optimum temperature range of 25 to 30°C for the growth of *P. nicotianae* var. *nicotianae*.

H-ion concentration : Lower pH may affect the degree of the dissociation of inorganic ions of the substrate, entry of essential vitamins and enzymatic reactions while higher pH may initially arrest its growth. Brinjal *Phytophthora* grew in a wide range of pH

(3.1 to 6.9) but it preferred acidic medium; optimum was pH 6.1 (Table 2.6). Lilly and Barnett (1951) observed that most of the fungi grew satisfactorily between pH 5 to 7, but the optimum generally lay on acidic side. Cameron and Milbrath (1965) found that most of the 8 species of *Phytophthora* studied apparently grew best on a buffered medium at pH 4.5 to 5.5.

Table 2.4. Growth and sporulation of *Phytophthora nicotianae* var. *nicotianae* on different agar media after 96 hours of incubation at 30°C

Medium	Growth (mm) ¹	Sporulation ²
Maize meal	87	+
Oat meal	85	++
Chick pea	89	++
Pea seed	81	++++
Brinjal fruit extract	61	++
French bean	76	++
Carrot extract	58	++
Soybean meal	67	++
Potato dextrose	56	+
Hemp seed	50	+++
Rye seed	21	++
Rape seed	19	++
Glucose asparagine	44	—
Mehrotra's	40	+++
Brown's	46	—
Glucose nitrate	34	+++
Richard's	30	—
Sabouraud's	41	—
Czapek Dox	42	—
Asthana & Hawker	23	—

1 : Average of 4 replicates; 2. Presence of zoosporangia and chlamydospore;

— = denotes nil + = very poor, ++ = Poor, +++ = moderate,
 + + + + = good, + + + + + = abundant S.E. m = 5.77 C.D. 5% = 16.32.

Table 2.5. Growth of *Phytophthora nicotianae* var. *nicotianae* on PDA at different temperature after 96 hours of incubation

Temperature(C)	Growth (mm) ¹	Temperature(C)	Growth (mm) ¹
10	0	30	56
15	6	35	38
20	26	36	5
25	35	37	0
28	52		

1 = Average of 3 replications

S. Em. = 0.443

C.D. 5% = 1.315

Table 2.6. Growth of *Phytophthora nicotianae* var. *nicotianae* on different H-ion concentration after 15 days incubation at 30°C

Initial pH	pH after autoclaving	Average dry wt. (mg) ¹
3.0	3.10	57.0
4.0	4.20	62.5
5.0	5.00	67.0
5.5	5.50	70.8
6.0	6.00	77.8
6.5	6.15	82.3
7.5	6.50	64.3
8.0	6.70	51.6
9.0	6.90	39.5

¹ - Average of 4 replication

S. E.m. - 1.37

C.D. 5% - 3.97.

Carbon and nitrogen nutrition : Out of 12 carbon sources tried, brinjal *Phytophthora* grew best on hexoses (glucose and fructose) followed by sucrose and maltose (Table 2.7).

Table 2.7. Growth of *Phytophthora nicotianae* var. *nicotianae* on different carbon sources, incubated for 15 days at 30°C

Carbon source	Av. dry wt. (mg) ¹	Carbon source	Av. dry wt. (mg) ¹
D-Glucose	78.5	Lactose	40.0
D-Fructose	75.0	Starch	39.5
Sucrose	67.5	Glycerol	32.5
Maltose	50.5	Cellulose	26.0
L-Sorbose	47.3	Mannitol	24.5
Dextrin	44.5	Control	6.0
L-Xylose	40.5		

¹ - Average of 4 replications.

S. Em. - 3.09

C.D.5% - 8.83.

The polysaccharides (starch and cellulose) and hexahydric alcohols (glycerol and mannitol) were found inferior sources of carbon. It is well known that carbon and nitrogen are the main essential elements required by fungi for functional and structural processes, but the structural differences amongst compounds providing the sources and fungus metabolism make their availability specific to certain fungi. Brannon (1923) observed that nearly all culturable fungi were able to use glucose and fructose and the growth was usually good with either of the sugars. Glucose and fructose were also found to be best carbon sources for several species of *Phytophthora*, (Lopatecki and Newton, 1956; Kassura and Nishioka, 1958; and Roncadori, 1965). Wills (1954) observed glycerol and other alcohol either poor or inhibitory to *P. parasitica* var. *nicotianae* as has been observed in the present study also. Among the inorganic nitrogenic salts, Ammonium nitrate supported the best growth of the brinjal fruit rot pathogen followed by ammonium sulphate and ammonium chloride. Potassium and sodium nitrate supported poor growth (Table 2.8). Several species of

Table 2.8. Growth of *Phytophthora nicotianae* var. *nicotianae* on different inorganic nitrogen salts, incubated for 15 days at 30°C

Nitrogen source	Average dry weight (mg) ¹
Ammonium nitrate	45.8
Ammonium sulphate	44.0
Ammonium chloride	41.0
Sodium nitrate	33.8
Potassium nitrite	27.0
Sodium nitrite	5.0
Control (no nitrogen)	9.5

1 - Average of 4 replications

C.D. 5% - 3.15.

SEM = 1.07.

Phytophthora are reported to prefer ammonium nitrate as nitrogen source (Roncadori, 1965; Cameron and Milbrath, 1965 and Mahendra Pal, 1974).

The fungus grew more luxuriantly on organic nitrogen compared to inorganic nitrogen sources with the only exception of cysteine hydrochloride. Chee and Newhook (1965) found glutamine as the best nitrogen source for *P. cinnamomi*. Leal *et al.* (1971) compared 21 amino acids as nitrogen source for the growth of *P. cactorum* and *P. heveae* and found that L-alanine, L-arginine, L-asparagine, glycine, L-aspartic acid and L-glutamine to be good nitrogen sources for both the fungi. During present investigations also glutamine was found to be the best source followed by L-alanine and L-asparagine for the growth of brinjal *Phytophthora* (Table 2.9).

Table 2.9. Growth of *Phytophthora nicotianae* var. *nicotianae* on different amino acids, incubated for 15 days at 30°C

Amino acids	Average dry weight (mg) ¹	Amino acids	Average dry weight (mg) ¹
Glutamine	90.8	Glycine	70.3
DL-Alanine	86.3	L-Phenylalanine	63.5
L-Asparagine	84.0	Methionine	50.0
DL-Valine	82.3	Tryptophan	48.5
Leucine	81.8	Cystine	48.0
Arginine	79.5	Cysteine HCl	34.0
DL-Aspartic acid	75.0	Control	10.3
Serine	71.3		

1 - Average of 4 replications.

S.Em. = 1.28.

C.D. 5% - 3.66.

Vitamin : Several workers found thiamine to be essential for *Phytophthora* species (Payette and Perrault, 1944, Sakai, 1957, Chee and Newhook, 1965 and Roncadori, 1965). Addition of various vitamins in the medium individually increased the growth of brinjal *Phytophthora*. Thiamine followed by biotin supported significantly good growth (Table 2.10). This fungus was observed to utilize thiamine at a concentration range between 5-50 µg per litre, optimum being at 20 µg per litre; the growth has significantly reduced in the absence of thiamine in the medium (Table 2.11). This indicates that although brinjal *Phytophthora* is partially deficient for thiamine it is not indis-

Table 2.10. Growth of *Phytophthora nicotianae* var. *nicotianae* on different vitamins, incubated for 15 days at 30°C

Vitamin	Concentration per litre (μ g)	Average dry weight (mg) ¹
Thiamine	100	73.7
Biotin	5	61.0
Pyridoxin	100	47.3
Riboflavin	5	45.3
Inositol	100	40.3
Control(no vitamin)	—	38.3

1 - Average of 3 replications.

S.Em. = 3.61.

CD 5% = 11.12.

pensible and can be substituted by biotin to a great extent. Reduction in growth, due to the absence of thiamine, was also noted in case of *P. colocasiae*, *P. parasitica* and *P. cinnamomi* by Washir (1969) and Ridings *et al.* (1969).

Table 2.11. Growth of *Phytophthora nicotianae* var. *nicotianae* on different dosages of thiamine, incubated for 15 days at 30°C

Thiamine concentration (μ g/litre)	Average dry weight (mg) ¹	Thiamine concentration (μ g/litre)	Average dry weight (mg) ¹
0.0	38.5	50.0	76.0
1.0	71.0	100.0	73.7
5.0	78.0	200.0	70.3
10.0	80.3	500.0	66.0
20.0	84.7		

1 - Average for 3 replications.

S.Em. = 1.31.

C.D. 5% = 3.94.

Pathological studies :

Inoculation methods : Infection developed within 48 hours in all the methods of inoculation (Table 2.12). However, the disease development was more pronounced in injured fruits, most of which rotted completely within 72 hours. Pin prick method was found to be the best. Infection started as water-soaked lesion followed by discoloration of fruit skin leading subsequently to the rotting of pulp. Initially infection was slightly delayed on uninjured surface, but once the infection got established the disease developed

Table 2.12. Relative efficacy of different methods of inoculation on the development of brinjal fruit rot by *Phytophthora nicotianae* var. *nicotianae* at 30°C

Treatment	Fruits inoculated	Fruits infected		Average lesion dia. (cm)	
		48 hrs	72 hrs	48 hrs	72 hrs
Without injury	4	1	4	0.3	4.3
Pin-prick method	4	4	4	5.9	R
Well method	4	4	4	5.2	R
Cut method	4	4	4	5.7	R
Injection method	4	4	4	2.2	6.8
Control (uninoculated, uninjured)	4	—	—	—	—
Control (uninoculated, injured)	4	—	—	—	—

(—) = denotes no disease; R = Complete rotting

at the same pace as in injured fruits. Thus the minor injury of fruits in nature during favourable conditions may result in quick rotting in the field or in storage.

Age of fruits : Fruits of various age group (1 to 13 days) readily developed infection when injured, but was delayed in older (9 to 13 days) uninjured fruits (Table 2.13). The fact that with length of time the fruits of all ages (injured or uninjured) developed infection shows that probably age of the fruit has no effect on disease development. Injury apparently helps in early establishment of the disease. The capability of *Phytophthora* to produce rotting in younger as well as older fruits makes it a potential threat to this vegetable crop.

Table 2.13. Effect of age of fruits on the development of fruit rot of brinjal by *Phytophthora nicotianae* var. *nicotianae* at 30°C

Age (days)	Average lesion dia (cm.)			
	48 hrs.		72 hrs.	
	I	W	I	W
1	R	R	R	R
3	3.8	2.8	R	R
5	4.2	2.0	7.7	5.9
7	4.2	1.8	7.2	6.4
9	4.6	—	6.4	3.8
11	4.4	—	5.8	4.2
13	4.2	—	6.0	3.0

(—) - denotes no disease; I - Inoculation on injured fruits
W - Inoculation on uninjured fruits
R - Complete rotting of fruits.

Temperature and Humidity : The most congenial temperature for disease development was found to be 30°C (Table 2.14) at which infection was initiated within 48 hours of inoculation whereas at other temperatures it took 72 hours in uninjured fruits. No disease developed at 10 and 40°C. These results correspond well with that of the optimum temperature requirement for the growth of this fungus in culture (Jain, 1976).

Table 2.14. Effect of different temperatures on the development of fruit rot of brinjal caused by *Phytophthora nicotianae* var. *nicotianae*

Temperature°C	Average lesion diameter (cm.)					
	48 hrs.		72 hrs.		96 hrs.	
	I	W	I	W	I	W
10	—	—	—	—	—	—
Room (17-21)	0.90	—	3.15	1.05	5.20	3.97
22.5	2.82	—	5.02	2.10	7.12	5.75
30	5.30	4.10	7.95	6.50	R	R
35	0.50	—	2.35	0.75	4.80	1.95
40	—	—	—	—	—	—

(—) - denotes no disease; I - injured fruits W - uninjured fruits R - rotted fruits.

Fruit rot of brinjal is directly related to high humid conditions. The disease did not develop at relative humidity below 80 per cent in both injured and uninjured fruits. However, for fast disease development high humidity was found essential (Table 2.15). These results are in agreement with the observations of Cochrane (1958) that the sporangia of *Phytophthora infestans* and other species of *Phytophthora* were extremely susceptible to desiccation. He also reported that when the RH dropped much below 100 per cent the sporangia were killed in a few hours. Trujillo (1965) recorded a total inhibition of sporulation in *Phytophthora colocasiae* at RH lower than 90 per cent.

Table 2.15. Effect of different relative humidities on the development of brinjal fruit rot caused by *Phytophthora nicotianae* var. *nicotianae* at 30°C

Relative humidity (%)	Average lesion diameter (cm.)			
	48 hrs.		72 hrs.	
	I	W	I	W
0	—	—	—	—
10	—	—	—	—
40	—	—	—	—
60	—	—	—	—
80	4.25	—	7.40	3.40
100	5.50	4.00	7.85	6.75

(—) = denotes no disease; I - injured fruits W - uninjured fruits.

Since the optimum temperature and humidity conditions prevail during rainy season in most part of the country, *Phytophthora* fruit rot may pose a threat to rainy season brinjal crop especially during the wet years.

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SURVIVAL OF INOCULUM OF THE LEAF BLIGHT FUNGUS
PHYTOPHTHORA COLOCASIAE INFECTING TARO, COLOCASIA
ESCULENTA IN THE SOLOMON ISLANDS*

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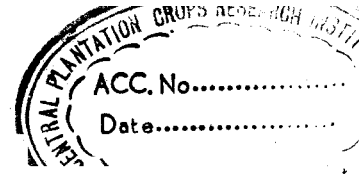
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ABSTRACT

Phytophthora colocasiae was successfully isolated by baiting with detergent-treated taro leaf discs 8 cm diameter placed on water slurries of soil, on suspensions of macerated infected leaf lesions or on the washings from petioles of harvested plants. Taro root tips, detached or left on corms, were not susceptible to zoospores of *P. colocasiae* nor were detached root tips of *Lupinus angustifolius*. Cubes of taro corm used as baits, and agar selective for *Phycomycetes* which was inoculated directly with soil, both became too heavily overrun by *Pythium splendens* to allow detection of *P. colocasiae*. Investigations indicated that inoculum on lesions of detached leaves and in soil remains viable for only a few days. Petiole bases which comprise the bulk of the 'tops' used for vegetative propagation, lost detectable natural inoculum rapidly (2 days) if stored dry, but less rapidly (14 days) if planted immediately in the field. Artificially augmenting surface inoculum with naturally produced sporangia considerably extended the periods of detectability, probably by increasing the chances that a few propagules would survive, especially during dry storage. Incubation of inoculated tops in high humidity led to active infection and sporulation on petioles, especially on cut ends, a situation that might be paralleled under suitable moisture conditions in the field. Of several aroid species tested by artificial inoculation only *Alocasia macrorrhiza* was susceptible. Natural infection of this plant has not been seen, making it an unlikely alternate host of *P. colocasiae* under field conditions. Thus perennation between taro crops is effected by short-lived surface propagules and possibly also by mycelium within lesions on petioles. Reduction of the former and prevention of the latter might be achieved by dry storage of tops for 2 to 3 weeks.

*For full text of the paper, vide *Ann. appl. Biol.* 94: 379-390, 1980.



SOURCES OF PHYTOPHTHORA PALMIVORA INOCULUM IN SOLOMON ISLAND COCOA PLANTATIONS*

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ABSTRACT

Data from a trial in which regular roguing prevented sporulation of *Phytophthora palmivora* on tree-borne cocoa pods indicated that stem cankers associated with flower cushions act as a continuing source of infection.

Other sources of *P. palmivora* propagules were identified as infected flowers and immature foliage such as volunteer seedlings, chupons, and leaves of flush shoots in the canopy. Mature leaves are rarely susceptible. Initial infection of a maiden plantation can result from inoculum brought in by rats and on harvesting and pruning tools.

Baiting with cocoa pod plugs indicated that propagules of *P. palmivora* could be dislodged by water from bark of trunks and especially jorquettes and branches. Water-transferable propagules were frequently detected on surfaces of healthy canopy leaves. Interception of falling leaflets of the shade tree *Leucaena leucocephala* increased the inoculum-holding frequency of cocoa canopy leaves. Freshly fallen *L. leucocephala* leaflets were susceptible to inoculation with the pathogen, which produced up to 200 sporangia per leaflet. Other shade tree species were shown to provide a similar hazard. Applying fungicidal sprays to the complete canopy as well as to pods might improve disease control.

DISCUSSIONS

Y. R. Sarma : Does the *Leucaena leucocephala* increase the disease in cocoa plantations in Solomon Islands?

ANSWER : It increases the inoculum in the area because this plant also is susceptible to the pathogen.

*For full text of the paper, vide *Trans. Br. mycol. Soc.* 71(2): 239-249, 1978.

PHYTOPHTHORA PALMIVORA IN COCOA PLANTATION SOILS IN THE SOLOMON ISLANDS*

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ABSTRACT

The most efficient of several soil-baiting methods for the detection of *Phytophthora palmivora* in soil proved to be the incubation of plugs of cocoa-pod tissue half-submerged in flooded soil in open Petri dishes. Negative dishes often gave positive results on a second baiting. In open-topped vials or in covered Petri dishes *Pythium vexans* frequently overran and obscured the presence of *P. palmivora*.

Soil populations of *P. palmivora* scarcely diminished with increasing distance from the trunks of the cocoa trees and direct microscopic observation of propagules inoculated into soil in the field or in Petri dishes, suggested that propagules remain dormant in the soil under the influence of fungistasis. No *P. palmivora* was detected in soil from a disease free young plantation about to mature its first crop but in a plantation with a long history of black pod disease, *P. palmivora* was detected in over half the soil samples tested 2 months after felling. Populations persisted at low levels for 34 months.

Fallen cocoa leaves were shown to be a potential source of splash-dispersed inoculum as were the fallen leaflets of the shade tree *Leucaena leucocephala*. *Phytophthora planivora* was also isolated readily from 'tents' of the ant *Technomyrmex detorquens* on cocoa pods.

*For full text of the paper, vide *Trans. Br. mycol. Soc.* 69(1): 31-38, 1977.

Saturday, 20 September, 1980
3.00 p.m. — 5.30 p.m.

SESSION 3

**TAXONOMY, PHYSIOLOGIC SPECIALISATION AND
RESISTANCE**

Chairman : Dr. H. S. SOHI

Rapporteur : Dr. P. CHIDAMBARAM

TAXONOMY OF PHYTOPHTHORA

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The genus, type species *Phytophthora infestans*, was established by de Bary (1876) in the Peronosporaceae, order Peronosporales, and later transferred to the Pythiaceae by Fitzpatrick because of the indefinite sporangiophore and development of sporangia in succession.

Hyphae are non-septate, except in older cultures, branching at right angles, the branches constricted at the point of origin. Various hyphal swellings are characteristic of some species. Chlamydospores are formed by some isolates. Sporangiohores, except in *P. infestans*, differ little from other hyphae, branching being sympodial, sub-sporangial or by internal proliferation, through the empty sporangium. Sporangia vary in size and shape. An apical thickening, the depth characteristic of species, protrudes in some to form a well defined papilla. Sporangia may be caducous, i.e. detached readily from the sporangiophore, with a pedicel of length depending on the species or non-caducous; the latter group includes all non-papillate, proliferating species germination may be direct, by a germ tube, or indirect, by differentiation of biflagellate zoospores within the sporangium.

Oogonia are spherical or tapering to the stalk, the wall usually smooth, occasionally verrucose. The antheridium may be paragynous or amphigynous. A single oospore usually develops, having a variously thickened inner wall. While some 'homothallic' species form sex organs readily in single cultures, in others ('heterothallic') the pairing of two compatible isolates, A¹ and A², belonging to the same or different species, may be necessary.

Combinations of morphological criteria, primarily relating to the sporangial apex, occurrence of oospores and antheridial relationships, were used by Waterhouse (1963) to classify *Phytophthora* spp. into six groups and to construct a key. The species now recognised can still conveniently be considered in these groups.

Group I. Apex of sporangium markedly papillate, exit pore narrow, sporangium not proliferating internally. Oogonia in single-strain culture, antheridia usually all paragynous. *cactorum*; *iranica*.

Group II. Sporangium as I. Oogonia formed by some isolates in single cultures, by others on pairing, antheridia amphigynous.

- palmivora* MF 1. Sporangium caducous with short, broad pedicel (<5 μ)
MF 2. As MF 1 but sporangium broader
MF 3. (*P. megakarya*) Pedicel thin, c. 12 μ . Oogonium tapering to base.
MF 4. Pedicel 20-250 μ . Sporangium elongated with tapered base.

- meadii* Sporangium often distorted, pedicel 10-20 μ . Oogonia sometimes in single cultures.
- botryosa* Sporangia very abundant, clumped, elongated ellipsoidovoid, tapering to base, pedicel slender, <10 μ .
- heveae*. Oogonia abundant in single cultures, usually tapering to stalk. Sporangia often irregular, pedicel 10 μ .
- castaneae* Similar to *heveae* but oogonia with rounded protuberances.
- arecae* Sporangia broad-nearly spherical, pedicel 1-6 μ .
- boehmeriae* Sporangia regular, ellipsoid-nearly spherical, pedicel 3 μ Oogonia formed readily in culture.
- capsici* Sporangia variable, often >1 apex, pedicel >10 μ . Max. temp. > 35°C.
- nicotianae* Oogonia on pairing or sometimes in single cultures. Max. temp. > 35°C, vars. *parasitica* and *nicotianae*.
- citrophthora* Cultures with fine, radiate growth. Sporangia variable. Oogonia not usually formed in single or paired cultures. Max. temp. 32°C.
- mexicana* Sporangia ovoid-ellipsoid, tapering to base. Oogonia usually in single cultures but not abundant.

Group III. Apex of sporangium less protuberant, thickening less than hemispherical, exit pore narrow. Sporangia not proliferating internally. Oospores in host or in culture, antheridia predominantly paragynous: *citricola*; *syringae*; *porri*; *primulae*; *vesicula*.

Group IV. Apex of sporangium as in III. Sporangium usually caducous, not proliferating internally. Oogonia not always in single cultures. Antheridia predominantly or all amphigynous: *infestans*; *colocasiae*; *hibernalis*; *phaseoli*; *melonis*.

Group V. Apex not protruding beyond general contour of sporangium (non-papillate), exit pore broad. Sporangia not caducous, generally produced only in water, proliferating internally. Oospores always produced in host and/or in culture; antheridia paragynous and amphigynous: *fragariae*; *megasperma* vars. *megasperma* and *sojae*; *quininea*; *verrucosa*.

Group VI. Sporangium as in V. Oospores not always produced in single culture. Antheridia all or mostly amphigynous: *cambivora*; *cinnamomi*; *cryptogea*; *drechsleri* vars. *drechsleri* and *cajani*; *erythroseptica*; *vignae*; *lateralis*; *japonica*.

While morphological criteria are accepted as a useful basis for classification in *Phytophthora*, lack of certain characters, as when isolates fail to form the necessary organs, may lead to difficulties in identification. The extent of variation within any one species, changes in individual isolates during culture and the possibility of hybridization between species are also problems to be considered. Other methods of investigating relationships between taxa have received increasing attention. Ultrastructural studies, particularly of the sporangial apex, confirmed the distinction between papillate and non-papillate spe-

cies. Cytology has proved particularly promising in revealing chromosome differences in the sex organs at meiosis; thus large or small chromosomes tetraploids and diploids were observed, distinguishing a number of species, and, in *P. megasperma*, separating varieties. Nutritional requirements can be employed to separate groups of species. Electrophoresis is a useful and reliable tool for distinguishing species by their protein and enzyme patterns. Some relationships between species or species groups have also been demonstrated serologically.

Most of this more recent work provides additional support for the maintenance of already established species. The further development of such studies is desirable in order that morphological data, especially when inadequate, can be supplemented, and to investigate possible basic factors related to morphological differences.

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DISCUSSIONS

P. S. Sohi : In case of powdery mildews the L/B ratio is used for identification. Is it the case in *Phytophthora* too?

ANSWER: Yes. It is also used as one of the criteria.

C. R. Ramesh : Is there parthenogenetic development of oospores?

ANSWER: No.

K. V. Chandrasekhara : Are there any species of *Phytophthora* (probably those with papillate sporangia) in which the sporangia possess an operculum?

ANSWER: Some isolates obtained recently from Mangrove Swamps seem to show a structure of this type.

K. V. Chandrasekhara : Two types of sporangiophores are described in *Phytophthora* spp. viz. (1) sympodial type and (2) umbellate type. Is this categorization based on ontogenetic study? I ask this because very often a basically sympodial development of

sporangia may (in the mature state) result in an umbellate type of structure due to production of sporangia in rapid successions. In other words in a truly umbellate type of sporangiophore, the sporangia should develop simultaneously.

ANSWER: Probably basically sympodial.

P. H. Tsao: Do you believe there are considerable similarities between *P. arecae* and the so called *P. palmivora* MF2?

ANSWER: I would not confuse them. In my experience the sporangia of *P. arecae* tend to be much nearer spherical than those of *P. palmivora* MF2.

MORPHOLOGY AND IDENTITY OF BLACK PEPPER *PHYTOPHTHORA* ISOLATES

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Controversy and confusion have existed for decades regarding the identity and nomenclature of *Phytophthora* isolates pathogenic on black pepper (*Piper nigrum*) and on other *Piper* species. Isolates from different geographic locations have been identified by different workers as *Phytophthora colocasiae*, *P. palmivora*, or various varieties of *P. palmivora* or *P. parasitica* (see references in Nambiar and Sarma, 1977, and in Tsao and Tummakate, 1977). In addition, an isolate from black pepper in Thailand with unique morphological features different from all the above named species was recently reported, but unnamed, by Tsao and Tummakate (1977). There might indeed be different species or varieties of *Phytophthora* infecting black pepper around the world. However, it is also possible that some or all of these above nomenclatures have resulted from incomplete studies, improper identification procedures, incorrect observations, and/or inadequate existing keys and species descriptions, all of which could have led to misidentifications and the resulting controversy. Turner (1969), after a comparative study, concluded that all *Phytophthora* isolates from *Piper* spp. in southeast Asia were similar and proposed that they be referred to, henceforth, as a typical strain of *P. palmivora*. The conclusion was based primarily on the taxonomic works of Waterhouse and Holliday (personal communications with P. Holliday, G. J. Turner and G. M. Waterhouse). The atypical strain, or *Piper* form, differs from the typical strain (morphological form 1) of *P. palmivora* in many respects, one of which is the noncaducous* nature of the sporangia in the atypical black pepper strain (Holliday and Mowat, 1963; Waterhouse, 1974). The Thai isolate studied by Tsao and Tummakate (1977), however, showed caducous nature of the sporangia which have a long pedicel, high length/breadth (L/B) ratio, tapered base, and umbellate ontogeny. It agrees well with the descriptions of the recently described '*P. palmivora*' Morphological Form 4 (MF4) (Al-Hedaithy and Tsao, 1979; Brasier and Griffin, 1979; Griffin, 1977; Kaosiri *et al.*, 1978; Zentmyer *et al.*, 1977).

A comparative study has been in progress in our laboratory in recent years employing a number of black pepper *Phytophthora* isolates obtained from various parts of the world, as well as various '*P. palmivora*' MF4 isolates from other hosts. Results have shown that

*Re-examination, by G. M. Waterhouse and P. H. Tsao, of the original isolates sent by P. Holliday from Malaysia to the Commonwealth Mycological Institute in the late 1950's has shown that sporangia of those isolates are indeed caducous and are with long pedicels, high L/B ratios, tapered base, and occasional double septa. The long pedicel was previously misinterpreted as being a broken hypha on which a persistent (noncaducous) sporangium had formed.

all black pepper isolates so far examined have similar morphological features which fit the MF4 descriptions. This paper summarizes the work up to the present which involves the collaboration of a number of researchers including A. Tummakate, R. Ugale, Pamela W. Tsao, and A. Alizadeh.

All *Phytophthora* isolates obtained from black pepper in South East Asia, Central America, and Africa were similar in many morphological features, and can be called '*P. palmivora*' MF4. They included the isolates from Thailand (13A-3, mating type A2), Malaysia (IMI-182710A, A2; IMI-182710B, A2; IMI-182711, A1; IMI-182714, A1), Guatemala (P412, A1), West Africa (P431, A1), and Brazil (P1199, A1). The sporangia of all these isolates were caducous, had tapered base when formed in the light, were elongated with a high L/B ratio (ranging from 1.6 ± 0.1 to 2.5 ± 0.4), and had a long pedicel (ranging from 84 ± 40 to $202 \pm 115 \mu\text{m}$). All isolates showed certain degree of umbellate ontogeny of the sporangia (Tsao, 1977; Tsao and Tummakate, 1977), and most of them produced a certain percentage of double-septate sporangia (Tsao and Tummakate, 1977). The black pepper isolate (S-1) and *Piper betle* isolate (S-4), both sent recently by Y. R. Sarma from India, also showed all the morphological features of '*P. palmivora*' MF4 (Alizadeh and Tsao, unpublished).

All the above *Piper* isolates examined were similar in sporangium morphology to several other MF4 cultures isolated from cocoa in Brazil (P253, A1; P622, A1; P624, A1), from macadamia in Hawaii (P1072, A2), and from rubber in Brazil (IMI-206790, A1). These isolates had sporangia of L/B ratios ranging from 1.5 ± 0.1 to 1.8 ± 0.2 . The pedicel lengths of the sporangia of these isolates varied from 36 ± 18 to $181 \pm 81 \mu\text{m}$, but fit well within the descriptions of MF4 (Al-Hedaithy and Tsao, 1979; Brasier and Griffin, 1979; Griffin, 1977; Tsao *et al.*, 1981; Zentmyer *et al.*, 1977). Double-septation and umbellate ontogeny have also been observed in certain sporangia of these MF4 isolates from non-*Piper* hosts.

The MF4 isolate from macadamia had been designated by Kunimoto *et al.* (1976) and Aragaki and Uchida (1980) as *Phytophthora capsici* Leonian. Some MF4 isolates from cocoa have also been considered as *P. capsici* by Zentmyer *et al.* (1979). A comparative study was therefore undertaken employing several authentic *P. capsici* isolates including Leonian's type isolate, P1091 (same as IMI-40502 or CBS 128.23), P504 (ATCC 32067), and P505 (ATCC 32068), all of which are pathogenic on *Capsicum* spp. These were compared with the eight black pepper isolates and the five other MF4 isolates from non-*Piper* hosts on standardized media and incubation conditions. Morphological variability within each isolate was also studied under varying conditions and with different methods of sporangium production. Also studied was the ability of all these isolates to grow at 35°C and to produce chlamydospores. Our results (Tsao *et al.*, 1980) so far have indicated that some black pepper isolates differ from the authentic *P. capsici* isolates which produce round-based sporangia when forming in the dark, exhibit irregular sporangium ontogeny, and grow at 35°C. Other black pepper isolates and the cocoa and rubber isolates have only some, but not all, of these *P. capsici* features. The macadamia isolate, like the black pepper isolate from Thailand, differs greatly from *P. capsici* in having sporangia with extremely tapered base and extremely long pedicel, umbellate sporangium ontogeny

and no growth at 35°C. In addition, the macadamia isolate differs from all other isolates in forming chlamydo-spores which are absent in *P. capsici*.

Descriptions of *Phytophthora* sexual reproductive structures (antheridia, oogonia, and oospores) in the literature have stemmed from work involving different methods, media, and incubation conditions, and are often made without regards to distinctions between the hybrid and selfed sexual structures. Sexual organs in many *Phytophthora* species are morphologically variable, not species specific in most cases, and are often useless in species identification. Our results have also shown a great variability in shape and size of these sexual structures among isolates within the black pepper group, within the MF4 group of non-*Piper* origin and within the authentic *P. capsici* group as well as among all these three major groups. Based on the morphology of sexual structures many of the black pepper isolates cannot be easily identified and designated as *P. capsici*.

In conclusion, all black pepper isolates from different geographic regions show similar morphological features identifiable as '*P. palmivora*' MF4. They are also morphologically similar to the MF4 isolates from certain non-*Piper* hosts so far studied. Some, but not all, of the MF4 isolates (including black pepper isolates) possess features of *P. capsici* sensu Frezzi (1950), but cannot be correctly keyed and identified to this species if one follows species descriptions by Leonian (1922), Tucker (1931), Waterhouse (1963) or Newhook *et al.* (1978). It appears that *P. capsici* needs to be accurately redescribed to include in the revised description some features and certain variability hitherto missing in all present species descriptions for *P. capsici*. Correct nomenclature of the black pepper *Phytophthora* isolates, therefore, might not be possible until such redescription of *P. capsici* is made.

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DISCUSSIONS

P. W. F. De Waard: In your studies on identification of species carrot medium was used. The original specimens could have been grown on a different medium like PDA. Will it make any difference?

ANSWER: Our experience does not show much difference.

D. H. Lapwood: What happens in nature? You do not know whether the sporangia are produced in light or dark? Can you use this criterion for identification?

ANSWER: Light is required just for a short interval of time of the production for sporangia in nature, and this is generally available.

F. J. Newhook: Will Dr. Tsao like to comment on the difficulties imposed on taxonomy by description of new species on the basis of a single type culture which can't take account of the range of variability of a species, though I would accept that his whole paper dealing with frustrating variability is adequate.

ANSWER: I agree with the point made. Hence I don't follow conventional procedures of taxonomists. I study many cultures, original descriptions, etc, compare them and finally decide.

D. N. Srivastava : Are you planning to examine the progenies of mating types?

ANSWER: These oospores are carefully chosen. They possess true morphology of the isolates. Many do the matings with unknown isolates. The unknown probably produces chemical stimulus for the known say *P. parasitica* to produce oospores.

GENETICS OF THE GENUS *PHYTOPHTHORA*

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The genetics of the genus *Phytophthora* is still not entirely understood though considerable information has been published on this topic for the last 15 years. These publications mainly concern the nature of the sexuality in *Phytophthora* species, the nature of the life cycle, and the possibility of interspecific hybridization throughout the genus.

According to Waterhouse's key (Waterhouse, 1963), the genus *Phytophthora* includes forty two species. Twenty seven of them are homothallic and form oospores in single culture, whereas twelve are considered as heterothallic since most of their isolates usually produce oospores only when they are paired with a compatible strain. The sexual reproduction of the three last species viz., *P. oryzae*, *P. lateralis*, and *P. gonapodyides* has not been observed.

Homothallic species may be divided in two groups (Savage *et al.*, 1968) (Table 3.1): species with predominantly paragynous antheridia, and species with predominantly amphigynous antheridia.

All heterothallic species form amphigynous antheridia and their isolates are divided into two compatible groups designated as A_1 and A_2 . Both A_1 and A_2 compatibility or mating types, first recognised in *P. infestans* (Gallegly and Galindo, 1958), are known to be present in all twelve heterothallic species. Sexual compatibility is independent of the species status: oospore formation occurs in most $A_1 \times A_2$ pairings.

Each A_1 or A_2 isolate is bisexual and self incompatible. However, relative degrees of maleness and femaleness have been observed among *P. infestans* A_1 and A_2 isolates (Galindo and Gallegly, 1960). Some isolates of each mating type act as strong male and form principally antheridia, when some isolates behave as strong female and produce mainly oogonia. Some isolates are intermediate in their relative sexual strength and are able to form both types of gametangia.

It has been shown that zygote formation in heterothallic species does not always result from the mating of two compatible A_1 and A_2 isolates. Some A_2 isolates can form oospores in single culture by self fertilization, in response to different chemical inductions. In this way, selfing of A_2 isolates of *P. palmivora* and other species may be induced by volatile substances from another fungus: *Trichoderma viride* (Brasier, 1975). In *A_2 P. cinnamomi*, selfed oospores are produced in presence of avocado root exudates

Table 3.1. Homothallism, Heterothallism and mode of formation of antheridia in the Genus *Phytophthora*

Homothallic species		Heterothallic species	Sexual stage
Antheridia predominantly paragynous	Antheridia predominantly amphigynous	Amphigynous antherida	
<i>P. cactorum</i>	<i>P. heveae</i>	<i>P. capsici</i>	<i>P. oryzae</i>
<i>P. macrospora</i>	<i>P. mexicana</i>	<i>P. palmivora</i>	<i>P. lateralis*</i>
<i>P. inflata</i>	<i>P. meadii</i>	<i>P. citrophthora</i>	<i>P. gonapodyides</i>
<i>P. citricola</i>	<i>P. boehmeriae</i>	<i>P. arecae</i>	
<i>P. syringae</i>	<i>P. phaseoli*</i>	<i>P. nicotinae</i> var. <i>nicotiana</i>	
<i>P. primulae</i>	<i>P. hibernalis</i>	<i>P. nicotiana</i> var. <i>parasitica</i>	
<i>P. porri</i>	<i>P. ilicis</i>	<i>P. infestans</i> f. sp. <i>infestans</i>	
<i>P. cyperi bulbosi</i>	<i>P. colocasiae</i>	<i>P. infestans</i> f. sp. <i>thalictri</i>	
<i>P. cyperi</i>	<i>P. verrucosa</i>	<i>P. cinnamomi</i>	
<i>P. lepironiae</i>	<i>P. quininea</i>	<i>P. cambivora</i>	
	<i>P. fragariae</i>	<i>P. cryptogea</i>	
	<i>P. megasperma</i> var. <i>megasperma</i>	<i>P. dreschleri</i>	
	<i>P. megasperma</i> var. <i>sojae</i>		
	<i>P. erythroseptica</i> var. <i>pisi</i>		
	<i>P. richardiae</i>		
	<i>P. vignae</i>		
	<i>P. erythroseptica</i> var. <i>erythroseptica</i>		

**P. phaseoli* which is considered as heterothallic in Waterhouse's key was found homothallic by Savage *et al.* 1968

P. lateralis is given as homothallic by Savage *et al.*, 1968

(Zentmyer, 1952). In A_2 isolates of *P. capsici*, the fungicide "chloroneb" induced sexual reproduction (Noon and Hickmann, 1974). None of those stimulating substances is effective on A_1 isolates.

Recently, it has been shown that sexual reproduction in *Phytophthora* is likely under hormonal regulation (Ko, 1978). Both A_1 and A_2 isolates of *P. cinnamomi*, *P. parasitica* and *P. palmivora* form oospores by selfing when they are paired with different mating types on opposite side of polycarbonate membrane. In this instance, the membrane prevents direct contact between hyphae of the two compatible isolates, and oospore formation on both sides demonstrates the production of diffusible substance like plant hormones, as found in related fungi (*Achlya* sp., Raper, 1951). At least two different hormones seem to be involved in sexual reproduction of heterothallic species. It has been proposed to designate the first one, produced by A_1 isolates, hormone α_1 , and the second, produced by A_2 isolates, hormone α_2 . α_1 induces oospores formation in A_2 isolates, and α_2 induces sexual reproduction in A_1 isolates only.

To date, little or nothing is known on the genetical regulation of the mating types, but it seems possible that they could be determined by a complex system of genes. The reason of that uncertainty is mainly that most genetical studies in the recent years have not been specifically undertaken to elucidate the mode of inheritance of mating types, but to provide information on the life cycle of the fungus.

The life cycle of *Phytophthora* species and consequently the ploidy level of vegetative nuclei have long been a matter of controversy. According to the classical concept, the diploid phase is restricted to the oospore which results from the fusion of haploid gametangial nuclei. Meiosis occurs in the oospore, before its germination, resulting in haploid vegetative mycelium. The alternative theory, proposed by Sansome (1961) and initially founded on cytological data, describes diploid somatic nuclei, and meiosis in the gametangia. Oospore formation results from the fusion of two haploid gametic nuclei and oospore germination results in diploid vegetative mycelium.

After Sansome reopened the debate on the position of meiosis in Peronosporales, several cytologists carried out histochemical studies on different *Phytophthora* species, and provided substantial support to the hypothesis of gametangial meiosis. None of these cytological works, however, was entirely convincing and it was necessary to confirm genetically the cytological evidence for diploidy. Such confirmation was difficult to obtain because genetical studies have long been hampered by difficulties in germinating oospores and in obtaining adequate mutants.

Despite these difficulties, several works were carried out on heterothallic species (Galindo and Zentmyer, 1967; Romero and Erwin, 1969; Laviola, 1969; Satour and Butler, 1968; Timmer *et al.*, 1970). They showed that when isolates differing in several characters are crossed, the progeny resulting from germinated oospores includes recombinants. With one exception (Laviola, 1969), the data also showed a lack of segregation among the progeny from individual oospores. This suggested meiosis in gametangia and germination of diploid oospore rather than zygotic meiosis. The latter would imply that only one of the four meiotic products survived in the germinating oospore: a somewhat complicated explanation. But most of the results remained open to interpretation on either hypothesis, gametangial or zygotic meiosis, and these genetical studies were inconclusive.

The first genetical evidence for diploidy was obtained with *P. dreschleri* Shaw and Khaki (1971). They crossed wild type with mutants resistant to drugs, and obtained segregation ratios consistent with the hypothesis that the somatic nuclei were diploid. It was not possible, however, to draw any definitive conclusion based on their data, since some features remained unexplained: for instance, the non-Mendelian segregation for the mating type that could be due to selfing of the parental strains.

Further informations were obtained from the study of homothallic species. In homothallic species, populations of progeny from sexual and asexual origin, derived from oospores and zoospores of the same clone, can be compared. Variation among both types of populations will be caused by mutation, by heterokaryosis of the parent

clone, by cytoplasmic factors, and by mitotic recombination if the parent clone is diploid. In that case, and if the parent clone is also heterozygous for some loci, the variation among sexual progeny will be greater than that among asexual progeny, since oospores result from fusion of different meiotic products, when zoospores only derive from mitotic division of somatic nuclei.

I found it to be so in *P. syringae* (Boccas, 1972) (Figure 3.1). Over three generations, I compared colony diameter in populations of sexual and asexual progenies and noted highly significant difference in variation between the two kinds of populations. The variation in growth rate among colonies from sexual origin was always greater than that in colonies derived from zoospores. If *P. syringae* were haploid, variations among sexual progeny should not then differ significantly from that among asexual progeny derived from the same parent clone. Therefore, the significant difference in variation between the two kinds of populations was most simply explained by genetic recombination in the oospores, following meiosis in gametangia.

In 1973, Elliot and Macintyre treated zoospores of the homothallic *P. cactorum* with a mutagen and showed that auxotrophic mutant characters, induced in heterozygous condition, segregated in mendelian fashion in subsequent generations. The patterns of inheritance obtained were entirely consistent with the hypothesis of diploid mycelium and meiosis in the gametangia.

More recently, Long and Keen (1977) dealing with another homothallic species, *P. megasperma* var. *sojae*, reached a similar conclusion. They constructed heterokaryons from two different auxotrophic mutants. By self fertilization, these heterokaryons produced parental type auxotrophic progeny and prototrophic progeny, but no double auxotrophic offspring. The selfing of these F₁ monokaryotic prototrophs segregated in F₂ four classes of progeny, i.e. prototrophs, two classes of single auxotrophs and double auxotrophs, as it was expected in a vegetatively diploid organism.

Today, cytological and genetical evidences for diploidy have been accumulated in more than forty species of biflagellate Oomycetes, and in particular in ten homothallic and heterothallic *Phytophthora* species: *P. erythroseptica* (Sansome, 1965), *P. cactorum* (Sansome, 1965; Elliot and Macintyre, 1973), *P. parasitica* (Huguenin and Boccas, 1970), *P. dreschleri* (Shaw and Khaki, 1971), *P. syringae* (Boccas, 1972), *P. cinnamomi*, *P. infestans* (Brasier and Sansome, 1975), *P. capsici* (Main, 1976; Sansome, 1976), *P. palmivora* (Legrand-Pernot and Pellegrin, 1976) and *P. megasperma* (Long and Keen, 1977). Therefore, it seems reasonable to end the controversy and to conclude that *Phytophthora*, like other Oomycetes, are diploid in their vegetative state, with meiosis occurring in oogonia and antheridia, prior to oospore formation.

Since heterothallic species can form oospores in interspecific crosses, the sexual stage is a potential source of variability in the genus *Phytophthora*. Possible exchange of genetic material in interspecific crosses might be an important factor in the evolution of natural *Phytophthora* populations and would raise fundamental questions on the validity of the species concept in the genus. However, until recently, the occurrence of interspecific

hybridization remained hypothetical due to the difficulty to germinate oospores formed in interspecific crosses and to establish viable colonies from such germinations.

I have studied two kinds of interspecific crosses (Boccas, 1980, Boccas and Zentmyer, 1976): crosses between *P. parasitica* and *P. cinnamomi*, which differ markedly in morphology, physiology and pathogenicity, and crosses between more closely related species belonging to the same morphological group of Waterhouse's key. (Table 3.2).

Table 3.2. Interspecific crosses studied

CROSSES BETWEEN SPECIES OF DIFFERENT MORPHOLOGICAL GROUPS		
<i>P. cinnamomi</i> A ₁ × <i>P. parasitica</i> A ₂	43	progeny
<i>P. cinnamomi</i> A ₂ × <i>P. parasitica</i> A ₁	33	progeny
CROSSES BETWEEN CLOSELY RELATED SPECIES		
<i>P. palmivora</i> A ₂ × <i>P. capsici</i> A ₂	18	progeny
<i>P. palmivora</i> A ₂ × <i>P. parasitica</i> A ₂	18	progeny
<i>P. megakarya</i> A ₁ × <i>P. parasitica</i> A ₂	65	progeny
<i>P. parasitica</i> A ₁ × <i>P. capsici</i> A ₂	15	progeny
<i>P. cinnamomi</i> A ₂ × <i>P. cambivora</i> A ₁	6	progeny

Table 3.3. Oospore production and germination in interspecific crosses

Crosses	Oospore production (no. per mm ³ of culture medium)	Oospore germination %
<i>P. cinnamomi</i> A ₂ × <i>P. parasitica</i> A ₁	2	0.2
<i>P. cinnamomi</i> A ₁ × <i>P. parasitica</i> A ₂	3	0.2
<i>P. palmivora</i> A ₂ × <i>P. capsici</i> A ₁	18	0.4
<i>P. palmivora</i> A ₂ × <i>P. parasitica</i> A ₁	62	0.5
<i>P. megakarya</i> A ₂ × <i>P. parasitica</i> A ₁	20	0.5
<i>P. parasitica</i> A ₁ × <i>P. capsici</i> A ₂	24	0.1

All the crosses formed oospores, but the rate of oospore production in crosses between closely related species was about ten times higher than that in crosses between species of different morphological groups. In both kinds of crosses, the percentages of oospore germination were extremely low (from 0.1 to 0.5%) (Table 3.3); nevertheless 198 single oospores isolates from seven crosses were harvested and genetically analysed.

Among that progeny, colony morphology, optimum and maximum temperature for growth, growth rate, pathogenicity and composition of soluble proteins, determined by acrylamide gel electrophoresis, were studied.

Most of the crosses resulted in phenotypically heterogeneous progeny which exhibited recombinations for morphological, physiological and pathogenic characters. This was first interpreted as an indication of interspecific hybridization between the species crossed.

That interpretation, however, was confirmed by the study of protein patterns for only one of the progeny isolates. This single oospore isolate from a cross between *P. capsici* and *P. palmivora* was the only one to exhibit a protein pattern qualitatively and quantitatively different from that of its parents. As it also differed from the parental strains by other characters, it was interpreted as an interspecific hybrid.

But all other phenotypically recombined progeny produced protein patterns parental types, and did not show any indication of recombination for that character, as it was expected in the hypothesis of true hybridization.

Since the composition in soluble proteins of an isolate is assumed to be a significant expression of its genome, the lack of any indication of recombination among the progeny patterns, strongly suggested that no exchange of genetic material occurred between the different species. Therefore, it was concluded, according to the simplest interpretation, that the parental strains were heterozygous for loci controlling those characters showing variation, and that all the progeny, but one, resulted from the self fertilization of the parental strains.

Self fertilization among heterothallic species of *Phytophthora* is a consequence of the potential bisexuality of these organisms. It has been shown that, although less common than hybrid oospores, selfed oospores are formed in both intra and interspecific crosses. However, in interspecific crosses, either between closely related or morphologically distant species most hybrid oospores appear unable to germinate and probably abort because they associate genomes which lack homology. Only selfed oospores may produce viable progeny. Thus, the mating of compatible strains of different species does not generally result in hybrid progeny, but leads to a reciprocal induction of selfing which may result in phenotypic variation of the progeny if the parental strains are heterozygous. From a more practical point of view, it is of particular interest to note that the crossing of two different species, if occurring in nature, may produce a wide range of progeny, greatly variable in morphology, physiology and in pathogenic aggressiveness. This might well contribute to the evolution of *Phytophthora* populations and especially to their pathogenic adaptation.

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PHYSIOLOGIC SPECIALISATION IN *PHYTOPHTHORA* SPECIES

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The genus *Phytophthora* has a wide adaptability and causes diseases of wild and cultivated plants belonging to many families of angiosperms and gymnosperms in tropics, subtropics and temperate areas. About 70 species of the genus have been described (Tucker, 1931; Waterhouse, 1963). In India nearly 20 species of *Phytophthora* have been reported to cause diseases of a number of higher plants (Bilgrami *et al.*, 1979). *P. himalayensis* Dast. was reported from the Simla hills (Dastur, 1948), but Waterhouse relegated it to *P. erythroseptica* (Waterhouse, 1963). However, Dastur could not find any 'Pink rot' in the potatoes affected by *P. himalayensis*. Recently, *P. erythroseptica* has been recorded to cause pink-rot of potatoes in Darjeeling and Simla (Phadtare, 1978 and Rai, 1979).

Some species of *Phytophthora* are very pliable and frequently develop new physiologic races. The best example is found in *Phytophthora infestans* in which a number of new races are recorded every year in different countries. On account of continuous appearance of new pathogenic races the potato varieties bred for blight resistance succumb to the disease within a few years of being brought under general cultivation. A knowledge of the range of variability is the basic requirement for breeding disease resistant varieties for the control of the disease. It will also help in the clarification of the taxonomic position of the species. Variability in some *Phytophthora* species has been reviewed (Erwin *et al.*, 1963). An attempt will be made here to summarise the work on different species in India while briefly mentioning the work carried out in other countries.

Physiologic races

Maximum development of races has been observed in *P. infestans* and it has been reviewed by several workers in almost every country (Graham, 1955; Doling, 1956; Black, 1957; Driver, 1958; Conroy, 1960; and Dutt 1965). Among the species recorded in India, the occurrence of physiologic races has been studied in some of the species. A detailed review of the work done on *P. infestans* besides a brief account of the work done on other important species is presented here.

Phytophthora infestans de Bary

The early works of Giddings and Berg (1919) and Berg (1926) have shown the existence of races, 'Potato race' and 'Tomato race' in *P. infestans*. These two groups of races have been further observed within the original potato race group. In India, the studies on races of the potato blight pathogen were initiated by the author in 1960 (Dutt, 1965) and these studies are now being made annually at Simla, Darjeeling, and Shillong.

Determination and classification of races. In inoculation tests it was observed that the fungus penetrated the tissues of leaves of all the varieties. In some varieties the organism developed and produced sporulation while in others it was localised at the point of infection by the rapid formation of a necrotic barrier. This hypersensitive reaction of some varieties against certain races was made use of in the identification of the races of the fungus. Different workers designated the races differently (Black, 1952; Mastenbroek, 1952; Mills and Peterson, 1952; Darozhkin and Ramneva, 1959; and Schick and Schick, 1959). In order to overcome this confusion an international system of nomenclature was developed (Black et al., 1953; Anonymous, 1954). According to this classification, the race '0' could attack only plants with no 'R' genes while races 1, 2, 3 and 4 attacked the single gene genotypes designated R_1 , R_2 , R_3 and R_4 respectively besides attacking the plants with no 'R' genes. In this system there were 16 different genotypes possessing the major genes single or in combination enabling the identification of 16 different races. The position now has changed with the location of genotypes possessing major genes like R_5 , R_6 , R_7 , R_8 , R_9 , R_{10} and R_{11} . Now there seems to be no end to the races that may develop in *P. infestans* as highly specialised races are appearing continuously.

Inoculation techniques and equipment. Various workers have adopted different techniques and equipments, for collection of isolates, sporulation of samples and inoculation of differential hosts to suit the local conditions. Periodic collection of blight affected leaves from the commercial crop and different hybrid varieties in the potato trials was found very useful for obtaining isolates for study of the racial picture. A bit of the blight lesion from the affected leaves was inoculated on a potato slice in a Petri plate. Heavy sporulation developed in about 5 days. A suspension of the sporangia was prepared in water and kept at 12°C for germination. After germination of sporangia filter paper discs (3 mm) were dipped in the zoospore suspension. On the ventral side of a leaflet of each differential a filter paper disc from the zoospore suspension was placed. The inoculated leaflets were kept in a tray lined with wet moss and kept in a chamber at a temperature of 15°C and relative humidity of about 90%. The observations on the reaction of genotypes were recorded on the third and fifth day for identification of the races.

In studies on determination of races, the condition of the leaves of the differential hosts played a very important part. The senescent and weak leaves of the genotypes possessing R genes may develop infection even by inoculation with race '0'. Tough leaves from plants raised in the field give the best results. However, when the blight occurs in the field it becomes difficult to make use of these plants. The plants raised in the glass-house are often weak and give erratic results.

Distribution of races. Before the late blight resistant varieties of potato were brought under commercial cultivation, race '0' was the common race in most of the countries. With the introduction of resistant varieties race '4' becomes predominant (Dolling, 1956 and Black, 1957). A similar shift in the population of races was observed in North America, South Rhodesia and Israel. In recent years highly specialized races developed with the cultivation of 'R' genotypes, as observed in Mexico and Eastern Canada. With the cultivation of *Solanum tuberosum* varieties not possessing major genes for resistance

more highly specialised races tend to disappear (Black, 1960). The position in India is somewhat similar (Dutt et al. 1973). In 1960, race '0' was determined in 72 samples and race 1 in four samples obtained from Punjab, Himachal Pradesh and Uttar Pradesh. During 1961-1962 race '0' continued to be dominant both in the hills and the plains. race '0' continued to be dominant both in the hills and the plains. Race 1 was observed in some samples from the Simla Hills. Race 1 alone was found in the Nilgiri hills during 1961 when the blight appeared there first time after a lapse of 83 years. Race 4 was determined in samples from Shillong. The position of races continued to be simple till 1966, though race 4 was becoming more common. In 1967 race 4 was prevalent in the Simla hills and race 3, 4 appeared in Shillong and Darjeeling. In the eastern hills specialised races appeared in Shillong and Darjeeling. In the eastern hills specialised races appeared in 1969. From 1973 onwards the occurrence of specialised races became an annual feature both in the western and eastern hills. The position became very serious by 1979 as highly specialized races were recorded (Dutt, 1965; Phadtare and Pushkarnath, 1968; Khanna and Vishwa Dhar, 1975; Bhattacharyya et al. 1976 and Khanna et al. 1977, 1978).

Phytophthora parasitica Dast.

P. parasitica has been recorded on several hosts from different parts of India and quite a few varieties have been recognized within the species which have been listed (Bilgrami et al. 1979). *P. parasitica* Dast. var. *nicotianae* Tuck. causes black shank of tobacco. It has been suggested that it should be changed to *P. nicotianae* var. *nicotianae* (Waterhouse, 1963). Isolates of the pathogen showed variation in their pathogenicity which helped in determination of race '0' and '1' (Apple, 1962). On the basis of resistance in different varieties of tobacco derived from *Nicotiana longiflora*, 3 races of the pathogen were identified (Valleau et al. 1960). Tests with varieties and *Nicotiana* species for their reaction to black shank pathogen made at the Central Tobacco Research Institute, Rajahmundry showed that most of the varieties were susceptible. However, three of the species *N. nesophila*, *N. suaveolens* and *N. wigemdiodes* were immune to the isolate (Anonymous, 1975). If these species are tested against a large number of isolates of the pathogen it may be possible to determine variability in their pathogenicity. Three other varieties of *P. parasitica* have also been reported from India. *P. parasitica* Dast. var. *macrospora* Ashby causes fruit rot of *Annona squamosa* L. and also infects a number of other hosts (Rao, et al. 1963). *P. parasitica* Dast. var. *sesami* Kale & Prasad is specific to *Sesamum orientale* (Kaley and Prasad, 1957). *P. parasitica* var. *piperina* Dast. affects Piper betle as well as a number of other hosts. The work on *P. betle* was reviewed in detail and studies with different isolates did not reveal any racial differences (Saxena, 1977).

Phytophthora palmivora Butler

P. palmivora has been recorded on a number of hosts. *P. arecae* (Colem.) Pethyb. has also been reported on a number of hosts but Tucker has described it as a synonym of *P. palmivora*. Thirty two isolates of *P. palmivora* cultured from various hosts were classified into two compatibility types A₁ and A₂ (Savage et al., 1968). Variations in pathogenicity to okra and castor was also recorded (Ramakrishnan and Seethalaxmi, 1956a). Turner (1960) recorded variation in growth rate of isolates of *P. palmivora* on cacao pods. Similar studies have been initiated at the Central Plantation Crops Research Institute,

Kasaragod. Six isolates were obtained from coconut, rubber, oil palm palmyra, pepper, and areca. The isolate from coconut was most pathogenic on cacao pods (Reddy *et al.* 1976). The pathogen caused blossom blight of *Gerbera* at Pune and was also found pathogenic to the seedlings of castor, cotton, tobacco, and cactus, but non-pathogenic to french beans and chillies (Rao and Ullasa, 1971). *P. palmivora* Butl. var. *piperis* Mull. was recorded on leaves of *Piper nigrum* (Sam Raj and Jose, 1966). The pepper isolates have been reported to infect *Piper longum*, fruits of *Areca catechu* and *Theobroma cacao* (Nambiar *et al.*, 1976).

Origin of physiologic races

The variability in a species of *Phytophthora* may result from adaptive parasitism, mutation, heterokaryosis and parasexuality, and sexuality. Very little work has been done on this aspect. The information available is briefly given here.

Adaptive parasitism. The development of the races on account of serial passage through foliage of varieties increasing in level of resistance has been explained to be on account of 'adaptive parasitism' (Mills, 1940) also known as 'Physiologic adaptation' or 'building up' of virulence (Reddick and Mills, 1938). The author inoculated old leaves of R₂ genotypes with races '0' of *P. infestans*. By serially culturing the isolates for six times on leaves of R₂ genotypes a very heavy sporulation was observed even on tough leaves of the genotypes. This was identified as race '4'. Similarly starting with race '0' race, 2, 4 was developed. The phenomenon has been explained on account of physiologic adaptation but it may not exclude the possibility of being due to mutation (Black, 1954). In nature also it has been observed that new races of *P. infestans* developed as soon as the blight resistant varieties were cultivated in the area. At Simla race '0' was the common race when *Solanum tuberosum* varieties were under cultivation, but with the introduction of a potato hybrid S. 1756, an R₁ genotype race 1 appeared in the crop. Similarly, later race 4 developed in the area. The racial picture changed in Shillong and Darjeeling from simple races to very highly specialized races with the introduction of blight resistant varieties like Kufri Jyoti and Kufri Naveen. Black (1952) indicated that the climatic factors might effect the vigour and reproductive powers of physiologic races of *P. infestans*. Some of the workers reported that there did not exist any correlation between physiologic races and climatic conditions (Kedar *et al.*, 1959). However, observations in India indicated that weather conditions had a great bearing on the occurrence of new races of the blight pathogen (Dutt *et al.*, 1973). In the eastern hills the wet and cool conditions continue for a few months from April onwards. Such conditions are ideal for the development of the blight and quick reproduction of the pathogen enabling completion of several infection cycles starting from the initial occurrence of the disease. Thus in this serial passage the pathogen gets a chance of building up, resulting in the development of a number of new races every year. On the other hand in the plains the weather conditions favouring the development of blight may not remain available for more than a week. The initially developed sporangia may not, thus, get a chance of serial passage and are not able to develop into new races. This may be one of the factors for racial position in the plains to continue to be simple.

Mutation. Variations in *Phytophthora* species may develop through mutation. Mutants have been induced in *P. cactorum* by subjecting zoospores to ultra-violet light and X-ray

(Buddenhagen, 1957). Similarly in *P. infestans* mutants were induced by exposing zoospores to ultra-violet light (Wilde, 1961). In *P. infestans* second or even third order races have arisen directly from the common race omitting the intermediate places (Black, 1954). Mutability in *P. infestans* resulting in the origin of very highly specialized races has been observed on blight resistant selections of potato and tomato (Graham *et al.* 1961). Similarly changes in the pathogenicity have also been observed to take place in opposite direction.

Heterokaryosis and parasexual recombination. The change in the epidemiology of the potato late blight fungus with the change in the varietal pattern may be due to heterokaryosis or parasexuality. The hyphae may anastomose and the nuclei of the two hyphae may intermingle (Wilde, 1961). New races were also obtained by inoculating simultaneously two races of the same mating type on potato plants. This may be as a result of parasexual recombination or heterokaryosis since no sexual recombination was involved.

Sexuality. The origin of new races is mainly due to genetic recombination on account of hybridization of the compatible group of races. Sexual stage of *P. infestans* was recorded in Mexico (Galindo and Gallegly, 1958; Niederhauser, 1956). Two compatible groups of races designated as A₁ and A₂ have been isolated and by pairing these, oospores developed in cultures as well as in foliage. It has been observed that all the potato blight races in British Isles, western Europe, Africa, North America and Asia including India belong to A₁ group and are sexually incompatible. This explains the absence of oospores of the pathogen in these areas. The oospores commonly occur in Mexico and South America and that may be one of the factors responsible for existence of maximum number of physiologic races in these areas. Heterothallic strains in the genus *Phytophthora* have been recorded in Mysore (Narasimhan, 1930). *P. parasitica* is heterothallic but is sexually compatible with other types or species. Sexuality was studied in detail in *P. infestans* and also in 30 other species of *Phytophthora* have been recorded on *Areca catechu* (Rama-krishnan and Seethalaxmi, 1956).

DISCUSSION

A good amount of work has been done on the host range of species of *Phytophthora* in India. However, there may be some confusion about the identification of the species on different hosts. *P. palmivora* was recorded on *Colocasia* and it was suggested that the same species may be affecting some of the hosts which had previously been recorded to be affected by *P. colocasiae* (Umabala and Ramarao, 1972). A good knowledge of the range of variability of the different species will help in solving the taxonomic position. The morphologic characters of the fungus on the different hosts and culture media have been studied but the different isolates need to be studied in detail by cross inoculation on varieties and genotypes possessing immunity or high degree of resistance towards specific isolates. These resistant types will form a good basic material for studying the racial position in the species.

Standardization of inoculation techniques taking into consideration the load of inoculum, mode of inoculation, temperature and humidity, use of different parts of the host

plants at different stages of growth will prove very useful in the study of physiologic races in different isolates.

Useful information on *P. palmivora*, *P. parasitica* and *P. colocasiae* has been secured by different workers at various places. A concerted effort at one centre using one particular species and isolates from different locations and varieties will help in clarifying the situation regarding the position of the different races. Thus one centre will form a testing ground for all the hosts affected by a species. For example, the entire work on the study of the variability in *P. palmivora* can be concentrated at the Central Plantation Crops Research Institute, some varieties of tobacco and *Nicotiana* species possessing resistance as also immunity have also been located at the Central Tobacco Research Institute, where the studies on the species and its varieties may be taken up as a whole in the form of a project.

The system of the designation of races has been very well developed in case of *P. infestans*. It should serve as a model for work on other species. An effort should be made to locate hosts with single gene to serve the need of classifying the races. In the absence of that, specific races may be designated on the basis of their reaction on particular hosts.

The mechanism of variability needs to be studied to form a correct idea about the origin of races. Study of induced mutations will indicate the possibility of variability and that may be correlated with the natural variation. However, care has to be taken to avoid the release of races developed in the laboratory. Studies on anastomosis of hyphae and zoospores will help in understanding heterokaryosis and parasexual recombination. Similarly the phenomenon of origin of races due to physiologic adaptation also deserves to be taken up in more detail. Studies on mating of different isolates and germination of oospores will also provide information on variability due to sexuality.

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DISCUSSIONS

H. S. Sohi: In certain crops the entire germplasm in the country is susceptible. How do we go about in finding races?

ANSWER: We may take up annuals, both wild and cultivated, inoculate the leaflets under controlled conditions and then try to locate the races.

S. Y. Padmanabhan: Certain amount of variability occurs in nature. Under favourable conditions the races get sorted out. Mutation may also occur. What is the use of inoculating plants which are not useful?

ANSWER: The purpose is to locate variation in isolates. It will also help in sorting out the species.

S. Y. Padmanabhan: Interesting academic pursuit; but may not be useful in breeding programme.

D. N. Srivastava: Overloading the plants with inoculum is not useful. However, areas to sort out the host range of different isolates should be pursued.

RACES OF *PHYTOPHTHORA INFESTANS* RECORDED IN INDIA

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ABSTRACT

The racial pattern of *Phytophthora infestans* (Mont.) de Bary was simple in this country until 1966. Only the races 0 and 4 were recorded from the North-western, 0; 1 and 4 in the North-eastern, 1 in Nilgiri hills and 0 in the northern plains. With the advent and adoption of cultivars having R-gene resistance after 1966, new and complex races started appearing. Two to three gene complex races were first observed in the North-eastern hills in 1967. Subsequently more complex gene races developed and predominated at Shillong, Darjeeling and Simla making them hot spots. So far 74 races of *P. infestans* have been identified. First appearance of a 7-gene complex race (1, 2, 3, 4, 5, 6, 7) was recorded from Shillong in 1976. Furthermore, 8-gene complex races were reported from Shillong and Darjeeling (1, 2, 3, 4, 5, 6, 7, 8) in 1978 and from Simla (1, 2, 3, 4, 5, 7, 8, 11) in 1979. In the plains, due to the cultivation of late blight-susceptible cultivars only simple races have been observed so far.

INTRODUCTION

Late blight disease of potato caused by *Phytophthora infestans* (Mont.) de Bary appears in epiphytotic form every year in the hills of India causing 20-70% losses in yield depending upon the weather conditions, time of appearance and varieties (Dutt *et al.*, 1978).

From time to time workers from different parts of this country reported several physiological races of the pathogen (Bhattacharyya *et al.*, 1976; Dutt, 1965; Dutt *et al.*, 1973; Khanna and Vishwa Dhar, 1975; Khanna *et al.*, 1977; Phadtare and Pushkarnath, 1968; Phadtare and Sharma, 1970; Phadtare *et al.*, 1971, 1973). Scattered information in different and easily unavailable publications on the spectrum of physiological races of the late blight pathogen in India has been compiled in the present paper. The latest unpublished findings have also been included.

MATERIALS AND METHODS

The blight-infested potato leaves were collected from different potato varieties and hybrids from different localities. The sporangia from the infected leaves were either taken directly for zoospore release for determination of races or a bit of the blight lesion from the affected leaves was inoculated on a potato slice in a Petri plate and incubated at $16 \pm 1^\circ\text{C}$ under high humidity. Heavy sporulation developed in about 4-5 days. The sporangia so obtained either on infected leaves or on tuber slices were kept in sterilized water at $13 \pm 1^\circ\text{C}$ for 1 to $1\frac{1}{2}$ hr for the release of zoospores.

The late blight differentials maintained at CPRI, Simla, CPRS-Darjeeling, Shillong and Ootacamund were employed for identification of the races. Tubers of susceptible variety inoculated with the pathogen in the plains were received at Simla for race identification.

Mature leaflets (at least three) of each of the 23 differentials (R_2 to R_{11} and combinations of R_1 to R_2) were taken and arranged in wet moss lined wooden/enamelled tray keeping the under surface upwards. Two filter paper discs (5 mm diam.) soaked in the zoospore suspension were placed on either side of the midrib. The tray was covered with a hard board and incubated at $16 \pm 1^\circ\text{C}$. After 5 to 7 days observations were recorded for disease symptoms. The inoculum taken again from infected leaflets was used for inoculation of all the late blight differentials to confirm whether it was a single simple or complex race or a mixture of races. The distribution of the races recorded from 1965 to 1979 at different places is presented in Tables 3.4a, 4b and 4c.

Table 3.4a. Year to year distribution of the races of *Phytophthora infestans* at different hilly regions in India

Races	Years												
	1965	1966	1967	1968	1969	1970	1971-1973	1974	1975	1976	1977	1978	1979
0	S	S	S,D, SH	S	S,O, SH	S,M, SH	S	S	S,D,	..	S,D	..	SH
1	..	S,O	SH,D	S	S,SH	..	D
3	D	S
4	..	S	S,D SH	S,D	S,D	S	S,D SH	S,D SH	S,D	SH, D	SH, D	S	S,D SH
7	S
8	S	..	S
10	S
11	S	S
1,2	SH
1,3	S,O
1,4	D,SH	..	D	S,D SH	D,SH	SH	SH	D
2,3	S
2,4	D	..	D	D	D	D	D	..	S,D	..	D
3,4	D	S	..	S
3,7	D	D	S	..	D
3,8	S
3,9	S
4,8	S
4,11	S	..
7,8	S
7,9	S
7,11	S
8,9	S
10,11	S
1,2,4	SH,D	D	SH,D	..	SH,D	SH	D	SH,D	D

S - Simla, SH - Shillong, D - Darjeeling, M - Mukteswar,
O - Ootacamund/Kodaikanal.

Table 3.4b. Year to year distribution of the races of *Phytophthora infestans* at different hilly regions in India*

Races	Years								
	1969	1970	1971-1973	1974	1975	1976	1977	1978	1979
1,3,4	D
1,3,7	S
1,3,8	O
1,4,5	SH	SH	SH
1,4,7	SH	SH
1,7,8	M
1,8,10	S
2,3,7	..	D
3,4,7	SH,D	D	SH	S,D SH	S,SH	S,SH	D,SH	S,SH	D,SH
3,7,8	S	SH	..
3,7,9	S
4,7,11	S	..
4,10,11	SD
7,8,9	S
8,10,11	S
1,2,3,4	S
1,2,4,5	SH
1,2,4,7	SH	SH	SH
1,3,4,5	SH
1,3,4,7	SH	SH	SH	SH	SH
1,4,10,11	S
2,3,4,7	SH	SH	SH	..	D
3,4,7,8	S,SH	SH	SH,D	..	SH
3,4,7,9	S
3,4,7,11	S	..

*Not observed between 1965-1968

S - Simla, SH - Shillong, D - Darjeeling, M - Mukteswar,
O - Ootacamund/Kodaikanal.

Table 3.4c. Year to year distribution of the races of *Phytophthora infestans* at different hilly regions in India*

Races	Years				
	1975	1976	1977	1978	1979
3,7,10,11	D
1,2,3,4,5	D	..
1,2,4,5,6	..	SH	SH
1,2,4,5,7	..	SH
1,2,4,10,11	D
1,3,4,7,8	SH
1,3,4,10,11	S
2,3,4,7,8	D	SH,D	D
2,3,4,7,11	D
3,4,7,8,9	S
3,4,7,8,10	S
3,4,7,8,11	S	..
3,4,7,10,11	D
1,2,3,4,6,7	SH
1,2,3,4,7,8	D	D
1,2,4,5,10,11	D
1,3,4,5,6,7	..	SH
1,3,4,7,10,11	D
1,2,3,4,5,6,7	SH	SH	SH
1,2,4,5,8,10,11	D
1,3,4,5,6,7,8	SH
1,2,3,4,5,6,7,8	SH,D	..
1,2,3,4,5,7,8,11	S

*Not observed between 1965-1974

S - Simla, SH - Shillong, D - Darjeeling, M - Mukteswar,
O - Ootacamund/Kodaikanal.

OBSERVATIONS

The racial picture in general up to 1967 (Table 3.4a) was very simple. Races 0, 1 and 4 were observed at Simla and Shillong and race 1 at Ootacamund. Complex races with two gene combination (2, 4 and 3, 4) were first recorded in Darjeeling in 1967. In 1969, 2-gene (1, 2 and 1, 4) and 3-gene (1, 2, 4 and 3, 4, 7) races appeared in Shillong and Darjeeling in addition to the existing races (0; 1; 3; 4; 2, 4;) but the race 3,4 was not observed later on. Races 4; 1, 4; 1, 2, 4 and 3, 4, 7 became more predominant in these areas and it appeared almost in all the years from 1969 onwards. At Simla, appearance of 2 and 3 gene complex races (1, 3; 1, 4 and 1, 3, 7) was first noticed in 1971-73 and race 3, 4, 7 in 1974. The racial picture in Simla became more complicated

since 1975 when 2 to 5-gene complex races were identified. In the same year four 4-gene complex races (1, 2, 4, 7; 1, 3, 4, 7; 2, 3, 4, 7 and 3, 4, 7, 8) were also identified from Shillong. The racial picture of Shillong became more complex since 1976 when 5 to 7-gene complex races (1, 2, 4, 5, 6; 1, 2, 4, 5, 7; 1, 3, 4, 5, 6, 7 and 1, 3, 4, 5, 6, 7, 8) were identified. However, except 1, 2, 4, 5, 6, the other three above mentioned races were not encountered in subsequent years. Races 1, 3, 4, 7, 8 and 1, 2, 3, 4, 5, 6, 7 appeared in 1977 and race 1, 2, 3, 4, 5, 6, 7, 8 was recorded in 1978 in addition to race 1, 2, 3, 4, 5, 6, 7. The racial picture in Darjeeling followed more or less a similar pattern as that of Shillong in 1978 and 1979. The most complex race recorded at Darjeeling and Shillong in 1978 was 1, 2, 3, 4, 5, 6, 7, 8 and at Simla was 1, 2, 3, 4, 5, 7, 8, 11 in 1979. However the most predominant races in latter years at Shillong were 4; 1, 4; 1, 2, 4; 3, 4, 7; 1, 3, 4, 7; 3, 4, 7, 8 and 1, 2, 3, 4, 5, 6, 7; and at Darjeeling 4; 2, 4; 1, 2, 4; 2, 3, 4, 7, 8 and 1, 2, 3, 4, 7, 8 in addition to the new races recorded during the year. At Simla races 4; 11; 3, 4, 7; predominated though more new races appeared in 1978 and 1979,

The racial pictures in Ootacamund/Kodaikanal and Mukteshwar hills could not be analysed properly due to non-availability of data/samples but 3-gene complex races were identified both from -Kodaikanal (1, 3, 8) and Mukteshwar hills (1, 7, 8) in 1975.

Races from the northern plains were analysed regularly and races 0 or 4 were identified from most of the places. Further, race 11 was also indentified from Jullundur and Patna in 1979.

It was interesting to note that only race '0' was identified from a locality in Shillong hills in 1979 where only late blight susceptible varieties were being cropped continuously up to 1979.

DISCUSSION

The complex races started appearing since 1966 when the late blight resistance screening programme started in this country incorporating major genes for resistance. The appearance of complex races was recorded first from the North eastern region where climatic factors were more congenial. Moreover the association of the host and parasite exists for a longer period, as the crop in this region is available from March to October with congenial temperature and high humidity from May to October enabling the fungus to produce several spore germination processes and establish in the host. Similar conditions are also prevailing in the Simla hills. The variety Kufri Jyothi which was resistant to late blight since 1965 broke down in 1973 in the north eastern hills and in 1975 the Simla hills indicating the adaptive mutation or vegetative hybridization of the pathogen (Mills and Peterson, 1952; Malcolmson, 1970). As more and more R-gene resistance was employed in the breeding programme more complex races developed in this country making the racial picture more complex. In the absence of A_1 and A_2 mating types in this country, the development of new races might have occurred due to adaptive mutation and hybridization (Graham *et al.*, 1961).

It is therefore, recommended to incorporate minor genes for imparting late blight resistance in the cultivars rather than using the major genes for resistance. This would enable the pathogen to survive on the host with very limited infection and would prevent development of complex races either by adaptive mutation or vegetative hybridization.

The prevalence of large number of highly evolved complex races in our country may be harnessed for using their endemic sites as hot spots for late blight screening especially for true and dependable degree of field resistance.

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DISCUSSIONS

D. H. Lapwood: The present trend is to go for multigene resistance. Is it not good to locate complex genes and subsequently go for resistance to these?

ANSWER: In our breeding programme also by the elimination of R genes, complex genes are developing.

D. H. Lapwood: By introducing more races, you may be eliminating many of them and finally you may be getting only one dominating gene and that may multiply.

THE EFFECT OF SOURCE OF ISOLATES ON THE VIRULENCE OF *PHYTOPHTHORA PALMIVORA* (BUTL.) BUTL.

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ABSTRACT

The time for the appearance of black pod lesions, sporangia production and girdling of cocoa pods wound inoculated with 97 isolates of *Phytophthora palmivora* (Butl.) Butl. were not affected by the susceptibility ratings of the parents from which the crosses were made and the type of cocoa as well as the ecological zone from which the isolates were collected. Lesions appeared on 86% of the 388 inoculated pods in 48 hr, but 58.8% 26.8% and 13.4% of the 97 isolates respectively formed sporangia in 4-5 days, 3-4 days and 5-6 days after inoculation. Upto 54% of the pods were girdled in 7 days. Lesions produced within three days were significantly different ($P < 0.05$) but the effects were not sustained over the experimental period. Isolates from Amelonado \times Trinitario crosses formed large lesions whereas those from Trinitario crosses formed small lesions. Similarly isolates from Resistant \times Susceptible crosses formed large lesions and those from resistant crosses formed small lesions.

INTRODUCTION

Progenies in the Obomeng black pod resistance trial were selected on the basis of the field performance of their parents to the cocoa black pod disease caused by *Phytophthora palmivora* (Butl.) Butl. (Wharton, 1962). As resistance to black pod is a heritable genetic character (Asare-Nyako, Amponsah and Nuamah, 1972) it was expected that progenies obtained by selfing a resistant parent or by crossing two resistant parents would be less diseased than those obtained by crossing resistant and susceptible parents or selfing susceptible parents. But analysis of ten years' records of the Obomeng trial (Dakwa, 1978) showed that black pod incidence was high on all progenies and differences, particularly during the epiphytotic period, were not generally significant. Apart from the weather and the inherent genetic characteristics, a factor that could probably and partly account for the high disease incidence at Obomeng compared to Tafo is the differences in virulence of *P. palmivora* occurring at Obomeng where the trial was sited and at Tafo where the progenies were selected. This paper reports the results of infection studies that compared isolates of the fungus collected from the two places.

MATERIALS AND METHODS

Isolates of *P. palmivora*: The cocoa progenies from which the 97 isolates of *P. palmivora* were collected and the number of isolates from each progeny tested are sum-

marized in Table-I. The isolates on A to Z were collected from Obomeng and those on WEA2 to TF1 were collected from Tafo.

Isolation and maintenance of isolates : The isolates were collected early in the black pod season (March/April) when environmental conditions precluded secondary infections from primary lesions. Pods with about one-third of their proximal lengths showing black pod symptoms were collected in April, 1974, from all the progenies in the Obomeng black pod variety trial (Wharton, 1962) and also from the second clonal trial at Tafo, Ghana (Wharton, 1961). The pods were then incubated in moist polythene bags for up to 48 h in the laboratory ($26 \pm 2^\circ\text{C}$) to induce sporangia production. After confirming the identity of the fungus by microscopic examination of the sporangia, each isolate was sub-cultured by transferring sporangia into wounds made into pods of the progeny from which the fungus was isolated. Cultures were thus maintained until inoculations were made in the field.

Inoculation : Four days' old nonsporulating pod cultures were used as inoculum. Holes were punched at the middle of attached mature green pods by removing tissues (5mm diameter) with No. 1 cork borer. The plug holes were immediately replaced by inoculum of the same dimension. The inocula were not protected from the environment.

Design of experiments : A randomised block design with four replications was used. Suitable pods on twenty cocoa trees, in two rows of ten, were inoculated. Healthy pods with almost the same size and age were, as far as possible, used. The isolates were completely randomised in each block so that it was possible for isolates from the same progeny to occur on the same tree.

Recording of virulence : Virulence was estimated by recording the time for (a) the appearance of lesions, (b) the production of sporangia (c) lesions to girdle pods and (d) lesion sizes along the proximal-distal axis (length) and along the circumference (width) of pods. The daily increases in lesion sizes were recorded for seven days when most pods had been girdled.

Statistical analysis of results : Analysis of variance was done on the daily untransformed lesion sizes to establish possible significant differences that might exist among (a) the 97 isolates (b) the four cocoa types (Trinitario, Trinitario x Amazon, Trinitario x Amelonado and Amelonado) (c) the four susceptibility ratings (Resistant, Resistant x Resistant, Resistant x Susceptible and Susceptible), (d) the thirty progenies (A-TF₁) and (e) between Tafo (WEA₂-TF₁) and Obomeng (A-Z), the sites from which the isolates were collected.

RESULTS

Time for the appearance of lesions, sporulation and girdling of pods : Lesions appeared in 48 h on 86% of the 388 pods inoculated and there were no differences between isolates collected from cocoa trees obtained from susceptible or resistant parents nor from Amelonado nor Trinitario parents. Similarly the time for producing spores was not influenced

by any of these parental characteristics as fifty seven of the ninety seven isolates from all the progenies formed spores 4-5 days after inoculation whereas twenty six and thirteen isolates respectively formed spores 3-4 and 5-6 days after inoculation. Only one isolate from WEA₃ formed sporangia in seven days. Some 54.3% of the inoculated pods were girdled but was unaffected by parental characteristics. There were no differences between Tafo and Obomeng isolates when any of the characters used to assess virulence was considered.

Lesion sizes :

Isolates : Significant differences in lesion sizes occurred between isolates three days after inoculation but these effects were not sustained over the experimental period. The two X isolates formed small lesions. Three of the Z isolates produced lesions that were

Table 3.5. Details of crosses from which *P. palmivora* was isolated

Cross	Code letter	Parent type	Susceptibility rating of parent*	No. of isolates studied
D70 × S27	A	T × T**	R × R***	3
S27 selfed	B	T	R	3
P30 × K5	C	Am × T	R? × R	3
D70 × Na32	D	T × Az	R × S	4
P30 × ACu85	E	Am × T	R? × R	2
Unselected Amelonado	F	Am	S	4
K5 selfed	G	T	R	3
Na 32 × S27	H	Az × T	S × R	3
K5 × Y44	J	T × T	R × R	2
P30 selfed	K	Am	R?	3
ACu85 × A12	L	T × T	R × S	4
D70 × A12	N	T × T	R × S	3
ACu85 × S27	P	T × T	R × R	4
D70 selfed	R	T	R	2
ACu85 × Y44	S	T × T	R × R	3
D70 × ACu85	T	T × T	R × R	3
ACu85 × K5	V	T × T	R × R	3
Y44 selfed	W	T	R	4
12B	X	T	R	2
A12 selfed	Y	T	S	4
Unspecified Amazon	Z	Az	S	4
T79/467 × S72: J114/49	WEA ₂	T × Am	Not classified	3
T85/799 × S84: E104/90	WEA ₃	T × Am	"	4
T16/613 × P3/7: J104/46	WEA ₄	T × Am	"	2
S84: E104/90 × T85/799	WEE ₁₁	Am × Az	"	4
T79/416 × E1: J92/70	WEB ₁	Az × T	"	4
T79/467 × E9: B31/95	WEB ₂	Az × T	"	4
T63/967 × W41	WEB ₃	Az × T	"	4
W41 × T63/967	WBE ₆	T × Az	"	4
TF1 selfed	TF ₁	T	"	2

*According to Wharton, 1962 **T-Trinitario; Am-Amelonado; Az-Amazon

***R-Resistant; S-Susceptible

bigger than one isolate but three of the Y isolates formed smaller lesions than one isolate. The smallest lesion of 24.3 mm was formed by a Y isolate while the biggest one was formed by a E isolate. This was the general pattern among isolates from the same progeny.

Progenies : Differences in lesion sizes formed by the isolates based on the 30 cocoa progenies (Table 3.5) were significantly different only three days after inoculation ($P=0.05$). The isolates from E,A,C,D, and WEA₄ produced large lesions (39–41 mm) whereas those from X and R formed small lesions (27–32 mm). The X isolates formed lesions that were significantly smaller than those from the other progenies; the R isolates also formed smaller lesions than A,C,D,E,J,K,L,N, WEB₁, WEB₂ and WEA₄. The E isolates however formed bigger lesions than the others. Certain isolates, particularly those from D,G,J,R, and WEA₃, maintained fast growth rates between the first and second or the second and third days (FIG. 3.2); others notably A,K,L,N,P,R and WEB₂, developed slowly within the same period. Isolates which showed slow rates of lesion development generally had an initial high rate (e.g. A,K,N,X and WEB₂) and those with fast rates showed an initial slow rate (e.g. G,J and R). While most isolates maintained a fairly consistent rate of lesion development, a few others such as WEB₃ and V fluctuated very much. These differences were not borne out by the cumulative lesion sizes.

Cocoa type : Isolates from the Amelonado x Trinitario crosses formed lesions that were significantly longer than the other three crosses three days after inoculation. All the isolates, except those from Amelonado (FIG. 3.3A), maintained an increased rate of lesion development between the second and third day but the rates dropped between the third and fourth day. The drop was particularly conspicuous in the Amelonado x Trinitario (FIG. 3.3C) and Trinitario (FIG. 3.3G) isolates.

Susceptibility ratings: Lesions produced in three days by isolates from R x R or R x S crosses were significantly longer than those from R or S crosses. The R x S isolates again formed longer lesions than R or S isolates four days after inoculation but these differences were not subsequently sustained. Isolates from the R and R and R x R crosses formed lesions that increased rapidly before slowing down particularly between the second and third days (FIG. 3.4). The R x S and S isolates, however, maintained a slow rate of lesion development.

Sites : Isolates collected from Tafo formed lesions that were not significantly different from those collected from Obomeng. The Tafo isolates formed lesions that were always slightly longer than the Obomeng isolates but the latter isolates formed slightly wider lesions than the former isolates.

DISCUSSION

Results of the inoculation studies showed that the times for the appearance of lesions, for sporangia production and for lesions to girdle wound inoculated cocoa pods were not affected by the source from which *P. palmivora* isolates were collected. However, lesions produced three days after inoculation were significantly different probably as a result of the differences in rates at which the isolates established themselves for these effects were

not sustained over the experimental period. It is, therefore, probable that when an inoculation technique which does not involve wounding the pod is developed, differences in the ability to establish infections may become apparent. It may also demonstrate clearly the significance of the epidermis in the infection process.

The rates at which lesions developed after infections were established differed very much (FIGS. 3.2, 3.3 & 3.4). Thus the rate of lesion development instead of the cumulative lesion sizes could be a useful variate in black pod infection studies.

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DISCUSSIONS

S. Y. Padmanabhan: You could not get resistance in the progenies more than the parents. This may be because no new or additional genes are present in the progenies. Only original genes might be present.

T. N. Sreenivasan: Your report is contradictory to an earlier one on the same aspect.

ANSWER: I studied isolates collected from cultivated farms, abandoned farms and forests. The isolates from cultivated farms were more virulent than those from abandoned farms or forests.

H. S. Sohi: In tomato, bell pepper etc. green fruits are susceptible. Once they change colour, they are not susceptible. Do you find anything like this in cacao.

ANSWER: No. Fruits of all ages are susceptible.

T. N. Sreenivasan: Mature fruits are more susceptible.

D. H. Lapwood: Are old pods susceptible?

ANSWER: Yes. If they are not harvested in time, the pods will get covered by the fungal growth.

Koti Reddy: Are there resistant varieties available?

ANSWER: Yes. Acu 84, Y44 and some of the crosses like S27. But these are not used, because they come to bearing rather late (about 7 years). We rely on hybrids which come to bearing in 3 yrs., but they are susceptible.

Sunday, 21 September, 1980
9.30 a.m. — 12.00 noon

SESSION 4



SCREENING METHODS AND CONTROL

Chairman: **Dr. C. S. VENKATA RAM**

Rapporteur: **Mr. THOMAS JOSEPH**

AGRONOMIC PROBLEMS OF COCOA PHYTOPHTHORA POD ROT CONTROL

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ABSTRACT

An attempt has been made to highlight the influence of environmental features such as rainfall, temperature, relative humidity and soil on the incidence, development and spread of Phytophthora pod rot (blackpod) disease of cocoa. The contributions of factors like habit of growth, vegetative behaviour, flushing, flower and fruiting characteristics to the epidemiology and control of the disease were also discussed. A critical appraisal of current control measures formed the basis of a number of proposals for future research on better disease control.

INTRODUCTION

The aim of this contribution is to discuss in general terms to what extent various agronomic factors influence the development of Phytophthora pod rot disease of cocoa and to highlight what efforts have been made at solving the problems arising therefrom.

Cacao (*Theobroma cacao* L.) is a typical crop of the humid tropics and is commercially grown within a fairly narrow belt about 15° North and South of the equator. Apart from this general zoning the crop has been successfully grown in some sub-tropical latitudes such as the lowlands of the State of Sao Paulo, Brazil at 23-25°C. Traditionally cocoa is grown in relatively low altitudes, usually below 300 m, but it has been found in higher altitudes in places near the equator, as for example in Chama valley of Venezuela (900m), in Colombia (1,000-1,200m) and in Uganda (1,100-1,300 m). The yield capacity of cocoa is determined by the structure and functioning of the tree which are in turn determined by the available environmental resources and conditions, the genotype and by the interactions. In order to maximise yield it is necessary to grow genotypes with high yielding potentials in an environment and with a system of management which allows the tree to express its genetic potential.

For successful establishment and spread a pathogen needs an accommodating (susceptible) host and suitable environmental conditions. Susceptibility of a plant to a pathogen is governed by some innate genetic factors which are invariably the end results of some physiological and biochemical activities within the plant. In practice the main ways by which such functioning of a particular genotype can be controlled are by adjusting environmental conditions. One can thus understand why management constitutes a very important aspect of crop production.

An outline of the environmental requirements and system of management

- (i) **Temperature:** The mean annual optimum temperature for cocoa is about 25°C. It is unusual for cocoa to thrive well below 21°C. While the average monthly minimum should be above 10°C (Alvim and Pereira, 1972). The minimum temperature for good growth of *Phytophthora palmivora* (Bult.) Bult. is between 8-11°C (10-11°C for MF1 and MF3; 8°C for MF4). The optimum is between 24-30°C (28-30°C for MF1 and MF4, and 24-26°C for MF3) (Gregory and Maddison, 1980). The temperature which supports good growth of cocoa thus appears to be very suitable for *P. palmivora*, especially MF3.
- (ii) **Rainfall:** It is accepted that the annual rainfall in most areas where cocoa is successfully grown lies between 1,400-2,000 mm. The annual minimum precipitation is said to be around 1,250 mm; rainfall is supplemented with irrigation or in areas where the ground water table is relatively high. One of the major disadvantages of high rainfall in cocoa growing areas is the maintenance beneath the canopy in the cocoa plantation of a humidity favourable for the development of cryptogamic diseases in particular *P. palmivora* (Braudeau, 1974). Spores of *P. palmivora* are known to be rain dispersed and readily germinate in water droplets on the pod surface to initiate fresh infections (Gregory and Maddison, 1980). Rainfall influences incidence and spread of *P. palmivora* not only directly but also by creating and sustaining high humidity which is essential for the development of the disease. In this regard Wood (1974) confirmed Dade's (1927) assertion that atmospheric humidity is probably the most important factor affecting the incidence of blackpod disease.
- (iii) **Soil:** A number of different types of soils are adequate for growing cocoa successfully provided they have some physical and chemical properties of particular importance for the crop (Hardy, 1960; Smyth, 1966). Such properties, especially physical ones, must be considered in connection with climate, particularly the volume and distribution of rainfall. According to Alvim and Kozlowsky (1977) an ideal soil for cacao must possess good moisture retention, good drainage and good aeration, these being the essential factors for unrestricted root growth. When there is adequate rainfall and vegetative cover over soil with good moisture retention, humidity within the plantation will be high and thus favourable for the development of blackpod disease.
- (iv) **Shade and canopy formation:** In Africa, cocoa is planted under controlled forest with utilization of natural cover. The forest is simply thinned and the forest undergrowth is underbrushed, leaving in place the trees needed to shade the future plantation. A number of the remaining forest trees are felled, at intervals, as the cocoa trees unite to form a continuous canopy which naturally eliminates all growth of weeds and the soil then become covered by a thick carpet of dead leaves. This layer of dead leaves prevents the soil and plant debris like pod husks in which *P. palmivora* thrives saprophytically, from drying off and thus serve as source of primary inoculum after the dry season.

In America and in some plantations in Nigeria the forest is first clean-felled before planting. Artificial shading is then necessary since it is known that during the early stages of growth the young cocoa plant needs, for optimum growth, relatively dense shade letting

through only about 25-50% of the total light. This shade serves to protect the soil and perhaps also provides adequate cover for the growth of microorganisms including *P. palmivora*. In this regard, Gorenz and Okaisabor (1971) observed a severe incidence of pod rot disease inspite of frequent fungicide sprays in an experimental farm with a good two-storied canopy whereas an adjacent unsprayed peasant cocoa farm with a thin open canopy had very little infection. Shade removal has thus been recognised as one of the control measures for blackpod disease. However, many farms in Nigeria often suffer from severe attacks by mirids and thrips resulting in complete destruction of the entire farm after complete shade removal. Some form of over-canopy shading is thus thought to be desirable even after the canopies of the trees in the plantation have met and interlaced with each other to provide significant self-shading. According to Alvim and Pereira (1972) there are an average of 79 ± 25 shade trees per hectare in traditional Bahia farms. Thus cocoa farmers must of necessity find solutions to blackpod disease problems arising from self-shading and over-canopy shading.

(v) **Plant density, habit and pod production pattern:** In its natural habitat the cocoa plant is a small tree which when planted as a seedling reaches full maturity after about ten years at which time it attains an average height of 5-6 metres. Some cocoa plantations are clonal but usually the crop is grown from seeds, which are more or less hybrids, thus introducing genetic variability. The traditional method of establishment is direct sowing of 2 or 3 seeds per hole. Plant densities are very variable but are generally lower in America and Oceania (500-1100 trees per hectare) than in Africa (1000-2500 trees per hectare). In many of the old plantations the spacings are very irregular and close spacing encourages the plants to grow exceedingly tall. This makes harvesting a nerve racking and tedious task as unusually long cumbersome poles are needed to reach pods in top branches of the trees.

Cocoa is an intermittently growing evergreen which exhibits periods of leaf flushes alternated with periods of vegetative rest. The branches are dimorphic but this feature is often obscured by pruning. Flowers and consequently pods are developed on the main trunk right from the soil level up to and including the branches of the canopy. Flowering occurs in discontinuous flushes; adjacent trees and indeed cushions being out of step. Although flowering periodicity in mature plants follows "seasonal" patterns there is hardly any time of the year when a plantation is devoid of flowers. Thus, like flowering pod setting, occurs throughout the year. There is consequently an overlap, in time, of pods of different ages within a plantation and on each tree. To develop to full maturity, when it is harvested as "healthy ripe", each pod remains on the trees for about 170 days after pollination. At any point in time, a typical cocoa plantation is thus made up of a collection of trees with variable numbers of pods on each tree. The pods being of different ages and scattered all over an enormous surface area made up of the tree trunk and branches. Such scattering of target products poses great problems in harvesting and disease control. Similarly, most features of the typical cocoa farm especially in Nigeria are bedevilled with attendant production and management problems of which the following readily come to mind.

Since new plantations are, in most cases, established by sowing seeds, genetic varia-

bility is generally high. The net effect is that most plantations contain trees which differ greatly in habit of growth, vegetative behaviour, flushing, flower and fruiting characteristics, potential yield and susceptibility to diseases. Such variabilities pose special problems in disease tolerance assessment, breeding work and in devising disease control measures.

Table 4.1 shows cumulative data on two trees in an F₂ Amazon cocoa plot at Gambari Experimental Station (GES) Ibadan, Nigeria. Although both trees originated from the same parent material W6/718 produced almost twice as many pods which also appeared to be more susceptible to *Phytophthora* pod rot disease than W6/358. Table 4.2 shows that such individual tree differences can occur even in clonal materials.

Table 4.1. Individual tree records from an F₂ Amazon plot at GES⁺

Tree No.	Genotype	Cumulative Pod Production*	% Blackpod
W6/358	T79/468	1950	8.9
W6/718	T79/379	3388	11.9

*Total pods produced within first ten years of planting.

+Extracted from CRIN Annual Report 1969/70 p. 74.

Table 4.2. Individual tree records from selfed ICS 1 clonal materials in a plot at GES

Tree No.	Cumulative Pod Production	% Blackpod
125	567	14.1
186	852	20.7
234	766	25.5
369	703	25.5
405	593	24.3

*Extracted from CRIN Annual Report 1969/70 p. 74.

*Total pods produced within the first eight years of planting.

Table 4.3 shows the influence of yield period on incidence of blackpod disease. In Nigeria, the period November to April is the drier half of the year when development and spread of *Phytophthora* pod rot disease is minimal. On the other hand pods on trees between April and November are at high "pod rot disease-risk." Thus, although T12/5 and T12/11 were raised from the same parent material, disease incidence is considerably lower (about a third) in the latter which yields mainly in the dry season than the former which yields mainly in the wet period. The same is true of T54/22 and T54/9 (see Table 4.3).

At an average height of 5-6 m (Table 4.4), a farmer either has to climb the tree or use harvesting poles to reach pods up the tree canopy. Experience has shown that many up-trees pods especially diseased unripe pods and cherelles are never harvested (Adebayo,

Table 4.3. Effect of yield period on Blackpod incidence⁺

Tree No.	Yield Period	% Blackpod
T12/5	April-June, Sept.-Dec.	17.7
T12/11	Mar.-May, Nov.-Dec.	6.0
T54/9	Mar.-June, Sept.-Dec.	18.5
T54/22	Mar.-May, Nov.-Dec.	8.8

⁺ Extracted from Weststeijn 1967. CRIN Memorandum No. 16 Ibadan. 1967.

Table 4.4. Effect of planting material on age and tree height interactions

Plot No.	Planting material	Age of plantation	Average height
N3/2	Clonal materials	29	5.90
N4/2	Seedlings	16	6.02

1974). Similarly, most pods above 4 m from ground level are never sprayed because chemicals never reach that height when the conventional pneumatic knapsack sprayer is used. Although the fungicide could be jetted to such heights with the aid of extension lances, peasant farmers usually feel reluctant to use them because the operation is slowed down, more laborious and wasteful in terms of chemical. Unharvested diseased cherelles and pods have been implicated in the carry-over of the disease from season to season. Such pods are also ready sources of secondary infection for the spread of the disease (Adebayo, 1974) since contact with wash-down or splash of spore-containing rain drops from a diseased pod often spells the doom for most other pods down the tree trunk. Since pod setting occurs throughout the year there is virtually no time of the year when the pathogen is short of suitable host. Each pod thus remains at risk any time environmental conditions, especially rainfall or relative humidity, are right for initiation and development of the disease. The situation is indeed complicated by the fact that many cushions bear clusters of flowers which give rise to an aggregation of pods. Since proximity favours spread of infection the risk is exceedingly high in such areas. Unlike crops like wheat and rice which bear seeds at the apex of the plant each active cushion of the cocoa plant is a potential pod producing site. This is a complicated situation since there are cushions all over a cocoa plant from soil level to the tip of the smallest branch in the canopy. The net effect is that each tree is often dotted all over with pods of varying ages and number. Although spread of disease is encouraged when pods are produced in clusters, control measures are easier to apply and harvesting less laborious than is the case when pods, on the same tree, are far apart.

The economic part of the pod is the dry bean which constitutes only about 6-8% of the fresh pod by weight. With such preponderance of unwanted weight in each pod the general practice is to leave the pod husks in the plantation. However, pod husks should never be left in the plantation since Wright (1931), Thorold (1955) and Okaisabor (1969) have shown that pod heaps are important in the survival of the pathogen through

the dry season. Adebayo (1974) advocated the use of fungicide to suppress the growth of the pathogen in the pod heaps. This practice however, adds to the cost of production and perhaps reduces even further the meagre profit margin of the farmer.

Soil is known to be a major source of inoculum at the onset of the rainy season (Wharton, 1955, Turner, 1965, Okaisabor 1969). Production of flowers and pods at the base of the tree is a common phenomenon in young cocoa plantations. This encourages initiation and spread of the disease as spores from the soil easily contaminate pods either by way of rain splash or by tent-building insects which move soil and other plant debris up the cocoa trees (Evans, 1971, Okaisabor, 1965, 1971). Such early season infections serve as sources of secondary infections and thus spread of the disease to other pods on the tree.

Application of basic disease control measures

With an estimated global loss of 20-30% per annum (Opeke and Gorenz, 1974) a lot of attention has been directed towards the involvement of suitable control programmes for the *Phytophthora* pod rot disease of cocoa. As is to be expected such efforts have concentrated on tackling factors, mainly agronomic, which encourage initiation and spread of the disease within the plantation.

As explained above, temperature, rainfall and shade play a major role in the initiation, development and spread of *Phytophthora* pod rot disease when in their combined effect high humidity is maintained under the cocoa tree for a considerable period. There is not much one can do about rainfall and temperature in as much as both are natural phenomena, more or less, dependent on location over the earth surface. The only factor that one can manipulate thus appear to be shade. In this regard it has been amply demonstrated that reduction of shade especially with judicious pruning of cocoa trees resulting in lowering of the relative humidity and a rise in under canopy temperature are essential aspects of pod rot disease control (Alvim, 1977). As complementary to this, it has been experimentally demonstrated in several countries that after passing its juvenile stage cocoa production is usually higher when grown with little or no shade than when added (Alvim, 1977). Removal of shade thus appears to serve other useful purposes. Recent findings have indicated, however, that increase in yield following complete shade removal does not usually last very long and is commonly followed by a rapid general decline of the plantation, many plants showing severe defoliation and dieback which is often associated with intensive insect attack, particularly capsids. The current thinking therefore is that some shade will still be necessary for cocoa cultivation. There is thus the need to include additional measures in combating the menace of pod rot disease in such lightly shaded cocoa plantations. Such measures include regular, perhaps daily, harvests and removal of diseased pods from the plantation. This can be a costly exercise during the rainy season when fresh infections develop daily. Removal of pod husks or spraying the same with fungicide is also recommended though the cost of either operation can be heavy.

One of the standard measures for combating plant diseases in economic crops is the use of prophylactic chemicals (fungicides). A number of fungicides, mostly copper based, have thus been used in the control of *Phytophthora* pod rot disease of cocoa. In

Nigeria, the following copper-based fungicides are in common use; Perenox(oxide), Caocobre Sandoz (oxide), Copper Nordox (oxide), Kocide 101 (hydroxide), Procida BBS (sulphate) and Lime Bordeaux Mixture (sulphate). Noncopper based fungicides in use include Brestan (Triphenyl tin sulphate) and Orthodifolatan(N-1, 1, 2, 2 tetrachloroethylthio) cyclohex-4-ene-1, 2, dithiocarboximide). Apart from recent reports from Brazil, where a measure of success has been attained with low volume fungicide application, cocoa farmers carry on high volume fungicide spraying using various types of pneumatic knapsack sprayers. Although such high volume fungicide applications ensure adequate coverage of pods within reach most up-tree pods are never sprayed. In Nigeria, the unprecedented increase in labour costs within the last few years has also made the use of pneumatic knapsack sprayers for fungicide application on expensive farm operation. Experiments are on to evaluate the efficiency and operational cost of high volume fungicide application using motorised knapsack sprayers as a prelude to work on low volume spraying.

Having established the fact that at least a phase of the life cycle of *P. palmivora* occurs in the soil, Okaisabor (1970) tried to control the disease by reducing the inoculum density of the pathogen in the ground with a number of soil fungicides. Regular application of insecticides has also been shown to reduce the incidence of pod rot disease (Evans, 1971, Okaisabor, 1971). To this end local farmers in Nigeria often spray a mixture of insecticide and fungicide. Okaisabor (1965) showed that removal of lower pods reduced disease incidence while Maddison (Personal Communication) advocated suppression of flowers at basal region of the tree or the use of tree base insecticides as means of creating a gap between the propagules in the soil and the pods up the tree.

As pointed out above, fungicidal control is beset with a lot of practical problems and often economically impracticable in areas with heavy rainfall where fungicide deposited on pods is quickly washed away. Resistance is thought to be the obvious answer and discovery or development of clones of *T. cacao* resistant to *P. palmivora* has been one of the primary approaches to control for many years. Such attempt at breeding resistant varieties is foreshadowed by problems of existence of morphotypes of the pathogen. At a workshop organised for cocoa *Phytophthora* pathologists in Harpenden in May 1976 a general regrouping of isolates of *P. palmivora* was carried. Thus four morphotypes (MFI-4) which differ in distribution, morphology and perhaps pathogenicity were recognised. Brasier & Griffin (1979) and Zentmyer *et al.* (1979) have advocated that MF3 and MF4 should be renamed *P. megakarya* and *P. capsici* respectively. With the existence of such pathologically different "morphotypes" "strains" or "species" one is not surprised that some cacao cultivars which exhibit some measure of tolerance to blackpod disease in certain countries succumb to the disease in another. And for obvious reasons exchanges of isolates between researchers in different cocoa producing countries is to be discouraged. Perhaps there is need to establish a general screening centre outside the cocoa growing areas. This is, however, hampered by the fact that there is at present no generally reliable laboratory or glasshouse screening methods for tolerance testing before pod production stage. The alternative is to send out bud wood of known clones to many countries. As pointed out by Imle (1966) however, much care is required to avoid simultaneously spreading exotic pests and diseases. This is more so with *P. palmivora* which is known to

attack cocoa stems and branches, producing cankers, whereby the local strains of the pathogen could easily be transferred even in bud woods.

Suggestions for future developments of research efforts.

The above account has perhaps outlined the extent to which the agronomy and management of cocoa can influence the epidemiology and control of the blackpod disease. Past and present efforts of scientists at curtailing the disease have also been enumerated. Like most biological problems it will be asking for too much to expect a final solution but one can at least hope for an improvement on present levels of research and consequently of disease control. Based on a critical appraisal of the complex situation the following proposals for further developments of the present research efforts are worth considering for the future:

(i) **Planting material:** When planted from beans, a cocoa tree attains a height of over 6 metres in less than 15 years (Table 4.4). On the other hand the average height of trees in a 29-year old plantation raised from rooted cuttings is less than 6 metres. Since it has been established that farm operations such as harvesting and spraying are hampered by height of trees, raising new plantations from rooted cuttings will ensure that most trees are within manageable heights. Establishing a plantation with rooted cuttings is understandably more expensive than by direct seeding but with more research it is envisaged that such costs could be reduced.

(ii) **Shade management:** It has been established that the net effect of excessive shading in a cocoa plantation is maintenance, beneath the tree canopy, of high humidity which encourages the incidence and spread of pod rot disease. However, complete removal of shade encourages pest attack which may result in heavy defoliation and destruction of whole plantation. Detailed studies of shade management which may entail planting at wider spacing and use of herbicides to control weed problems arising therefrom should form an integral part of new programmes for blackpod control.

(iii) **Screening of cultivars:** Absence of a quick and reliable method for assessing the tolerance level of cocoa cultivars to *P. palmivora* hampers breeding work. There is thus an urgent need to develop improved methods for screening seedlings in a way that results of laboratory or glasshouse trials can give a better correlation with field results. This will quicken breeding work. In addition, scientists outside the cocoa growing areas will be able to assemble and assess the reaction of different cocoa cultivars to each of a collection of different strains of the pathogen.

(iv) **Fungicide application:** The obstructive canopy formation of a typical cocoa plantation does not permit fungicide application to pods in the canopy from measurable distances from the tree base. This reduces the maximum height attainable to only a few metres hence fungicides hardly get to most up-tree pods. Research into improved methods of fungicide application which could include field trials with motorised knapsack and tractor mounted motorised sprayers will yield fruitful results. Apart from the need to save money on operational costs Gorenz's (1974) finding that as much as 90%

of the fungicide could be wasted when a pneumatic knapsack sprayer is used is further evidence that there is need for a change of style, equipment or both.

(v) **Fungicide formulation:** The increasing cost of copper which is putting the use of fungicides almost out of the reach of an average farmer is now considered to be a major constraint in Nigeria. Many farmers only manage to apply fungicides thrice in a year as against 6-10 applications per year recommended for effective control of the disease. When such inadequate protection does not give the desired results in one year such farmers do not bother to apply any fungicide in subsequent years. In an apparent bid to alleviate this problem the Nigerian Government started a fungicide subsidy scheme but experience has shown that the subsidy is hardly enjoyed by the farmers for whom it is meant. While efforts are on to rectify this anomaly there is a clear need for less expensive fungicides which will, per force, not be copperbased since it is not likely that there can be an appreciable downward trend in world price of copper. It will be necessary for pathologists and chemical companies to work hand in hand to attain this objective.

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DISCUSSIONS

K. K. N. Nambiar: Is there any correlation between the intensity of shade and incidence of the disease?

ANSWER: Shade plants provide for maintenance of high humidity which in turn facilitates disease incidence. Light shades and heavy shades have correlation with the rate of incidence.

Y. R. Sarma: How many sprayings are required for successful control?

ANSWER: 8-14 applications in a year.

Koti Reddy: Will not light shade help disease control?

ANSWER: Reduction of shade helps fall in humidity and hence less incidence of disease.

SCREENING CACAO CULTIVARS FOR BLACK-POD RESISTANCE

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ABSTRACT

Seven cacao clones, three Amazon-Trinitario hybrids and West African Amelonado (Standard) were screened for blackpod resistance. The pods were inoculated with infected pod-discs. The rates of spread of lesion after inoculation were found to be significantly different in various cultivars. They also showed significant differences in the number of days taken to reach 25%, 50%, 75% and 100% infection, once the pod is inoculated.

Among the 11 cultivars tested, clone W5/47 (an introduction from Nigeria) seems to be promising in view of the slower rate of spread of infection.

INTRODUCTION

The genus *Phytophthora* de Bary, according to the recent taxonomic treatment, contains 38 species, which affect a wide range of annual and perennial plants, shrubs and herbaceous plants from many families of flowering plants and conifers. The 'blackpod' disease of cacao is caused by *P. palmivora* (Butl) Butl. The disease occurs in all the major cacao growing areas of the world. Crop losses due to black-pod disease (*Phytophthora* pod-rot) in the various cacao-producing countries varies from almost nil up to 80%. In Papua and New Guinea it was 1.2% (Hicks, 1967), while in the Cameroons the average loss was placed as high as 50% (Muller, 1971). The world average crop loss is estimated (Gregory, 1969) in the range of 20 to 30 per cent. This fungus is also frequently associated with other serious diseases of cacao, ranging from canker to wilt and die-back affecting branches or whole trees (Thorold, 1967). Although blackpod was first distinguished in Guyana and the West Indies, stem canker caused by *Phytophthora* in cacao was first recognised in Sri Lanka (Firman, 1974). Carruthers (1898a, 1898b, 1898c), working in Sri Lanka, was able to connect canker with black-pod.

Spraying experiments in Nigeria on the control of *Phytophthora* pod-rot with various chemicals have not been successful in lowering the amount of losses caused by the disease to acceptable levels (Hislop and Park, 1962; Weststeijn, 1968). The continuation of profitable growing of cacao in the humid tropical regions depends, to a large extent, on the level of control of black-pod disease. Although chemical control of black-pod disease is being practised in many countries, the most efficient way to control this disease is to

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incorporate some degree of resistance in the cultivars. Thus acceptable levels of control can be achieved through chemical sprays along with crop sanitation when the cultivars possess some degree of resistance (Jacob *et al.*, 1973).

In their efforts to develop cacao varieties resistant to *P. palmivora*, plant breeders and pathologists have developed various methods to assess the resistance or susceptibility of cultivars. In screening cacao cultivars for resistance to *P. palmivora*, various types of inoculum such as standard-sized fragments of necrosed tissues cut out from the edge of a diseased area of a pod that had been previously infected, standardised discs of mycelium and zoospores have been used. Various methods of testing used in the resistance studies include infecting attached and detached pods, wounded and unwounded pods, leaves, stem and roots (Blaha, 1974). Weststeijn (1969) reported that pod inoculation was generally preferred in most of the countries.

MATERIALS AND METHODS

Eleven cultivars of cacao, five upper Amazon clones (W5/47, W6/374, W5/566, W6/438 and W5/385), two local selections (B40 and B2) and three Trinitario-Amazon hybrids (ICS1 × NA32, NA32 × ICS95, SCA6 × ICS6) and West African Amelonado (standard) were used in this study. The upper Amazon (Toxopeus, 1964) clones were introduced into Sri Lanka in 1975 from Nigeria under the UNDP/FAO Project on Minor Export Crops (Jacob, 1977).

Four healthy full-sized but immature (about 18 weeks old) pods from each cultivar were selected. The pods were harvested and their length and diameter were recorded. From the middle portion at the side of each pod a uniform-sized disc of tissues was taken out using a cork-borer of 0.5 cm diameter. With the same cork-borer, a disc of tissues (husk) from diseased pod was taken out and inserted into the cavity created by the excavation of tissues on the pod for screening. The inoculum was taken out from the actively advancing peripheral zone of the infected area of pods kept in the laboratory for producing inoculum. The inoculated pod was kept hanging inside a wire-cage of 12 × 12 × 22 cm size, by using a pin on the pod-stalk and tying with a string into the frame of the cage. Then the cage containing the inoculated pod is enclosed by a polythene bag containing about 10 ml of water to provide high humidity inside.

As the lesions were more or less circular, the shape was determined by measuring the infected area along the length and breadth of the pod and the average taken. Rate of spread of lesion and the time taken to reach 25%, 50%, 75% and 100% infection in the inoculated pods were recorded based on visual observation.

RESULTS

Among the 11 cultivars, ICS1 × NA 32 scored the highest mean values for pod length (17.55 cm), pod diameter (8.03 cm) and pod length × pod diameter index (140.93) as shown in Table 4.5. The lowest values for these parameters were obtained in the Amazon clone W6/438. The cultivars showed significant differences in these parameters.

Table 4.5. Mean pod length (L) and pod diameter (B) and L × B index in the 11 cultivars

Cultivars	Type	L(cm)	B(cm)	L × B
W5/47	Amazon Clone	10.83	6.25	67.62
W6/374	Amazon Clone	11.53	6.90	79.58
W5/566	Amazon Clone	10.98	6.25	69.66
W6/438	Amazon Clone	8.08	6.13	49.50
1CS1 × NA32	Trinitario			
	Amazon hybrid	17.55	8.03	140.93
B40	Clone local			
	selection	13.33	6.40	80.57
B2	Clone local			
	selection	16.73	7.05	118.22
W5/385	Amazon clone	14.40	6.55	94.33
NA32 × 1CS95	Trinitario			
	Amazon hybrid	13.93	6.65	92.85
SCA6 × 1CS6	Trinitario			
	Amazon hybrid	15.75	6.33	100.03
Amelonado	West African	12.53	6.78	86.23
	Mean	13.24	6.67	89.05
	Variance due to cultivars	31.974**	1.160**	510.095*
	Variance due to pods	1.443 ^{NS}	0.197 ^{NS}	236.550 ^{NS}
	Error variance	1.574	0.198	185.225

Table 4.6. The mean spread of lesion up to 4 days after inoculation (cm.)

Cultivars	1 day	2 days	3 days	4 days
W5/47	0.05	2.28	4.78	7.45
W6/374	0.63	2.45	5.05	6.48
W5/566	0.60	3.43	5.33	7.25
W6/438	0.60	3.23	5.90	8.18
1CS1 × NA32	0.43	3.28	6.28	8.73
B40	0.68	2.03	4.25	7.60
B2	0.10	2.50	5.03	6.85
W5/385	0.13	3.45	6.78	9.45
NA32 × 1CS95	0.45	3.45	6.25	8.78
SCA6 × 1CS6	0.28	1.88	4.95	7.90
Amelonado	0.58	2.60	4.75	6.80
	Mean	0.45	2.78	5.40
	Variance due to cultivars	0.218**	1.449**	2.494**
	Variance due to pods	0.04 ^{NS}	0.4000 ^{NS}	0.053 ^{NS}
	Error variance	0.051	0.338	0.493

The data on the spread of lesion up to 4 days after inoculation are given in Table 4.6. The cultivars showed significant differences in the spread of lesion on the inoculated pods. Upper Amazon clone W5/47 had the lowest spread of lesion expressed as diameter (0.05 cm), one day after inoculation. The highest value was observed in the locally selected clone B40 (0.68 cm).

Two days after inoculation, hybrid SCA6 × 1CS6 recorded the lowest mean values for the spread of the lesion, while local clones B40 which scored the highest mean spread one day after inoculation scored the lowest mean value for the spread of the lesion 3 days after inoculation. Four days after inoculation, Amazon clones W6/374 and W5/385 scored the lowest (6.48 cm) and the highest (9.45 cm) spread of lesion respectively. The latter scored the highest value for mean spread of lesion 3 days after inoculation as well.

Number of days taken for the spread of the black-pod lesion to reach 25%, 50%, 75%, and 100% of the surface area of the pod in the cultivars are presented in Table 4.7.

Table 4.7. Number of days taken for the spread of lesion to reach 25%, 50%, 75% and 100% pod-area after inoculation.

Cultivars	Number of days taken for the spread of infection to reach			
	25%	50%	75%	100%
W5/47	4.25	5.25	6.25	8.00
W6/374	5.00	6.50	8.00	8.75
W5/566	3.50	5.50	7.25	9.00
W6/438	3.25	4.75	6.00	7.00
1CS1 × NA32	4.25	6.25	8.25	10.50
B40	4.00	5.00	6.00	7.00
B2	5.00	6.50	7.25	10.00
W5/385	4.00	5.00	6.00	7.75
NA32 × 1CS95	4.00	5.00	7.00	8.50
SCA6 × 1CS6	4.50	5.75	6.75	8.25
Amelonado	5.25	6.50	7.75	9.00
Means	4.27	5.64	6.95	8.52
Variance due to cultivars	0.085**	0.076**	0.092**	0.132**
Variance due to pods	0.016 ^{NS}	0.019 ^{NS}	0.023 ^{NS}	0.017 ^{NS}
Error variance	0.017	0.015	0.020	0.018

(data analysed after square root transformation.)

Amazon clone W6/438, was the first to reach 25%, 50%, 75, and 100% infection levels. Two more cultivars, B40 and W5/385, reached 75% level of infection in 6 days along with clone W6/438. The clone B40 also reached 100% level of infection in 7 days along with the clone W6/438. 1CS1 × NA32 took a mean of 10.5 days to reach 100% infection. The same hybrid took the longest time (8.25 days) to reach 75% level of infection. It is

interesting that the West African Amelonado took the longest time to reach 25% (5.25 days) and 50% (6.50 days) infection levels. Two other clones B2 and W6/374 also equalled West African Amelonado at 50% infection level.

DISCUSSION

There is general agreement that the susceptibility of detached pods is greater than that of the attached pods (Medeiros and Rocha, 1964; Rocha and Mariano, 1969; Blaha, 1967; Tarjot, 1969). However, the degree of resistance of detached pods, under laboratory conditions, could be used as an indication of the far higher resistance to be expected from the fruit on the trees (Lellis and Peixoto, 1960). One difficulty in using pods on the tree for screening is in the interpretation of the results when a number of variable environmental factors, which cannot be adequately controlled, are involved.

In general, the percentage success in experimental infection, the number of lesions and the spread of the infected spot on the entire pod are the criteria used for the classification of cacao varieties as resistant or susceptible. Tarjot (1967a, 1967b, 1969) developed a 'susceptibility index' which took into account simultaneously the percentage of successful infection and the time taken for the commencement of the spot (spread of lesion). Blaha (1971a, 1971b) used, as a criterion, the percentage of successful infections and the progress in the daily average spread of the disease by noting the increase in size of the spot in mm² per 24 hours.

Although, clone W5/47 had slower rate of spread for the lesion, the time taken to reach 25%, 50%, 75% and 100% infection was comparatively lower. The smallness of pod size in this clone partly explains why this cultivar which had low rate of spread for the lesion reached the above levels of infection at comparatively shorter durations. In screening studies, more reliable criterion is to assess the rate of spread of lesion. Time taken for the infection to reach various percentages of the surface area of the inoculated pod must be interpreted with caution.

Clone W5/47 is one of the Amazon clones selected in Nigeria for high yield (110 pods per year) and 'field resistance' to black pod disease (less than 5% pod loss per year) based on studies for a period of 10 years. This clone is the same as T85/876 introduced from Ghana into Nigeria. However, the assessment of resistance under field condition is exposed to criticism. Field resistance should be taken only as an approximate indication. Thorold (1953) had already emphasised the difficulty of classifying varieties according to their resistance in the field.

In this study, variability in the rate of spread of lesion exhibited by the same cultivar at 1, 2, 3 and 4 days after inoculation is an aspect which demands further detailed investigation.

In assessing the resistance/susceptibility status of a cacao cultivar, it is desirable to employ two or more methods of screening in addition to field resistance. Thus a more comprehensive assessment of the cultivars can be achieved.

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DISCUSSIONS

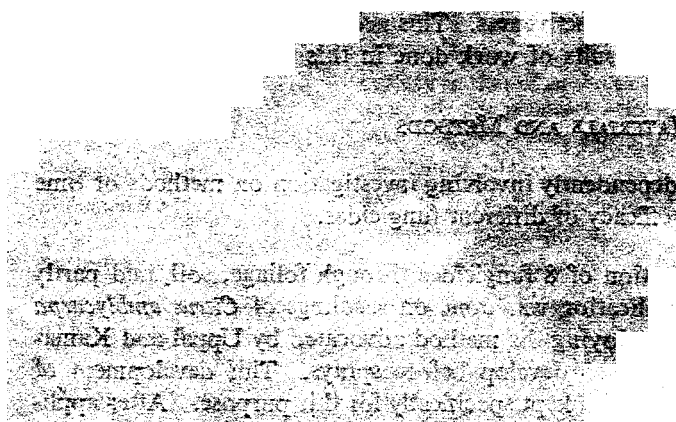
H. S. Sohi : You have been wounding the pods before inoculation. Was this necessary?

Chairman : *Phytophthora* is not a wound parasite and so needs no wound for entry.

A. A. Adebayo : Inoculation should be made without making wounds.

H. S. Sohi : The pathogen was introduced into the pods through cork borer wounds. This might induce susceptibility in all the test pods. Would it not adversely affect correct assessment ?

T. S. Sreenivasan : This might help in comparing the reaction in the test pods and so might be (correctly) practised.



CHEMICAL CONTROL OF PHYTOPHTHORA DISEASE OF CITRUS

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ABSTRACT

Invasion of *Citrus sinensis* by *Phytophthora* spp., causing root-rot and gummosis, has been considered to be one of the causes for its decline. Absence of effective and timely control measures can lead the plant to a state of decadence and unproductivity. This necessitated the present investigations.

The studies under controlled conditions did not display any variation between the methods of fungicide application. The efficacy of the tested fungicides showed considerable variation. The subsequent trials repeated under controlled and field conditions revealed superiority of Bavistin 0.1%, Dithane Z-78 0.2%, and Difolatan 0.3% in controlling incidence of root-rot and gummosis. Aureofungin was equally effective. Dithane M-45, Blitox and Bordeaux mixture were less effective.

INTRODUCTION

Phytophthora diseases are of common occurrence throughout the citrus growing countries of the world. *Phytophthora palmivora*, *P. parasitica* and *P. citrophthora* are some of the species associated with citrus decline through either girdling, leaf-fall, root-rot or foot-rot. *P. palmivora* seems to be more commonly associated with gummosis disease (Rama-krishnan, 1954 and Govinda Rao, 1954). Ravages of *Phytophthora* in citrus have been described in detail by Uppal and Kamat (1936). Mali and Choudhari (1976) considered *Phytophthora* diseases to be one of the important factors in causing decline of sweet oranges and further stated that absence of effective and timely control measures could lead the plant to a state of decadence and unproductiveness. This necessitated the present investigations and the paper presents the results of work done in this regard.

MATERIALS AND METHODS

Three trials were laid out independently involving investigation on methods or time of application of fungicides, and efficacy of different fungicides.

First trial consisted of application of 8 fungicides through foliage, soil, and partly through foliage and soil. The application was done on seedlings of *Citrus amblycarpa* pre-inoculated with *P. palmivora* employing the method advocated by Uppal and Kamat (1936). The symptoms were allowed to develop before sprays. This development of symptoms was tested from one of the lots kept specifically for this purpose. After application of fungicides, the observations in respect of collar-rot, root expansion, damage to roots etc. were recorded at periodic intervals. For this purpose, the seedlings from

individual treatments and replications were uprooted taking care to avoid any damage to root system. The seedlings were then washed thoroughly applying water sprays and percentage of damaged roots worked out.

Second trial was aimed at finding out the proper time for fungicidal application. For this trial, ten-year-old mosambi plants severely infected with *Phytophthora* gummosis were selected. Gum lesions on all these trees were counted and area of the lesions measured and marked before fungicidal application. Sprayings were started during January in one set and during June in second set. In each set three sprayings were given at fortnightly interval and reduction or increase in gum lesions measured.

Third trial was laid out to standardize a spraying schedule for rainy season and to find out the most effective fungicide for control of gummosis. This trial accommodated six fungicides. For this trial, 11-year-old mosambi plants with severe gum lesions were selected and the gum lesions measured and marked before first spray of fungicides. The spray was repeated thrice at fortnightly interval starting from 16 June and the reduction or increase in gum lesions was measured.

RESULTS AND DISCUSSION

The results in respect of method of application and different fungicides applied to plants preinoculated with *Phytophthora* are given in Table 4.8. Method of application of fungicides did not show any significant differences. There results are in agreement with

Table 4.8. Influence of application of fungicides against *Phytophthora* by various methods.

Name of the fungicides with concentrations	Active ingredient	% of damaged roots			Mean	
		F	F+S	S		
Benlate 1000 ppm	Methyl, 1-(butyl carbomoyl) -Z- benzimidazol.	2.41	9.03	5.00	5.48	
Aureofungin 50 ppm	heptane antifungal antibiotic produced by <i>Streptomyces cinnamonens</i> Var. <i>terricola</i> Thir.	8.27	12.89	6.08	9.08	
Bordeaux mixture 1%	Copper fungicide.	17.98	18.42	10.75	15.72	
Difolatan 0.3%	80% N (1, 2, 2, tetrachloroethyl thiotetra-hydrothalimide).	13.08	26.31	21.79	20.39	
Blitox 0.4%	Copper Oxychloride.	23.69	21.98	20.60	22.09	
Copper oxychloride 0.4%	Copper Oxychloride.	23.28	18.54	46.41	29.41	
Dithane Z-78 0.2%	Zinc ethylene bisdithiocarbamate.	27.49	30.84	30.68	29.67	
Dithane M-45 0.2%	Manganese ethylene bisdithiocarbamate.	43.68	21.63	31.61	32.31	
Control (Water Spray)	—	38.55	36.58	29.49	34.87	
F - Foliar, S - Soil,		S.E.	NS	NS	NS	± 7.02
		C.D.				20.85

the reports of Knauss (1974). The fungicides, however, differed from each other significantly. Benlate 1000 ppm proved most effective and was on par with Aureofungin 50 ppm and Bordeaux mixture 1%. However, Bordeaux mixture did not differ statistically from other chemicals and control indicating thereby that Benlate and Aureofungin were the only chemicals effective against *Phytophthora* diseases in citrus.

The results regarding disease development and efficacy of fungicides in controlling gummosis are presented in Table 4.9. It can be seen from the data in respect of control that the disease develops very slowly during January, while at a very fast rate during rainy months. The minimum and maximum temperatures observed at this research station during January to March were around 16°C and 25°C. This range was 22°C and 28.5°C during June onwards to August. The observation on the spread of disease is completely in agreement with the reports of Uppal and Kamat (1936) who observed that the gummosis pathogen required an optimum temperature of 27°C to 32.5°C. The results though point out to the necessity of sprays during rainy months, suggest that initial reduction in gum lesions, resorting to the fungicidal sprays in January may help in reduction of the initial lesions during rainy months. This may increase the efficacy of the fungicides. The comparison of the fungicides points out the superiority of Bavistin followed by Aureofungin, Dithane Z-78 and Difolatan. This was irrespective of the seasons when the fungicides were tested.

Table 4.9. Influence of spraying fungicides during January and June against *Phytophthora*.

Name of fungicides with concentrations	% reduction in gum-lesions at fortnightly intervals during					
	January spray			June spray		
	1st	2nd	3rd	1st	2nd	3rd
Aureofungin 50 ppm.	65.5	92.3	99.8	77.4	96.6	99.8
Dithane Z-78 0.2%	47.4	91.3	97.4	72.9	88.1	96.9
Bavistin 0.1%	60.0	87.7	100.0	81.5	98.8	100.0
Bordeaux Mixture 1%	31.2	61.0	77.1	19.0	29.8	52.3
Difolatan 0.3%	49.3	71.8	91.5	66.1	96.2	97.5
Control (Water Spray)	+3.8	+19.0	+19.0	+50.0	+100.0	+200.3
S.E.	1.29	1.70	1.27	1.64	1.80	32.38
C.D.	3.89	5.11	3.84	4.93	5.43	97.58

Table 4.10 indicates the reduction in gum lesions brought about by some of the fungicides sprayed in the month of June. Considering the entire gum lesions to be 100, percentage reduction at each interval has been worked out. It can be seen that in the absence of the control measures the area of gum lesions is increased by 144 per cent in two months. The best possible reduction was available with Bavistin closely followed by Dithane Z-78 and Difolatan, Bordeaux mixture, which was observed less effective, was statistically on

Table 4.10. Mean percentage reduction in gum lesions as influenced by some fungicides sprayed in June

Name of fungicides with concentrations	Mean percentage reduction after		
	1st spray	2nd spray	3rd spray
Bavistin 0.1%	54.8	21.0	7.2
Blitox 0.3%	76.2	46.2	25.2
Difolaton 0.3%	53.0	22.6	11.4
Bordeaux Mixture 1.0%	82.7	47.8	21.0
Dithane M-45 0.2%	77.9	65.6	40.5
Dithane Z-78 0.2%	60.9	20.0	10.5
Control (Water Spray)	111.2	128.2	143.6
S.E.	4.5	5.1	4.9
C.D.	13.4	15.0	14.6

par with Blitox. Dithane M-45 did not show much efficacy. These results are in agreement with the results of Suit and Cohen (1958) and Desai *et al.* (1966).

All these three trials point out to the damages caused by *Phytophthora* in the absence of very specific control measures. Any of the benomyl compounds, Difolatan or Aureofungin can be considered effective against *Phytophthora* diseases. Dithane Z-78, though have given good results under field conditions, did not muster much efficacy in experiments laid out under controlled conditions.

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DISCUSSIONS

P. H. Tsao : The term "brown rot" is not correctly used. It should be used for "fruit rot of citrus" and should not be used to designate the lesion on the cambium and wood of the trunk.

If Benomyl (Benlate) controls the disease, the causal organism involved is unlikely to be *Phytophthora*. The etiology needs to be examined further.

H. S. Sohi : Description of symptoms indicates the association of organisms other than *Phytophthora*. Also, Bavistin etc. are hardly known to control *Phytophthora*.

Abicheeran : Deep lesions help protection of the pathogen from the reach of contact fungicides. Hence the report on control needs reexamination.

D. N. Srivastava : As tristeza virus is known to be distributed widely among the plants its presence may also contribute to the decline.

P. H. Tsao : Benomyl etc. induces early growth of *Phytophthora* as they are inhibitory to contaminating fungi whose absence benefits *Phytophthora*.

F. J. Newhook : When there are well established stem cankers in which *Phytophthora* is active, I understand that Terrazole 1% a. i. in Xylene would effectively control the disease.

EVALUATION OF GERMPLASM AND CHEMICAL CONTROL OF *PHYTOPHTHORA PARASITICA* VAR. *NICOTIANAE* ON TOBACCO

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ABSTRACT

Phytophthora parasitica var. *nicotianae* is one of the important pathogens on tobacco causing leaf-blight and black-shank. It causes severe damage in commercial nurseries in Andhra Pradesh and Karnataka States. Transplanted tobacco of F. C. V., Natu, Bidi, Chewing and Cigar types are susceptible to this pathogen. Both indigenous and exotic collections of *N. tabacum* and *N. rustica* as well as wild species were evaluated with a view to identifying resistant donors for this important pathogen. Out of 496 collections of *N. tabacum* and *N. rustica* screened by artificial inoculation of *Phytophthora*, only a cigar variety Beinhart 1000-1 was found resistant while McNair-12 was tolerant to this pathogen. Among the 33 *Nicotiana* species, four species viz; 1) *N. longiflora*, 2) *N. nesophila*, 3) *N. plumbaginifolia* and 4) *N. suaveolens* were found resistant and the rest were susceptible. Work has been initiated to develop black-shank resistant varieties utilising the resistant donors Beinhart 1000-1 and *N. plumbaginifolia*.

For chemical control of leaf-blight disease in nurseries nine fungicides were evaluated. Among them, Ridomil @ 0.1%, 0.2%, and 0.3% concentrations were the best in giving 100 per cent control of the disease. Next best fungicides in order of performance were Difolatan 0.2%, Fytolan 0.2% and Bordeaux Mixture 0.4%.

In vitro tests indicated that Ridomil @ 0.1%, 0.2%, 0.3% and 0.4% gave 100 per cent inhibition on growth and sporulation of *Phytophthora parasitica* var. *nicotianae*.

Pot and microplot trials have shown that two applications of Ridomil @ 0.15%, 0.2% and 0.3% at monthly interval gave total protection against leaf-blight and black-shank diseases in nursery.

INTRODUCTION

In India tobacco is grown in about 4,11,000 ha and earns foreign exchange to the tune of over Rs. 110 crores a year. Tobacco crop is susceptible to several diseases both in the nursery as well as in the main field. *Phytophthora parasitica* var. *nicotianae* is one of the important soil borne pathogens on tobacco causing leaf-blight in nurseries and black-shank both in nurseries and on transplanted tobacco (FIG. S. 4.1 & 4.2). All the varieties viz; F. C. V., Natu, Bidi, Chewing and Cigar types are susceptible to this pathogen. In United States estimates of losses due to the dreadful disease have been made and in North Carolina, black shank causes an estimated annual loss of approximately 7 million dollars. In India so far such estimates on damage caused by this pathogen has not yet been made, however 5% annual loss is observed in severely diseased tracts of Andhra Pradesh and

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Karnataka states. Sometimes the cultivators have to abandon the entire crop especially in some pockets of these two states where the pathogen persists in an endemic form.

This paper presents results of studies taken up at this Institute on:

- a) Screening germ plasm and *Nicotiana* species to black shank disease caused by *Phytophthora parasitica* var. *nicotianae* with a view to identifying resistant cultivars and wild *Nicotiana* species.
- b) Chemical control of leaf-blight in nurseries caused by *Phytophthora parasitica* var. *nicotianae* using systemic and non-systemic fungicides.

Studies on Ridomil, a systemic fungicide, for the control of plant diseases were extensively made in other parts of the world for the control of *Peronospora tabacina* causing blue mould on tobacco by Schattauer and Schipfer (1978), Johnson *et al.*, (1979) and Worden (1978) and for the control of *P. parasitica* var. *nicotianae* in tobacco by Vasilakakis *et al.* (1979) and *P. infestans* on tomato plants by Cohen *et al.* (1979). Edgington *et al.*, (1980) observed that this fungicide was an outstanding one against *Pythium* and *Phytophthora*.

In India, Ridomil 25 w. p. @ 0.4% and 0.8% concentrations was found effective for the control of black-shank pathogen on tobacco at Hunsur, Karnataka state (Elias, personal communication).

MATERIALS AND METHODS

Seedlings of 496 collections of *N. tabacum* and *N. rustica* and 33 *Nicotiana* species were raised in sterilised soil. Duplicate pots were kept for each variety/species. Spore and mycelial suspension of 6 days old virulent culture of *Phytophthora* grown on oats meal agar was prepared in distilled water by macerating in a Waring blender and inoculated at the base of the plants uniformly. Inoculated plants were kept in humid chambers providing 90-95% humidity for the development of infection. Periodical observation on leaf blight and stem infection were taken. External stem lesion length was taken into consideration and reaction was evaluated as follows: 0 to 5 mm Resistant; above 5 mm Susceptible.

Laboratory trial : In the laboratory, Ridomil 25 w.p. (Methyl D, L-N- (2, 6-dimethyl-phenyl)-N-(2' methoxyacetylalaninate) was mixed with oats agar in 9 cm petridishes to obtain 0.1, 0.2, 0.3 and 0.4% concentrations. Three replications were maintained. The petridishes were inoculated with uniform inoculum discs (5 mm). The dishes were then incubated at the room-temperature (26° — 30°C). Extent of growth was recorded, sporangial and oospore production was examined.

Microplot trial : Nine fungicides viz; 1) Bafin, 2) Bavistin 3) F. M. Spray, 4) F.M. Spray F. 55, 5) Difolatan, 6) Bordeaux mixture, 7) Fytolan, 8) Neem Kernel suspension and 9) Ridomil 25 w.p. were screened in 0.25 sq. meter microplots in a randomised block design with four replications.

Fungicides were sprayed uniformly on 8-week old seedlings. Twenty four hours after the application of fungicides the beds were uniformly inoculated with a virulent culture of *Phytophthora*. Moist hessian cloth tents were erected to provide shade and humidity for the development of leaf-blight. Symptoms of blight appeared within a period of 4-6 days. Four days later incidence of leaf blight, stand of transplantable seedlings and healthy seedlings were recorded in 0.25 sq. meter beds. Ridomil 25 w.p. was applied thrice, first as a pre-sowing soil drench and the second and third as sprays at an interval of 25 days while the other fungicides were sprayed 6 times at 3 days' intervals besides the soil drench.

RESULTS AND DISCUSSION

Among the 496 collections of *N. tabacum* (Flue-cured, Air-cured, Burley, Bidi, Cigar, Cheroot, Wrapper, Chewing, Natu, Turkish and Fire cured) and *N. rustica* (Hookha and Snuff), only a cigar variety Beinhart 1000-1 was found resistant and one Flue-cured type McNair-12 was tolerant to black shank pathogen *P. parasitica* var. *nicotianae* while the remaining types were found susceptible. No symptom of leaf blight and black-shank was observed on Beinhart variety, while in McNair 12, no leaf blight was observed but the stem infection was less than 1 mm length.

The resistance found in Beinhart 1000-1 has been incorporated in flue-cured types and crosses made in this direction were thoroughly tested for further screening to black shank resistance. Backcrosses were also made and black-shank resistant plants were isolated.

Out of the 33 *Nicotiana* species tested, resistance was noticed in four species viz; *N. plumbaginifolia*, *N. nesophila*, *N. longiflora* and *N. suaveolens*. Rest of the species were highly susceptible (Table 4.11).

Foster (1943), reported *N. longiflora* and *N. plumbaginifolia* to be highly resistant. In our studies, similar results have been obtained. However, the resistant reaction of *N. nesophila* and *N. suaveolens* in the present studies were not recorded by any of the above workers and subsequently by Litton *et al.* (1970). Similarly species like *N. repanda*, *N. rustica*, *N. nudicaulis*, *N. paniculata* and *N. undulata* which were found resistant to moderately resistant by the above workers were found susceptible to our isolate of *P. parasitica* var. *nicotianae*. This explains the possibility of existence of a new race of the black shank pathogen apart from races 0 and 1 reported by Stokes and Litton (1965).

Black-shank resistance obtained in *N. plumbaginifolia* has been incorporated in *N. tabacum* and further back crosses were made with Laburely-21 and GSH-3. Black shank resistant plants have been isolated from the breeding material obtained from *N. plumbaginifolia*.

Laboratory evaluation of Ridomil 25 w.p. indicated that the growth and sporulation of *P. parasitica* var. *nicotianae* were totally suppressed by Ridomil @ 0.1, 0.2, 0.3 and 0.4% concentrations. There was no growth of the fungus even after 21 days in the treated petri plates. In untreated control, maximum (9 cm) growth was observed by the fifth day and heavy sporangial development by the seventh day.




Table 4.11. Reaction of *Nicotiana* species to black shank of tobacco.

<i>Nicotiana</i> species	External lesion length in mm	Reaction	<i>Nicotiana</i> species	External lesion length in mm	Reaction
<i>N. alata</i>	20	S	<i>N. occidentalis</i>	25	S
<i>N. amplexicaulis</i>	30	S	<i>N. palmeri</i>	C	S
<i>N. benthamiana</i>	C	S	<i>N. paniculata</i>	"	S
<i>N. debneyi</i>	"	S	<i>N. plumbaginifolia</i>	0	R
<i>N. excelsior</i>	"	S	<i>N. repanda</i>	C	S
<i>N. exigua</i>	20	S	<i>N. rosulata</i>	"	S
<i>N. glauca</i>	C	S	<i>N. rotundifolia</i>	"	S
<i>N. glutinosa</i>	30	S	<i>N. rustica</i>	"	S
<i>N. gosoei</i>	"	S	<i>N. simulens</i>	"	S
<i>N. hesperis</i>	"	S	<i>N. solanifolia</i>	20	S
<i>N. ingulba</i>	"	S	<i>N. sylvestris</i>	C	S
<i>N. langsdorffi</i>	"	S	<i>N. suaveolens</i>	0	R
<i>N. longiflora</i>	0	R	<i>N. tabacum</i>	C	S
<i>N. maritima</i>	C	S	<i>N. trigonophylla</i>	20	S
<i>N. megalosiphon</i>	15	S	<i>N. umbratica</i>	C	S
<i>N. nesophila</i>	0	R	<i>N. undulata</i>	C	S
<i>N. nudicaulis</i>	C	S			

S = Susceptible R = Resistant. C = Whole plant collapsed

Results of the seed bed trial are presented in Tables 4.12 and 4.13. Ridomil 25 w.p. in all the 3 concentrations viz; 0.1, 0.2 and 0.3% gave total control of leaf-blight in nursery beds. Next in order were Bordeaux mixture, Difolatan and Fytolan.

Maximum number of transplantable seedlings were obtained in beds treated with Ridomil (0.1, 0.2 and 0.3%) and Difolatan 0.2%. Bordeaux mixture 2 : 2 : 500, Fytolan 0.2% and Bafin 0.04% also yielded more number of transplantable seedlings. Fungicides Ridomil, Difolatan, Bordeaux mixture and Fytolan were superior in the production of healthy seedlings.

It is concluded that the systemic fungicide Ridomil 25 w.p. is the most effective one for the control of leaf blight of tobacco in seed-beds. Among the non-systemics, 0.4% Bordeaux mixture, 0.2% Difolatan and 0.2% Fytolan are the most promising fungicides. Bavistin and Neem kernel suspension were the least effective treatments.

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Authors are thankful to Dr. N. C. Gopalachari, Director for his keen interest and encouragement. Cooperation and help received from the Division of Genetics and Plant Breeding are gratefully acknowledged.

Table 4.12. Chemical control of leaf-blight on tobacco seed-beds.

Treatment	Mean no. of plants affected with leaf blight in 0.25 M ²		
	1977—78	1978—79	1979—80
Bafin seed treatment + 0.03% spray	21.75	52.75	23.00
Bafin 0.04%	21.50	74.75	21.00
Bafin 0.06%	6.25	75.00	28.25
Bavistin 0.1%	63.75	124.25	18.75
F. M. spray 1.0%	51.00	72.50	11.00
F. M. spray 1.5%	42.50	83.0	11.50
F. M. spray F. 55. 1.5%	33.75	49.0	7.75
Difolatan 0.2%	—	32.25	15.25
Bordeaux mixture (0.4%)	16.75	14.00	6.25
Fytolan 0.2%	17.50	78.75	18.50
Neem kernel suspension spray 1.0%	71.75	111.75	7.25
Ridomil 25 w. p. 0.1%	—	—	0.00
Ridomil 25 w. p. 0.2%	—	—	0.00
Ridomil 25 w. p. 0.3%	—	—	0.00
Control (Inoculated)	83.00	207.5	46.75
S. E. M.	11.05	15.36	0.57
C. D. at 5%	31.91	36.10	1.58
C. D. at 1%	42.92	41.99	2.08

Table 4.13. Chemical control of leaf-blight on tobacco seed beds.

Treatment	Mean no. of transplantable seedlings in 0.25 M ²			Mean no. of total healthy seedlings in 0.25 M ²		
	1977—78	1978—79	1979—80	1977—78	1978—79	1979—80
Bafin seed treatment + 0.03% spray	160.75	35.75	26.25	331.75	63.75	37.00
Bafin 0.04%	129.50	26.25	30.25	309.50	35.50	43.25
Bafin 0.06%	37.75	34.00	26.00	68.25	52.50	39.50
Bavistin 0.1%	104.75	32.25	11.75	195.25	51.00	22.25
F. M. spray 1.0%	154.00	67.75	7.75	189.75	110.25	12.00
F. M. spray 1.5%	87.50	69.75	10.00	119.25	91.50	12.00
F. M. spray F. 55. 1.5%	56.75	70.00	6.25	79.75	105.75	7.50
Difolatan 0.2%	—	75.00	50.50	—	155.75	82.50
Bordeaux mixture (0.4%)	74.50	61.00	47.25	195.25	143.75	72.25
Fytolan 0.2%	111.75	47.00	49.75	209.75	194.25	73.25
Neem kernel suspension spray 1.0%	32.75	18.75	33.50	54.75	36.75	61.25
Ridomil 25 w. p. 0.1%	—	—	76.50	—	—	105.00
Ridomil 25 w. p. 0.2%	—	—	70.25	—	—	112.00
Ridomil 25 w. p. 0.3%	—	—	67.00	—	—	117.75
Control (Inoculated)	15.00	33.50	15.00	37.75	46.50	26.75
S. E. M.	21.02	11.13	0.58	38.84	21.50	0.92
C. D. at 5%	60.69	22.65	1.62	112.15	43.75	2.56
C. D. at 1%	81.75	30.43	2.13	151.05	58.78	3.36

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DISCUSSIONS

Chairman: How long does the resistance last?

ANSWER: For a long time. Hitherto the propagating materials have not demonstrated disease development.

T. N. Sreenivasan: Ridomil at 0.02% concentration was found effective against *Phytophthora* spp. Why is it that you have used higher concentrations for its control.

ANSWER: Lower concentrations also will be tested.

STUDIES ON THE TARO LEAF BLIGHT FUNGUS *PHYTOPHTHORA*
COLOCASIAE IN SOLOMON ISLANDS: CONTROL BY
FUNGICIDES AND SPACING*

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ABSTRACT

With mist blower application, copper oxychloride at 2.25 kg/ha was effective in controlling *Phytophthora colocasiae* but mancozeb at 3.6 kg/ha was not. Phytotoxicity from captafol at 1.8 and 3.6 kg/ha nullified any potential gain in yield from control of blight. Leaf removal from healthy plants to maintain four leaves per plant for 90 days, to simulate roguing of leaves of disease control, was shown to cause no loss in yield. However, regular roguing of diseased leaves over the same period in plots affected by a severe epiphytotic did not eradicate the pathogen. When roguing ceased at 90 days disease increased rapidly to epidemic proportions that seriously affected final corm yield.

An attempt to reduce the effect of blight by wider than traditional spacing was unsuccessful. Under continuous epiphytotic conditions disease had the maximum possible impact regardless of spacing. Whereas plants free from competition normally bear six to seven leaves, this number was reduced by severe disease to three or four, the same number as was borne by plants under the competitive conditions of closer-than-traditional spacing. Instead of traditional 20-30,000 plants/ha, it was shown that density can be doubled under conditions of high leaf blight hazard and increased yields still be obtained.

DISCUSSIONS

Chairman : This may be useful in small home gardens.

D. H. Lapwood : Will not this sort of spacing lead to competition for final space?

ANSWER: Judicious adjustment of foliage in utilising the aerial space by permitting thin and heavy foliage alternatively, would avoid such competition.

*For full text of the paper, please see *Ann. appl. Biol.* 96:(1): 1-10, 1980.

REACTION OF CITRUS ROOTSTOCKS TO PHYTOPHTHORA DISEASES

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Citrus Dieback Disease Scheme, Shirampur

ABSTRACT

Phytophthora diseases are highly damaging and have positive correlation with slow decline of citrus. Fungicidal control though effective becomes costly. Search for the rootstock for resistance to *Phytophthora* diseases, was therefore undertaken.

The studies comprised 155 rootstocks from 28 citrus and related species. Based on apparent symptomatology of the diseases caused by viruses, mycoplasma and fungi it was possible to eliminate 119 types. Remaining 36 cultivars which showed promise from the angle of tolerance to viruses, were inoculated with *Phytophthora*.

The tests and repeat tests carried out under controlled conditions revealed *Poncirus trifoliata* and Cleopatra mandarin Morocco (*C. reshini*) to be resistant to *Phytophthora* diseases. High levels of tolerance were evident in 6 cultivars from *C. jamberi*, two from *C. limonia*, two from *C. reshini* species. Remaining cultivars showed susceptibility, though variation in its levels was evident.

Cultivars from jamberi, limonia, reshini groups and a few citranges only appear to be promising from the angle of *Phytophthora* tolerance. Cultivars from all other species showed susceptibility.

INTRODUCTION

Sweet orange decline has been attributed to complex factors involving viruses, mycoplasma and *Phytophthora* species. Mali and Choudhari (1976) and Fraser (1967) considered combination of viruses and *Phytophthora* disorders to be more deadly than the individual factor. Various workers (Uppal and Kamath 1936; Choudhari, 1951; Govinda Rao, 1954 and Kapoor and Bakshi, 1957) however, reported that *Phytophthora* individually caused various disorders viz. gummosis, brownrot, foot-rot and root-rot, resulting in serious injury to citrus.

Phytophthora disease produces symptoms of decline through either rotting of rootlets, girdling of trunk and dropping of blighted leaves. Control measures either by adopting manipulation in cultural practices or through use of fungicides, though have been indicated, are costlier and are of recurring nature.

Rootstocks resistant to these pathogenic disorders appear to be the only long range and economic proposition for getting an effective control. The existing germplasm available at this research station was, therefore, screened for resistance to all the pathogenic disorders with special emphasis on *Phytophthora*. These studies were initiated in 1974 and continued till 1979.

MATERIALS AND METHODS

The experimental material comprised 155 cultivars from 28 citrus and related species planted in between 1963 to 1966. These were screened for apparent symptoms of the viruses, mycoplasmal and fungal disorders (Chadha *et al.*, 1970). The cultivars which showed susceptibility to more than three disorders were discarded. Following this procedure of elimination, the cultivars which were symptomless under field condition were indexed for viruses following the methods of Childs (1968). The seedlings which were observed promising in the aforesaid preliminary studies, were grown under controlled conditions. After 6 months, root-inoculation with *Phytophthora* was done adopting the method of Uppal and Kamat (1936). For this purpose identified and tested cultures were raised on sand maize media. Measured quantity of culture per unit area of the soil was added. The seedlings were subjected to repeated inoculation after a period of six weeks.

Periodical observations were recorded on the growth of seedlings, collar-rot, root length and number and length of damaged roots. The susceptibility or resistance was graded into 5 categories on the basis of damage to feeder and tap roots:

1. No visible symptoms after repeat-inoculation (R).
2. A few roots observed damaged after repeat-inoculation (HT).
3. Damage to some roots with no ill effects on the growth of the plants (L).
4. Root system damaged up to 50 per cent (WS).
5. Root system either decayed or missing (HS).

RESULTS AND DISCUSSION

One hundred and nineteen cultivars out of the lot of 155 studied showed apparent susceptibility to more than two viral disorders, greening and *Phytophthora* during the 3 years of study.

The details in respect of species and cultivars so rejected are given at Table 4.14. Thirty six cultivars which showed freedom from the apparent symptoms of any of the pathogens were indexed for tristeza, psorosis, exocortis and greening. The indexing helped in eliminating 16 cultivars which showed susceptibility to damaging disorders. The details of these 16 cultivars are given in Table 4.15.

Twenty cultivars which showed tolerance to virus disorders were inoculated with *Phytophthora*. The results in respect of indexing and inoculation have been listed in Table 4.16. The cultivars showed various levels of resistance/tolerance. Cleopatra mandarin Morocco was observed resistant to the viral disorders and had very low incidence of greening. This rootstock even after repeated inoculations showed resistance to *Phytophthora*. Cleopatra mandarin Narana showed almost the same trend in respect of resistance. However, the third strain from the same species viz., Cleopatra

Table 4.14. Species and cultivars of *Citrus* susceptible to *Phytophthora*, greening and viral diseases.

Citrus species	Cultivars rejected because of susceptibility to more than 3 pathogens.
<i>Citrus reticulata</i>	Coorg santra, Kodaikithulli, Orlando tangelo, Dancy tangerine, Kara mandarin, King of Siyam, Coorg orange, Laddu coorg, Emperor mandarin, Scarlet mandarin, Rangatra coorg, Citrus China, Kinnow mandarin, Apple by sour orange, Japanese Str. of orange.
<i>C. sinensis</i>	Satgudi, Mosambi, R-4 Mosambi like, Mediterranean sweet orange, Sweet orange simons, Rubimalta, Blood red malta, Common malta, Homossosan orange, Orange tenerif, Paramatta orange, White sylette orange, St. Mitchel malta, Hamlin A. P., Tangalo malta, Dessert orange, Pineapple Nagpur orange, Smooth sour orange.
<i>C. jamberi</i>	Limao Russado Brazil, Rough lemon Australia, Limao Cravo Brazil, Rough lemon Coorg, Jamberi Bombay, Seville orange, Sohmyndong, Citrus Limao Socilno.
<i>C. rerna</i>	Karnakhatta.
<i>C. medica</i>	Bengal citron, Majiphal sweet, Mahalunj.
<i>C. mederaspatana</i>	Kichilli.
<i>C. grandis</i>	Pummelo red sour, Pummelo red sweet, Pink pummelo, Watson pummelo, Turanj.
<i>C. paradisi</i>	Grape fruit, Grape fruit Dunkan.
<i>C. aurantifolia</i>	Coorg lime, Western key lime, Mexican lime, Kagzi lime.
<i>C. limon</i>	Lemon galgal, L-8 seedless lemon, L-15 Brazilian lemon, Rajamundry lemon, Malta lemon, Pat lemon, Lemon long, L-12 Eureka lemon, Citrus lemon.
<i>C. aurantium</i>	Badvapuli, Gajnimma.
<i>C. madurensis</i>	Calamandin, Calamand in R-I.
<i>C. lijettoides</i>	Sweet lime.
<i>C. nobilis</i>	King mandarin.
<i>C. semipeflorencia</i>	Lime Sadaphal.
<i>C. latipes</i>	Sokhymphorashi.
<i>C. tiwanica</i>	Citrus tiwanica.
<i>Atlantia monophylla</i>	Atlantia.
<i>Ferria limpra</i>	Wood apple.
Other <i>Citrus</i> species	Gabbuchini, Kamquat, Molepuli, Limao Bugossodo Brazil, Westim Brazil, Quinshin, Froskmarsh, Two citrus species, Olive groove sweet lime, Mexicalime, Limao comerage Brazil, Keen sour, Miniola, Citremon, Tangerine ling ming ping, A strain of orange.
<i>Poncirus trifoliata</i> and its hybrids.	Morton citrange, Citrange rusk Morrocco, Troyer citrange, Troyer citrange Punjab, Carrizo citrange, Carrizo citrange Punjab, Troyer citrange Australia, Troyer citrange Morrocco, Citrange A. P., Tangerine Punjab, Carrizo Punjab, Tangerin Pan, Trifoliata trifosta, Troyer citrange Brazil, Troyer citrange A. P., Rubidox, Pomeroy trifoliata, Citrange Kodur, Troyer Punjab, Sevege citrange.

Table 4.15. Indexing of cultivars of *Citrus* for tristeza, psorosis, exocortis and greening

Name of rootstock	Tolerance to viral and mycoplasmal diseases			
	Tristeza	Psorosis	Exocortis	Greening
<i>Citrus limonia</i>				
Rangpur lime Australia	+	+	+	L
Nematanga Assam	+	+	+	M
<i>C. reshini</i>				
Cleopatra mandarin Australia	+	+	—	M
Cleopatra Coorg	+	+	—	M
Tangerine Cleopatra	+	+	—	L
<i>C. jamberi</i>				
Jamberi Local	+	+	—	L
Jamberi Saoner	+	+	—	L
Rough lemon Tharsa	+	+	—	L
Rough lemon South Africa	+	+Q	—	M
Moognimbe	+	+	—	M
<i>C. aurantifolia</i>				
St. Bordon red lime	+	+	+	L
<i>C. grandis</i>				
Rabab tanga Assam	+	+	—	L
<i>C. paradisi</i>				
Grapefruit Marsh	+	+	—	L
<i>C. mederaspatana</i>				
Wadapudi mutant	+	+	—	L
<i>C. monophylla</i>				
Narangi coorg	+	+	—	L
<i>Trifoliata</i> hybrids				
Tangelo	—	+	—	L

INDEX : + = Presence; — = Absence, M = Medium, L = Low.

mandarin Grabstan showed susceptibility to *Phytophthora*. Except L-2 Rangpur lime, other three strains from *C. limonia* group showed high tolerance to *Phytophthora* disorders. Among the 7 strains of *C. jamberi* group under study, Rough lemon 58-III-IV and Rough lemon M. P. displayed resistance. Smooth lemon Assam was observed highly susceptible, while other cultivars displayed some degree of tolerance.

Table 4.16. Performance of promising rootstocks to viral and mycoplasmal diseases, and to repeated inoculations of *Phytophthora palmivora*.

Name of rootstock	Tolerance to viral and mycoplasmal and fungal diseases				<i>Phytophthora</i>
	Tristeza	Psorosis	Exocortis	Greening	
<i>Citrus reshini</i>					
Cleopatra mandarin Morrocco	R	R	R	L	R
Cleopatra mandarin Narana	HT	R	R	L	R
Cleopatra mandarin Grabstan	R	TH	R	L	WS
<i>C. limonia</i>					
Rangpur lime	T	HT	R	L	HT
Marmalade orange—	G	HT	R	L	HT
L-2 Rangpur lime	R	HT	S	R	WS
B-19 Rangpur lime	T	T	T	L	HT
<i>C. jamberi</i>					
Rough lemon M. P.	HT	HT	R	L	R
Rough lemon Chettali	T	HT	R	L	T
Rough lemon 58-III-IV	T	R	R	L	R
Rough lemon limonaria	T	R	R	L	T
Florida rough	T	R	R	R	T
Smooth lemon Assam	T	R	R	L	HS
Jamberi Kodur	T	R	R	L	T
<i>C. amblycarpa</i>					
Nasnaran	HT	R	R	L	HS
<i>C. macroptera</i>					
Satkara	T	T	R	R	HS
<i>C. species</i>					
Severonia	T	T	R	L	WS
<i>C. aurantifolia</i>					
Philippine red lime	S	T	R	R	WS
<i>C. mederaspatana</i>					
Belladikithulli	R	T	R	R	WS
<i>Poincirus trifoliata</i>					
Trifoliolate orange	R	R	R	R	R

INDEX: R = Resistant; HT = Highly tolerant, T = Tolerant, S = Susceptible, WS = Weakly susceptible, HS = Highly susceptible, L = Low.

Cultivars viz. Nasnaran, Satkara, Sevaronia, Philippine red lime and Bell adikithulli, showed susceptibility to *Phytophthora*. Trifoliolate orange was resistant (Table 4.17).

Table 4.17. Performance of promising rootstocks to repeated inoculations of *Phytophthora palmivora*

Name of rootstock	Percentage of plants observed in various grades after repeated inoculations				
	1(R)*	2(HT)	3(T)	4(WS)	5(HS)
<i>Citrus reshini</i>					
Cleopatra mandarin Morrocco	60	40	0	0	0
Cleopatra mandarin Narana	80	20	0	0	0
Cleopatra mandarin Grabstan	0	0	20	40	40
<i>C. limonia</i>					
Rangpur lime	40	40	20	0	0
Marmalade orange	40	40	20	0	0
L-2 Rangpur lime	20	20	0	0	60
L-19 Rangpur lime	60	40	0	0	0
<i>C. jamberi</i>					
Rough lemon M. P.	80	20	0	0	0
Rough lemon Chettali	40	40	20	0	0
Rough lemon 58-III-IV	80	20	0	0	0
Rough lemon limonaria	60	20	20	0	0
Florida rough	0	60	0	0	40
Smooth lemon Assam	0	0	0	20	80
Jamberi Kodur	20	20	40	20	0
<i>C. amblycarpa</i>					
Nasnaran	20	0	0	20	60
<i>C. macroptera</i>					
Satkara	0	0	0	20	80
<i>C. Species</i>					
Severonia	0	20	20	20	40
<i>C. aurantifolia</i>					
Philippine red lime	20	20	0	20	40
<i>C. mederaspatana</i>					
Belladikithulli	40	0	0	40	20
<i>Poncirus trifoliata</i>					
Trifoliolate orange	60	40	0	0	0

* Index as in Table 4.16

The studies revealed that Cleopatra mandarin Morrocco, Cleopatra mandarin Narana and trifoliolate orange possessed all the desirable traits as root stocks. Choudhari and Patil (1976) have, however, postulated unsuitability of trifoliolate orange under alkaline soil conditions. The characters of resistance available in this cultivar can, however, be used in breeding programme. Two cultivars from *C. reshini* thus possess the potential as a rootstock for sweet orange.

The studies thus pointed out the resistance of cleopatra mandarin Morrocco, cleopatra mandarin Narana, Rough lemon M. P., Rough lemon 58-III-IV and trifoliolate orange to *Phytophthora*. All other cultivars from 28 *Citrus* species were considered to be unsuitable for employment as root stock in citrus. The studies also revealed that resistance differed from cultivar to cultivar of the same species and using the vague terms like Jamberi or Rangpur lime in employment of the root stocks would be disastrous for sweet orange industry with concrete basis. The results in respect of the resistance or susceptibility of the cultivars, observed in present investigations, are in general agreement with those reported by Deshmukh(1976).

The entire studies indicate the superiority of some of the cultivars from *C. jamberi*, *C. limonia* and *C. reshini* which possess the potential of a rootstock for sweet oranges under the existing conditions. Some of the strains like trifoliolate orange which have a marker gene, can be used in breeding programme, while the strains like Cleopatra mandarin, Morrocco and Narana, Rangpur lime, Marmalade orange, L-19 Rangpur lime, Rough lemon M. P., Rough lemon 58-III-IV can be used in large scale rootstock trials under varying soil conditions so as to arrive at specific conclusions regarding their suitability or otherwise as root stock for sweet orange as well as mandarins.

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DISCUSSIONS

T. S. N. Reddy : Which is the method used for rootstock inoculation?

ANSWER : Mass culture of *Phytophthora*.

T. S. N. Reddy : How would it be known that the fungus has entered the rootstock?

ANSWER : By observing the symptom development.

D. N. Srivastava : Have you studied the role of scion in carrying over the disease?

ANSWER : This will be looked into.

REACTION OF CITRUS AND RELATED GENERA TO *PHYTOPHTHORA NICOTIANAE* VAR. *PARASITICA**

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ABSTRACT

Young nucellar seedlings of sixty two different varieties and selections of *Citrus* and related genera were tested for resistance to *Phytophthora nicotianae* (B. de Haan) var. *parasitica* (Dast.) Waterh. in the greenhouse by inoculating their root system with suspension of zoospores in the aerated water culture tanks. Biometric data on growth and extent of feeder root and tap root damage were recorded after 120 days of inoculation and used for evaluation. High degree of resistance was found in all the three genera tested namely, *Citrus*, *Poincirus* and *Severinia*. Out of 16 species evaluated in the genus *Citrus*, highly resistant clones were found in *Citrus aurantium*, while the clones in the other species showed varying degree of resistance ranging from highly susceptible to moderately resistant. Most of the trifoliolate clones and its hybrids showed high degree of tolerance. Citrus scions such as Mosambi, Sathgudi, Blood Malta, Kinnow mandarin, Coorg orange, Nagpur orange, acid lime and lemons which are extensively grown in India were highly susceptible. Among the various root stocks tested, certain clones in Rough lemon, Rangpur lime and Cleopatra mandarin showed moderately resistant type of reaction.

INTRODUCTION

Root rot and gummosis of citrus incited by *Phytophthora* spp. continue to be the primary factors for poor growth and death of trees of all ages in many citrus growing areas of the world (Fraser, 1942; and Rossetti, 1969). In India, Kamath (1927) reported gummosis caused by *P. palmivora* Bulter as a serious disease whereas, Chowdhury (1951) stated that, *P. parasitica* Dastur was the cause of gummosis in Assam. Now, *P. nicotianae* var. *parasitica* is considered to be more predominant in Karnataka (Ullasa *et al.*, 1978) and Maharashtra (Khumbhare and Moghe, 1976) which causes root-rot, leaf-fall and fruit rot diseases. Programmes for testing the relative susceptibility of root stocks to *Phytophthora* spp. were in operation in several citrus producing countries of the world (Uppal and Kamath, 1936; Rossetti, 1947; Carpenter and Furr, 1962; Broadbent *et al.*, 1971; Hutchison and Grimm, 1972; Cinar and Tuzcu, 1976). However there is not much information available in India about the resistance of citrus cultivars to this pathogen. Hence the present studies were undertaken to locate sources of resistance, which in turn may help in breeding programme.

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*Part of the Doctoral thesis submitted to the University of Agricultural Sciences, Hebbal, Bangalore.

MATERIALS AND METHODS

Isolation of the pathogen and production of inoculum:

The fungus *P. nicotianae* (B. de. Haan) var. *parasitica* (Dast.) Waterh. was isolated from naturally infected Coorg orange trees from Citrus Experiment Station, Gonicoppal, Karnataka. The method used for isolation of the fungus was that of Grimm and Alexander (1973). Pathogenicity of the fungus was verified by root inoculation of Coorg orange and sweet orange (CV Sathgudi) seedlings. The zoospores for inoculation were obtained as per the method described by Tsao and Garber (1970) and modified by Grimm and Hutchison (1973). The fungus was grown on bean seed broth in 100 ml Erlenmeyer flasks incubated at $25 \pm 1^\circ\text{C}$ for 7 days. The mycelium from incubated cultures was transferred to petridishes flooded with sterilized pond water (1 part pond water + 2 parts tap water). Production of zoospores was accelerated by flooding the mycelium with deionised water which was cooled previously to 15°C . Three isolates of the pathogen from different sources were combined for inoculation.

Production of test seedlings :

Seeds of the various varieties/species of citrus and related genera were collected from Citrus Experiment Station, Gonicoppal, Horticultural Experiment Station, Chethalli, and also obtained from Agricultural University, Ludhiana, India, Australia and California. Seedlings were separately raised in 24×15 cm polythene bags for 5 months, till they were about 25-30 cm high. Plants were taken out from polythene bags and root system was washed with tap water to free adhering soil.

Method of inoculation :

The method used in the present studies for inoculation of the seedlings was of Klotz *et al*; (1958), Carpenter and Furr (1962) in which the entire root system and collar zone of seedlings were immersed in aerated inoculation tank. Amount of inoculum used was at the rate of one culture flask (100 ml capacity) per 5 seedlings. Seedlings were supported on glass rod and held for 24 hours in the inoculation tank. Inoculated seedlings were replanted individually in polythene bags filled with steam sterilized soil.

Disease rating :

Disease rating was determined based on the severity of symptoms of the pathogen on tap root and feeder roots. These were rated separately with numerical value of 1 to 5 as follows :

(i) **Feeder roots** : 1-No visible symptoms, 2-A few roots with symptoms of rotting, 3-Majority of roots rotted and loss of some roots, 4-Most of the roots rotted, cortex sloughed off from major roots, and 5-All roots were dead.

(ii) **Tap root** : 1-No visible symptoms, 2-Rotting symptoms confined only to the tip of the tap root, 3-Rotting in $\frac{1}{4}$ of the tap root from the tip, 4-Rotting in more than $\frac{1}{4}$ of the tap root from tip, and 5-Complete rotting of the tap root.

Observations on amount of feeder root and tap root damage, and root and shoot weights were recorded 4 months after inoculation. Based on these parameters five distinct groups, ranging from highly resistant to highly susceptible, were noticed.

RESULTS

Reaction pattern of 62 varieties of citrus and their relatives are presented in Tables 4.18 and 4.19.

Table 4.18. Reaction of Citrus species, varieties and related genera to *Phytophthora* root-rot disease.

Species	Variety	% loss			Overall reaction
		Root weight	Shoot weight	Disease rating	
<i>Citrus</i>	Coorg orange	48.56	40.43	7.33	HS
<i>reticulata</i>	Mandarin Laddu	52.48	50.00	6.75	HS
Blanco	Mandarin Lahore	32.08	38.60	5.50	S
	Nagpur orange	29.13	32.54	5.66	S
	Satsuma	24.66	23.77	4.33	MS
	Tangerin Dancey	30.89	29.71	5.16	MS
	Karamandarin	42.88	32.19	6.83	HS
	Kinnow mandarin	27.86	22.84	5.30	MS
<i>C. sinensis(L.)</i>	Sathgudi-Kodur	51.34	46.44	7.00	HS
Osbeck	Sathgudi-Coorg	45.02	42.13	5.33	HS
	Malta Blood	31.17	29.48	5.33	S
	Paramatta Malta	11.12	14.62	3.86	MR
<i>C. limon(L.)</i>	Malta lemon	55.43	41.61	7.16	HS
Burn.	Assam lemon	44.98	44.43	5.60	S
	Nakoor lemon	19.01	20.32	4.76	MS
<i>C. paradisi</i>	Grape fruit	34.26	31.92	4.66	MS
Mach.					
<i>C. aurantium L.</i>	Sour orange	4.82	9.63	3.00	HR
	Sevelli orange	6.23	8.29	3.16	HR
	Smooth Sevelli(Aust.)	3.83	5.92	3.16	HR
	Karun Jamir	15.29	17.42	4.33	MR
<i>C. pennivesiculata</i>	Baduvapuli	40.17	41.86	5.75	S
Tan.					
<i>C. limetoides</i> Tan	Sweet lime	31.16	27.89	4.67	MS
<i>C. limonia</i>	Rangpurlime - Tirupati	30.38	33.13	5.20	MS
Osbeck	Sreerampur	16.96	20.18	4.16	MR
	Chethalli	18.67	23.10	4.00	MR
<i>C. karna Raf.</i>	Lime Karna	29.67	31.28	5.16	MS

Contd.

(1)	(2)	(3)	(4)	(5)	(6)
<i>C. jambhiri</i> Lush	Khatta	28.74	29.82	5.33	MS
	Khatta Jamir	12.44	16.75	3.83	MR
	Jatti Khatti-Punjab	19.22	18.06	4.33	MR
	Jatti Khatti-Coorg	15.08	19.22	4.50	MR
	Jallendar Khatti	11.98	15.80	3.83	MR
	Pooney Jamberi	30.28	24.83	5.33	MS
	Jamberi-Bombay	43.31	42.93	6.32	HS
	Rough-lemon-Assam	29.34	26.77	3.90	MR
	Rough lemon-Coorg	28.01	32.08	4.81	MS
	Saoner-Jamberi	35.63	41.34	6.60	HS
	Rough lemon-Australia	17.40	11.68	4.50	MR
	Rough lemon-Tharsa	14.24	17.62	4.21	MR
	Rough lemon-Florida	34.58	36.44	6.50	HS
	Rough lemon-Chethalli	14.28	16.29	4.70	MR
	Jamberi-local	43.35	30.68	6.30	HS
	Jamberi-M. P.	14.00	15.30	4.10	MR
	Rough lemon-S. Africa	15.09	17.18	4.20	MR
Jamberi-Kodur	43.19	45.51	6.70	HS	
Sohmyndong	39.00	39.10	6.90	HS	
<i>C. reshini</i> Tan.	Cleopatra mandarin	26.61	18.19	4.70	MS
	Citrus China	28.39	26.64	4.51	MS
<i>C. maderaspatna</i> Tan.	Kichli	19.16	29.67	5.70	MS
<i>C. madurensis</i> Lour	Hazara	36.18	21.68	5.00	MS
<i>C. myrtifolia</i> Raf.	Chinatto(Aust.)	19.87	4.46	4.16	MR
<i>C. Aurantifolia</i> (Christm.) Swing	Acid lime	44.14	34.67	6.80	HS
<i>C. medica</i> (L.)	Citron	56.61	48.35	6.90	HS
<i>Poncirus trifoliata</i> (L.) Raf.	Pomeroy Trifoliolate	10.19	9.56	3.00	HR
	Trifosta Trifoliolate	31.00	29.96	4.52	MS
<i>P. trifoliata</i> X <i>C. sinensis</i>	Troyer citrange-Kodur	8.56	8.42	3.66	HR
	Troyer citrange-Aust.	9.33	10.49	3.66	HR
<i>C. sinensis</i>	Craizo citrange-Kodur	9.23	7.87	3.50	HR
	Craizo citrange	12.07	8.42	3.08	HR
<i>P. trifoliata</i> X <i>C. paradisi</i>	Citrummelo-Chethalli	16.25	17.14	4.00	MR
	Citrummelo-Swingle(USA)	18.15	15.48	3.87	MR
	Citromon	11.29	10.05	3.50	HR
<i>Severinia duxifolia</i> (Thumb.) Swing.	Box orange	8.62	10.31	3.00	HR

Disease rating = Feeder root + Tap root rating

HR = Highly resistant, MR = Moderately resistant, MS = Moderately susceptible,

S = Susceptible, HS = Highly susceptible.

Table 4.19. Grouping of different citrus varieties based on reaction to *Phytophthora nicotianae* var. *parasitica*.

Reaction	Varieties/Clones
Highly resistant	Sour orange; Smooth Seveli (Australia) Seveli orange; Pomeroy trifoliolate; Troyer citrange from Kodur and Australia; Carryzo citrange; citromon; Box orange.
Moderately resistant	Paramatta Malta; Citrummelo-Swingle (USA); Citrummelo (Chethalli); Rangpur lime (Chethalli); Khatta Jamir; Jattikhatti (Punjab); Jattikhatti Coorg); Jullunder Khatt; Rough lemon (Assam); Rough lemon (Australia); Rough lemon (Tharsa); Rough lemon (Chethalli); Jambiri (M.P.) Rough lemon (South Africa); Chinatto (Aust.)
Moderately susceptible	Tangerin Dancey; Satsuma; Kinnow mandarin; Grape fruit; Trifosta trifoliolate; Citrus China; Sweet lime; Rangpur lime (Tirupati); Khatta; Lime Karna; Pooney Jamberi; Rough lemon (Coorg); Cleopatra mandarin; Kichili; Hazara. Nakoor lemon.
Susceptible	Mandarin Lahore; Nagpur orange; Malta Blood; Assam lemon; Baduvapuli.
Highly susceptible	Coorg orange; Mandarin-Laddu; Karamandarin; Sathgudi (Kodur); Sathgudi (Coorg); Malta lemon; Sanerjamberi; Jamberi (Bombay); Florida-Rough lemon; Jamberi local; Jamberi (Kodur); Shomyndong; Acid lime, Citron.

Most of the varieties in *Citrus reticulata*, *C. sinensis*, *C. limon*, *C. pennivesiculata*, *C. aurantifolia* and *C. medica* showed susceptible type reaction. However a clone Paramatta Malta in *C. sinensis* obtained from Australia was found to be moderately resistant. Moderately susceptible reaction was observed among the varieties in *C. paradisi*, *C. limetoides*, *C. karna*, *C. reshini*, *C. madurensis* and *C. maderaspatna*. In all 19 different Rough lemon varieties in *C. jambhiri* were evaluated and wide variation in reaction pattern was observed. Shomyndong, Jamberi-kodur, Jamberi-local, Saoner-jamberi, Jamberi-Bombay, Khatta and Rough lemon-Coorg showed susceptible or moderately susceptible reactions while other varieties were found to be moderately resistant.

Out of 4 sour orange clones tested, 3 showed high degree of resistance while Karunjamir showed moderately resistant type reaction. Among 9 varieties of trifoliolate oranges and their hybrids tested, only Trifosta trifoliolate, was classed as moderately susceptible, *C. trummelo* as moderately resistant and remaining as highly resistant. Box orange showed high degree of resistance.

DISCUSSION

Generally two different methods of inoculation procedures in citrus *Phytophthora* have been used, one to test the susceptibility of roots of young seedlings by root inoculation and the other, the susceptibility of the stem bark of grown up trees to gummosis

(Klotz and Fawcett, 1930, Uppal and Kamath, 1936, Carpenter and Furr, 1962, Broadbent *et al.* 1971). In the present studies, seedling inoculation method was used to conserve the space, labour and time in handling large number of plants of different varieties. This is the common method employed in evaluation of citrus germplasm against *Phytophthora* root rot disease (Carpenter and Furr, 1962; Hutchison and Grimm, 1972).

In the present investigation, 62 citrus clones including 16 different species of Citrus, 7 hybrids and 2 related genera were evaluated against *P. nicotianae* var. *parasitica* at seedling stage. Growth parameters such as root and shoot growth and extent of feeder and tap root damage were used as criteria for determining relative tolerance. Carpenter and Furr (1962) used percentage of mortality of inoculated seedlings as a measure to determine the degree of tolerance to root inoculation. The susceptibility of clones in mandarin, sweet orange, lime and lemon groups to root inoculation under laboratory conditions confirmed the field observations. High degree of susceptibility was shown by most of the sweet orange seedlings, while Paramatta Malta, a clone from Australia was moderately resistant. These findings are in agreement with the results obtained by Broadbent *et al.* (1971) from Australia.

In Rough lemon clones, wide variation was noticed to *Phytophthora* infection and this perhaps may be due to existence of variation among the varieties within the species. Hutchison and Grimm (1972) found similar variation in resistance to *P. parasitica* among different Rough lemons after inoculation. This was attributed to the existence of strains which exhibited a wide range of morphological differences mainly with regard to the fruit colour. Resistant type reaction was noticed in sour orange, trifoliate and citranges. A clone Trifosta trifoliate in *Poincirus trifoliata* showed susceptible reaction while other clones showed highly resistant type reaction. This confirms the observations of Klotz *et al.* (1958) and Carpenter and Furr (1962) who reported survival of trifoliate clones to be highly variable following inoculation with *P. parasitica*. Field testing of grown up trees by bark inoculations revealed high tolerance in trifoliate, citranges and sour orange which are also highly resistant to root inoculations (Whiteside, 1973; Klotz *et al.*, 1967). Ullasa *et al.*; (1978) evaluated several root stocks used for Coorg mandarin scion under field conditions and reported high susceptibility in Coorg orange, Baduvapuli, Naichakotha, and moderate resistance in Rangpur lime, Rough lemon, Cleopatra mandarin and high resistance in trifoliate and citranges. Similar reaction was observed in the present study, when these were tested at seedling stage in the green house by root inoculation method.

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DISCUSSIONS

Koti Reddy: Have you counted the number of rotten roots?

ANSWER: No, root weight was taken.

H. S. Sohi: How old was the culture used for inoculation?

ANSWER: Fresh.

Y. R. Sarma: Do the selected varieties lose resistance later?

ANSWER: They continued to be resistant when grown up.

K. K. N. Nambiar: Could selected resistant materials be used as rootstocks?

ANSWER: Yes, this is possible.

Tuesday, 23 September 1980
9.00 a.m. — 1.00 p.m.

SESSION 5

FOOT ROT OF BLACK PEPPER

Chairman: **DR. R. S. MEHROTRA**

Rapporteur: **DR. R. NAIDU**

FOOT ROT DISEASE OF BLACK PEPPER (*PIPER NIGRUM* L.)

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Foot rot (Quick wilt) is the major disease affecting black pepper (*Piper nigrum* L.) causing severe crop loss, thus limiting pepper production in all pepper growing tracts of the world. The fluctuations in production and consequent changes in market prices have been partly attributed to the crop loss incurred due to foot rot. The problem has been reviewed previously by Muller (1936) Holliday and Mowat (1963) and Nambiar and Sarma (1977).

History and distribution of the disease.

The first report of sudden collapse and death of the pepper vines came from Lampung (Indonesia) in 1885. It was Muller (1936) who identified the causal agent as *Phytophthora* sp. and named it as *P. palmivora* var. *piperina*. In Sarawak (Malaysia) the disease was reported in 1941 (Newman, 1941; Thompson, 1941) and the severe out-break occurred in 1952 (Miller, 1953). In India the report on the incidence of root diseases of black pepper dates back to 1902 (Menon 1949) when Barber (1902, 1903, 1905) and later Butler (1906, 1918) investigated the disease in Wynad (Kerala); however these investigations were inconclusive. Although *Phytophthora* sp. isolation was reported from black pepper in Mysore area (Venkata Rao, 1929) the first authentic record of *Phytophthora* wilt of black pepper in Kerala came from Samraj and Jose (1966) who adopted Muller's identification of *Phytophthora*. The disease was also reported from Puerto Rico (Gregory, Almeyda and Theis, 1960), Brazil (Holliday, 1965, Albuquerque, 1966, Alconero *et al.*, 1972), Jamaica (Leather, 1967) and Thailand (Tsao and Tummakate, 1977). Involvement of *P. palmivora* in the large scale deaths of pepper vines in Sambirano river in the north western part of Malagaican of Malagasy Republic has been reported (de Waard, 1979).

Crop losses

The disease is soil borne, associated with high soil moisture and spreads through soil water. It is more destructive in areas of monoculture as in Sarawak. In India Samaraj and Jose (1966) recorded vine death up to 20% in Cannanore District (Kerala) while Nambiar and Sarma (1977) recorded 25-30% loss in some gardens in Cannanore and Calicut districts. A similar loss was recorded in Sarawak (Robertson, 1955). In Sarawak during 1953-56 the pepper loss was about 7000 tons amounting to £ 1.7 million (Holliday and Mowat, 1963). Vine death of about 10% was recorded in West Borneo (Leefmans, 1934.) An out break of foot rot occurred during 1967-68 in Lampung area destroying 40-50% of pepper area (de Waard, 1979). The overall loss due to foot rot in major pepper growing countries of the world, when estimated at 3-5% loss of the total planted area, would amount to US \$ 4.5-7.5 million per annum (de Waard, 1979).

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Symptoms

There is a general similarity of the disease symptoms in all pepper growing countries. All parts of the plant are susceptible and as such exclusive root rot, collar rot, aerial vine death, leaf infection or spike infection either alone or in different combinations may occur. Foliar infection is not fatal as compared to collar and root rot.

(i) **Foliar infection** : Direct infection of the foliage often occurs apart from foliar symptoms expressed as a result of collar and root rot. Leaf infections generally appear in the lower region of the bush, and might be due to soil splashing during heavy rains. However the authors observed leaf infection even at 3-4 m height of the bush in some plantations in Kerala and Karnataka. These are in conformity with the observations of Turner (1969b). Leaf infection can start from any part of the lamina and appears as water-soaked lesion with smooth or fimbriate margins, which advance rapidly. The mature lesion will be about 0.5 cm or more and dark, some times covering up to half of the lamina. They are either uniformly smooth or may show concentric zonations. Muller (1936) failed to observe the zonate pattern in lesions which were reported from Sarawak (Holliday and Mowat 1963). Turner (1969b) observed that the different types of lesions reflected different conditions of incubations. Under continuous humid conditions fimbriate lesions develop and, when wet and dry conditions alternate, zonate pattern results. Irrespective of the type of lesions heavy defoliation occurs due to leaf infection. Immature leaves are highly susceptible than mature ones and the lower surfaces of leaf is more susceptible than upper surface (Turner 1969b).

Spike infection results in their heavy shedding. The infection occurs at stalk portion resulting in its necrosis which progresses along the floral axis. Occasionally berries also get infected. Aerial infection of the stem occurs at any point of the vine, even at a height of 2-3 M. The infected region turns dark due to wet rot. The foliage of the infected twig turns yellow and drops off. The rotting progresses both upwards, and downwards and often results in die-back.

(ii) **Collar and root rot infection** : The infection of collar and root goes undetected until the appearance of foliar yellowing. Infection occurs either at the collar or just above or below the soil level. The infected portion appears slightly dark at the point of attack. When the cortex of the affected portion is chopped off, it shows the healthy yellow patch followed by a necrotic area. The vasculature of such infected stems turns darker. Vascular discolouration up to 0.5 m beyond the point of infection has been observed in many cases but not consistently (Nambiar and Sarma 1977). With progress of the disease the cortex gets disintegrated and peeled off. The rotting progresses further into soft medullary tissues leaving the xylem strands loosened. The infection of the collar gradually progresses downwards and spreads to the root system. Occasionally one vertical half of the stem alone is involved in the infection leaving the other half normal. A careful study is needed further to understand the mechanism underlying such type of rotting. In some bushes a single vine dies leaving the adjacent vine on the same standard unaffected. The general absence of foot rot in young vines might be due to disease escape rather than to juvenile resistance, since young vines succumb when inoculated artificially.

Apart from collar rot infections which spread to the roots, exclusive root infection occurs. The infection generally starts on tender lateral roots and progresses towards the mature root reaching finally the underground stem. Random infections at irregular intervals on the lateral roots are also seen, possibly due to the fungal invasion at some injured points. The number of roots affected and the extent of rotting determine the speed of death of the infected vine (Muller, 1936; Holliday and Mowat, 1963; Nambiar and Sarma, 1977). In general the affected vine may succumb within 15 to 45 days after infection depending upon the severity of the damage of the affected parts.

Intervenal chlorosis is the first foliar symptom of root and collar rot. Foliar yellowing, flaccidity, defoliation, breaking off of the stems at nodal regions and spike shedding are the general aerial symptoms noticed in the case of root rot and collar rot. Holliday and Mowat (1963) reported that cultivation of pepper in mounds favoured root rot infection. Both collar and root rot infection are common in India, where mound cultivation is not the practice. While collar infection is more common in slopy lands, root rot is common in the plains on level lands.

Isolation of organism

Holliday and Mowat (1963) reported successful isolation of *Phytophthora* from fresh lesions of foot rot affected stems and roots of black pepper using plain agar medium. Isolation from the tissues of advanced stage of infection were always negative. Turner (1964) reported successful isolation from the soil using apple bait technique. The authors experienced difficulty in isolation of *Phytophthora* both from infected tissues and soil using apple, castor seed, cacao pod and pepper leaf disc baits. This might be due to quick colonisation of infected tissues by saprophytic bacteria, *Fusarium*, *Rhizoctonia* and *Pythium* sp.. The authors were successful in selective isolation of *Phytophthora* from foot rot affected tissues and from soil with pepper leaf disc baits using PVPH medium of Tsao and Guy (1977).

Taxonomy and biology of the causal organism

Except in Brazil where *Fusarium solani* var. *piperi* appeared to be the major causal agent of foot rot (Albuquerque, 1961), *P. palmivora* has been identified as the causal agent of foot rot of black pepper in all other countries. Recently *P. capsici* has been reported from Indonesia (Kasim, 1978; de Waard, 1979).

In recent times taxonomy of *Phytophthora* in general and *P. palmivora* in particular received a greater attention with respect to the sporangial morphology, caducity and chromosome type (Tsao, 1977a; Griffin, 1977; Kaosiri, Zentmyer and Erwin, 1978; Brasier and Griffin, 1979). Turner (1969b) referred isolates from *Piper betle* and *P. nigrum* as atypical strains of *P. palmivora*. Waterhouse (1974) recognised black pepper isolates (BP) as atypical (Piper form) of *P. palmivora* which differed from morphological forms 1 and 2. The sporangia of black pepper isolates are ovoid, obovoid, pyriform or fusi-form which are caducous with long occluded pedicels. However isolates from Sarawak were reported to be non-caducous with no pedicel (Holliday and Mowat, 1963). The black pepper isolates from Thailand showed unique sporangial morphology in that they are highly caducous with long pedicels, with biseptate sporangia and umbellate type of

sporangial arrangement. The sporangia with tapered base showed L. B. ratio of 2.5 and that with rounded base 1.8. Further the isolates from Malaysia, Central America and Africa had similar morphology as that of Thai isolates and are considered to be Morphological form 4 (MF 4) of *P. palmivora* (Tsao and Tummakate, 1977). Caducity of black pepper isolates was influenced considerably by the medium. Studies on length of the pedicel in 4 isolates showed a range from 37.3 to 73.4 (AL-Hedaithy and Tsao, 1979a, 1979b). Idosu and Zentmyer (1978) opined that BP isolates are similar to MF4 of *P. palmivora* of cacao. The isolates from Thailand probably have chromosome number $n = 9-12$ (Brasier and Medeiros, 1978). According to Brasier and Griffin (1979) the BP isolates from Sarawak, Thailand and Puerto Rico showed petalloid type of colonies with dense aerial mycelium, sporangia with shallow papilla with base tapered towards the stalk, with long occluded pedicels and are similar to MF4 of *P. palmivora* and opined that it should be given a species status.

The fungus grew luxuriantly at 25-28°C on oats agar (Turner, 1969b) and growth was absent at 35°C. Sporulation was maximum at pH 6.0 and absent at pH 3.0 (Turner 1969a). Zoosporangial germination in sporulating discs in water was noticed even at 20-24°C; however when cold shock was given at 10°C for 15-20 min, the germination was enhanced (Sarma and Nambiar, unpublished). Turner (1962) reported oospore formation in BP isolates of Sarawak when the cultures were stored for 2-3 months. Homothallic oospore formation in single A1 or A2 compatible types in 3 out of 7 BP isolates was noticed when aged inoculum of about 12 months old, was plated on oats agar (Tsao, 1979). Ageing of the culture appears to have direct relation to sex organ formation. Holliday and Mowat (1963) obtained oospore formation when BP isolates of Sarawak were paired with *Phytophthora palmivora* isolates from cacao or *Citrus reticulata*. The authors observed the oospore formation in BP isolates when paired with *P. palmivora* isolates from cacao or rubber. When two compatible types of BP isolates inoculated on pepper leaves and incubated at 20°C in dark, oospore formation was noticed and was absent at 30°C (Brasier, 1969a; 1969b). Brasier opined that oospore formation was likely to occur in nature, though rarely and their production was confined to woody tissues or debris, which were protected from light. *Trichoderma viridae* stimulated oospore formation in *P. palmivora* in BP isolates (Brasier, 1972). Presence of both A1 and A2 compatible types was noticed in BP isolates in Sarawak (Brasier, 1978). Formation of oospores by *Phytophthora* in response to *Trichoderma* has been hypothesised as a defense response to potential antagonist or competitor and is important from the point of survival of the fungus. The *Trichoderma* effect was confined to A2 mating types and the stimulus was volatile in nature. Out of 4 BP isolates tested 3 showed mixed response to "Trichoderma effect" (Brasier, 1975, 1978). Oogonia in BP isolates were morphologically similar to 'S' chromosome (MFI) type, often oval, and on carrot agar golden coloured and turned dark when treated with acetorcin (Brasier and Griffin, 1979.)

Toxin

The pathogen being a necrotroph, primarily colonises the parenchymatous tissues of cortical region and brings about their disintegration and creates a condition similar to that under water stress resulting in death of the vine. Typical vascular browning is noticed in the stems and roots of affected vines. The fact that vascular browning is noticed beyond

the point of infection suggests the possible involvement of toxemia in disease syndrome. Incidentally Lee(1973) reported toxin production by BP isolates and it was related to virulence. He used toxin as a marker to screen pepper varieties for foot rot resistance. Keen *et al.* (1975) reported the presence of mycolaminarins and B-1 glucans in the mycelium of *P. cinnamomi*, *P. palmivora* and *P. megasperma* var. *sojae* and they were phytotoxic to soybean, cacao and tomato shoots. The authors found that cell free culture filtrates from BP isolates of *P. palmivora* could induce vascular browning and flaccidity of leaves in cut shoots of pepper (Anonymous, 1977a).

Spread

Flow of surface water and root contact between healthy and infected plants appear to be the primary mode of disease spread. In hilly terrain, the vines in the valley get infected early and the infection spreads gradually, more downwards, supporting the view that spread is mainly through soil and water. In the plains, infection occurs in a sporadic manner and spreads to the adjacent vines. The spread is rapid in neglected plantations with infected vines around.

We studied the spread of the disease in an arecanut plantation having pepper as an intercrop and observed that within two years, about 250 vines died. Regular irrigation as well as cultivation in the gardens could have aided the quick spread of the pathogen. Incidentally the authors could isolate *Phytophthora* from drainage water in some arecanut gardens intercropped with pepper where foot rot incidence was noticed in Sirsi area of Karnataka. Role of irrigation water in causation and spread of disease in betel vine plantations has been reported (Singh and Chand, 1973).

The rate of spread appears to be much more rapid in Sarawak, and it has been ascribed to the prevalence of continuous wet seasons coupled with application of large amounts of organic fertilizers and bare soil cultivation without any weed growth. Movement of farm personnel from diseased to healthy gardens and usage of same farm implements in these gardens also help in disease spread (Holliday and Mowat, 1963). They have proposed that the grass cover and weed growth in Indian pepper plantations may be impeding the spread of the pathogen.

Vines of all age groups are infected under field conditions. However, the incidence is less common in vines during the first three years after planting and maximum incidence is noticed after the first five years.

Muller (1936) and Holliday and Mowat (1963) have suggested that infection of leaves in the lower region of the bush might be due to rain splash only. Turner(1969b) observed that number of leaves exhibiting leaf lesions was more at lower heights of the vines than at the top and felt that heavy rainfall and wind could contribute to aerial spread of the disease. Aerial transmission of zoospores of *P. palmivora* has been reported in coconut (Britton-Jones, 1940), rubber (Chee, Lim and Wastie, 1967) and cacao (Thorold, 1952).

Many agents aid the dispersal of the zoospores of *Phytophthora* (Turner, 1964, 1967). *Phytophthora* has been recovered from the faeces of the giant African snail, *Achatina fulica*, which often feeds on infected foliage. He opined that this might serve as an effective

mode of spread during the hotter months of the year. Spores of *Phytophthora* have been isolated from the ant runs of *Crematogaster* (Turner, 1972). The ants carry the spores with soil while constructing tunnels on posts supporting pepper vines and thus spread the spores. The authors isolated *Phytophthora* from the soil deposited by termites on the live standards in a few infected bushes. This perhaps suggests the possible passive spread of the disease propagules during off season. Viable chlamydospores of *P. cinnamomi* have been found in the intestinal tracts of termites (*Nasutitermis exitiosus*) and faeces of two species of forest birds indigenous to West Australia Jarrah forests and thus serving as vectors of the pathogen (Keast and Walsh, 1979). Studies are warranted on various biological agents as disease carriers and these are of epidemiological importance for a better understanding of the disease spread during off season.

Climatic factors

The disease incidence is generally high during the South-West monsoon (June-September), in Kerala, India when rainfall (2270-2990 mm) and relative humidity (91-99%) are high and minimum temperature (19-23°) is low. In Sarawak, maximum disease incidence was observed during October-March when the mean maximum and minimum temperatures are about 26°C and 21°C. The mean rainfall during the period is 318-653 mm and daily mean sunshine 4.3 hr (Holliday and Mowat, 1963). The pathogen was successfully isolated in 60% of cases during October-March, but only in 43% during the dry months (April-September). Sarawak does not have a long dry season as the west coast of India has. On overcast days with low mean sunshine hours and heavy rainfall, the temperatures generally fall, and such conditions are conducive for spore release.

The ambient temperature plays an important role in the infection process. When the pathogen is inoculated on rooted cuttings, symptoms of root necrosis and foliar yellowing appear in 3-4 days when inoculated plants are incubated at 20-25°C. The symptom expression is delayed when the temperature is 28°C and above (Nambiar and Sarma, 1977). In one year old cuttings, symptoms appeared only after 35 days at 28-30°C. Selvaraj (1966) working on betel vine wilt in Tamil Nadu found that wilt incidence was maximum (100%) at low soil temperature (20-23°C). The incidence of betel vine wilt was maximum during December-January period when the temperatures fall below 23°C (Rao, Vidyasekharan and Narasimhan, 1969).

In pure pepper plantations in India, the disease generally becomes apparent during the south-west monsoon period. In mixed cropping systems such as pepper in arecanut gardens (which are generally irrigated during the dry period), the disease is noticed during the post monsoon period (November-January) also. Since during the winter months, the minimum temperature falls to 16-21°C and soil water content remains high due to frequent irrigations (once in 4-5 days), the microclimatic factors are very congenial for fungal growth, sporulation and zoospore emission.

Soil factors

In India, Nambiar *et al.* (1965) observed heavy incidence of the disease in neglected pepper gardens where inorganic fertilizers were not being applied. Kliejunas and Ko (1974) reported that deficiency of inorganic nutrients contributed to the heavy incidence of Ohia

decline (*Metrosideros collina* (Forst.) Gray subsp. *polymorpha* (Gang.) (Rock) associated with *P. cinnamomi*. Broadbent and Baker (1974) reported that exchangeable Ca, Mg, N and organic matter were high in soils suppressive to root rot of avocado caused by *P. cinnamomi* as compared to soils conducive to the incidence of root rot. Application of super phosphate had a suppressive effect on betel vine wilt in Tamil Nadu (Thyagarajan *et al.*, 1972). Working with quick wilt, Nambiar *et al.* (1965) reported that surface soils in diseased gardens contained lower levels of Ca, Mg, and K with high N. They suggested that if the ratios of K/N, available K/N and $\text{CaO} + \text{MgO} + \text{K}_2\text{O}/\text{N}$ fell below 1.14, 0.05 and 3.80 respectively, then the area would be prone to the disease. Huber and Watson (1974) found that the type of nitrogenous source also determined disease incidence in several root diseases involving Pythiaceae fungi.

Since the final outcome of root rot and foot rot caused by *Phytophthora* is gradual wilting simulating a stress syndrome, information on the plant and soil water relationships of healthy and diseased plants, and the effect of water potential on the fungus are essential, for a better understanding of the disease etiology and management in black pepper wilt. Although the major pepper crop is rainfed, pepper intercropped in arecanut gardens with regular irrigation, is more vulnerable, and regulation of soil moisture becomes more important to reduce the disease. In a recent study on *Phytophthora* root rot of avocado on water relation, the symptoms were similar to water stress resulting from low xylem pressure potential due to increased resistance to flow in the soil plant system even in well watered soils (Sterne, Kaufmann and Zentmyer, 1978). At present no information is available on soil water relations and the effect of nutrition on the incidence of pepper wilt. Hence such studies are warranted in this line.

Survival

Holliday and Mowat (1963) observed that *P. palmivora* from pepper survived for about 15 weeks in naturally infected underground stems. The authors could not isolate the fungus from the infected soil stored for 4 months. The pathogen, however, has a low saprophytic ability (Holliday and Mowat, 1963). Survival of *P. palmivora* on rubber for about 32 weeks in soil was reported (Chee, 1973). Brasier (1969a) observed oospore formation *in vitro* when two compatible types of the pathogen were present under congenial conditions of low temperature (20°C), darkness and adequate food supply. Oospore formation has been presumed to occur in nature though rarely, in woody tissue or debris not exposed to light. We have not however been able to notice oospores so far in infected tissues. Holliday and Mowat (1963) also did not observe any fusion organ in nature. Although the survival of the fungus is reported to be for about 15 weeks, the exact mode of survival is not known. Fluorescent labelling techniques with brighteners (Tsao, 1970) or fluorescent antibody techniques (Macdonald and Duniway, 1979) might be useful in studies on survival of the fungus, since oospore formation is not noticed in infected tissues. The mode of perennation and population build up of the pathogen during different seasons and under varied soil, water, and temperature regimes are to be studied in depth for developing effective control measures.

Host range

P. palmivora has been recorded to infect 138 species belonging to different families

of angiosperms (Chee, 1969) Turner (1971a) reported that the isolates of *P. palmivora* from pepper in Sarawak were highly host specific and none of the 43 species from 40 genera, belonging to 20 families other than the Piperaceae, was susceptible. Out of 32 *Piper* species tested, 30 species including *P. betle* were susceptible. All the seven *Peperomia* spp. were resistant. Further the leaves of *Lycopersicon esculentum*, *Solanum melongena* and *Vinca rosea* and fruits of *Areca catechu* and *S. melongena* were also occasionally infected under laboratory conditions. The authors found that *P. palmivora* isolates from pepper in Kerala infected roots of *P. betle*, *P. longum*, *P. attenuatum*, cacao pods, tender leaves of rubber, castor, and caused mild rotting of capsules of cardamom. *Phytophthora* isolates from cacao, cardamom, betel vine, palmyrah, oil palm, areca and *Ficus* showed differential reaction on leaves and root system of black pepper. Pepper gardens in the vicinity of rubber plantations usually show heavy incidence of pepper wilt. The information on positive cross inoculation of different *P. palmivora* on pepper, though important, is not of epidemiological significance unless the same strains are isolated from the infected pepper tissues. The pepper isolates examined so far by the authors are distinct from the *P. palmivora* from rubber, cacao, palmyrah and cardamom. Holliday and Mowat (1963) reported that *Phytophthora* isolates from *Colocasia* and cacao did not infect leaves of pepper, while an isolate from citrus did. According to Muller (1936), *Phytophthora* isolate from pepper in Indonesia showed similar characters in culture to those from coconut, rubber, cacao and papaya. Isolates of pathogen from pepper were, however, less virulent to the other host plants than those from the respective hosts.

Role of other associated organisms.

Holliday and Mowat (1963) have often isolated *Rhizoctonia solani* and *R. bataticola* from the roots and stems of pepper. They appear to be the earliest colonisers of *Phytophthora* infected tissues. The present authors have also made similar observations. We have isolated *Pythium* sp. from tender discoloured roots of pepper vines showing foliar yellowing and also from roots of quick wilt affected vines. Incidentally, Holliday and Mowat (1963) have frequently isolated *P. splendans* from small roots of pepper in Sarawak causing damping off in pepper seedlings.

The authors have isolated *Trichoderma* sp. from the roots of healthy pepper vines and also noted lysis of mycelium of BP isolate of *Phytophthora* when *Trichoderma* sp. overgrown over the former in the culture plates. *Trichoderma* as biological control agent of betel vine wilt (Tiwari and Mehrotra, 1958), root rot of avocado (Zentmyer, 1963, 1967) and of many other *Phytophthora* diseases has been reviewed (Baker and Cook, 1974). However the reports of Brasier (1978) regarding the response of *Phytophthora* to *Trichoderma* in oospore formation has been opined as of 'survival value' and needs further investigation on its utility as a biological control agent.

The plant parasitic nematode *Meloidogyne incognita* and *Radopholus similis* are being increasingly observed in pepper plantations. Holliday and Mowat (1963) observed that infestation by *Meloidogyne* sp. did not significantly enhance the susceptibility of pepper vine to foot rot. Selvaraj (1966) also made similar observations on betel vine wilt in Tamil Nadu. However, critical studies of the fungus-nematode interaction (Powell, 1971)

are required to be carried out in the case of pepper under conditions of both pure cropping and mixed cropping.

Resistance

Muller (1936) reported the black pepper variety Belantung from Indonesia as resistant to foot rot. Holliday and Mowat (1963) found the Indian pepper cultivar Uthirankotta and the Indonesian varieties Djambi and Belantung possess appreciable resistance. However, the present authors tested 40 Indian cultivars including Uthirankotta and 45 wild types adopting root dip inoculation technique (Sarma and Nambiar, 1979) and found all of them to be susceptible. Uthirankotta has also been found to be susceptible in Puerto Rico (Alconero *et al.*, 1972) and Sarawak (Turner, 1973a). Turner (1973a) however, found Balancotta to be highly resistant. The observations of differences in reaction of a particular type may be attributed to the differences in virulence of the isolates of the pathogen.

As already reported, Turner (1971a) screened 32 *Piper* species and found that *P. colubrinum* and *P. obliquum* var. *eximium* were resistant. Albuquerque (1968a, b) had earlier reported resistance in *P. colubrinum* in Brazil. In Ghana, *Piper guineense* has been reported to be resistant (Anonymous, 1977b). Ruppel and Almeyda (1965) reported that out of five *Piper* species tested, *P. aduncum*, *P. scabrum*, and *P. treleanum* showed partial resistance. Several workers in Puerto Rico, U.K., Brazil, and Malaysia (Sarawak) have successfully grafted *P. nigrum* on to a number of *Piper* spp., both resistant and partially resistant (Gaskins and Almeyda, 1969; Garner and Beakbane, 1968; Albuquerque, 1968a, b; Turner 1973a). However, field establishment has been reported only in the combination involving *P. nigrum* and *P. colubrinum* (Albuquerque, 1968a, b., Gaskins and Almeyda, 1969). Grafts with root stocks of pink form of *P. colubrinum* with 'Kuching' variety as scion, grew much faster as compared to root stocks of green form of *P. colubrinum* and no signs of degeneration of graft union was noticed even after 4 years (de Waard, 1979). In some combinations involving *P. colubrinum* and cultivars like Balancotta, Kalluvalli, and Singpuri, Alconero *et al.* (1972) observed that longitudinal cracks developed at the graft union after a normal growth of four years. The incompatibility of the grafts might be due to anomalous secondary growth and consequent poor callus growth.

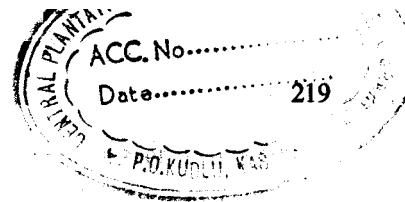
Control

Only limited success has been achieved in controlling the disease with fungicides in trials carried out in Malaysia (Sarawak) and India. Holliday and Mowat (1963) reported that heavy doses of a copper oxide (Perenox) reduced the disease incidence slightly when it was forked into the soil in the basins of the vines. Using Actidione was also suggested to control foot rot and root rot of pepper (Lee and Verghese, 1974.) In Indonesia, Harper (1974) recommended copper fungicides against *Phytophthora*. Among the ten different commercial formulations of fungicides tested by the present authors as foliar sprays and soil drenches around the vines, before and after South West monsoon, only Bordeaux mixture spraying and application of Bordeaux paste to the stem from the collar region to a height of about one meter reduced the incidence. In tests against betel vine wilt in India, spraying and drenching the soil with Bordeaux mixture alone checked the disease (Narasimhan *et al.* 1976). None of the 29 chemicals tested by Turner (1973b) was fungicidal.

He opined that concentration-volume-time interaction determined the efficacy of the formulation and soil permeability would be a major factor in penetration. In Sarawak, captafol has been reported to be useful as a soil drench against *P. palmivora* (Anonymous, 1972). We, however, did not find it to be effective. Recently, Noveroske (1975) reported pyroxychlor (Dowco 269) which is known to have basipetal translocation, to be effective against *P. parasitica* in tobacco. But in our *in vitro* studies this fungicide was ineffective even at 2000 ppm concentration. Suppression of root rot of avocado caused by *P. cinnamomi* by alfalfa meal (Zentmyer, 1963), suppression of *P. cinnamomi* and *P. parasitica* in 0.1% urea amended soils (Tsao and Zentmyer 1979) and also suppression of root rot of citrus caused by *P. nicotianae* var. *parasitica* using several organic amendments (Tsao, 1977b) are of greater relevance to the black pepper foot and root rot problem. In view of high rainfall conditions at a stretch for 3-4 months during South West monsoon period in India and consequent leaching off of the soil fungicides applied as a prophylactic measure, greater stress should be on biological control aspects, manipulating microbiological status of the soil suppressive to the pathogen, and thus ensuring a better protection and boosting up the health of the vines. Agronomic practices like earthing up around the vine ensures greater root regeneration, and consequently the health of the vine, since ratio of root regeneration to root degeneration determines the health of the vine (Nambiar and Sarma, 1977). Better drainage conditions helped in reducing the foot and root rot of pepper in Sarawak (de Waard 1979). Lysis of mycelium of *P. cinnamomi* and sporangial abortion was reported when the organic matter of the soil was 50% or more (Nesbitt, Malajczuck and Glenn, 1979).

Holliday and Mowat (1963) and Nambiar and Sarma (1976) have stressed the need for adopting phytosanitary measures under field conditions to reduce the inoculum in soil and thus check the disease. These included isolation of infected plants from the surrounding healthy vines, ensuring better drainage facilities and burning infected pits or drenching them with Bordeaux mixture before replanting. Chemical control measures are meaningless without adequate phytosanitary measures to check the disease spread. Turner's (1969a) observation of complete inhibition of sporulation and disease is theoretically possible in soils with low PH, but it will not be possible to rule out nutritional disorders under such circumstances (de Waard, 1969). In India, where the pepper soils have a PH of 4.5-5.8, Nambiar *et al.* (1965) recorded low K, Mg, and Ca levels in diseased soils and recommended application of lime, magnesium and potassic fertilizers in balanced amounts to prevent the disease.

In the absence of a highly resistant variety to foot rot disease at present (Nair, 1978) an integrated system of disease management involving phytosanitary measures, chemical and biological control methods, combined with efficient agronomic practices that boost up the vigour and health of the vine, are of greater relevance in the present context for tackling this serious scourge of black pepper. While priority should be given to screening wild types of *Piper* spp. for locating resistance to the pathogen, ecological studies on the factors that predispose the vines to infection under different cropping systems and on the possible existence of strainal variations are also to be made. This will enable us to develop an efficient forecasting system to make the control measures more effective and meaningful.



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DISCUSSIONS

T. N. Sreenivasan : What was the height of the graft? Any reference on use of root-stock for pepper?

Answer : About 3-month-old grafts are transplanted in the field. *Piper colubrinum* has been used as a root-stock both in Puerto Rico and Brazil. But the field establishment is poor. Longitudinal cracks developed at the graft union.

F. J. Newhook : How much time a grown up vine takes to express the foliar symptoms such as yellowing, defoliation etc.?

Answer : We have not worked out the time taken to express foliar symptoms in mature vines after inoculation. In rooted cuttings foliar yellowing can be seen in 8-15 days after inoculation.

P. H. Tsao : What type of symptoms you observe with 'Foot rot' disease?

Answer : Type of symptoms depends on the situation in which pepper is grown. When pepper is grown in hilly slopes with good drainage condition, collar rot or foot rot is more common. When pepper is crop-mixed with arecanut, coconut and under high soil moisture and thick shade, root rot symptom is generally prevalent. The leaves start drooping and branches at nodal region break. The whole foliage drops off leaving a bare vine.

P. H. Tsao : Early detection of infection is not possible in perennial crops like citrus, avocado and other tree crops, because after damaging certain amount of root system, then only the infected tree expresses foliar symptoms. In pepper also death may not occur immediately after infection. It may take 6-8 years to express symptoms. If we are able to detect the infection at an early stage, we can prevent the death of the vine.

J. Subbaya : Are the two symptoms, i.e. collar rot and 'foot rot' distinct? Can we save the affected vines by fungicidal treatment?

Answer : Infection of mainstem occurs at the soil level and some times just below the soil level. Collar and foot rot are one and the same. Application of

Bordeaux paste after scraping off of the affected portion may help in saving the vine, if detected at an early stage.

P. H. Tsao : Host range studies with MF4 isolate is very useful.

P. W. F. de Waard : *P. palmivora* seems to infect large number of hosts including arecanut and the pathogen could survive on these hosts and may serve as a source of infection.

Answer : The pepper isolate has not been isolated from these hosts so far.

S. Y. Padmanabhan : If early detection is not possible why Bordeaux paste has been recommended for infected vines? Is it not a waste of money?

Answer : The recommendation is to apply Bordeaux paste as a prophylactic measure of control. However, if symptoms are detected at an early stage itself, the infected portion can be scraped off and Bordeaux paste applied.



SOME ASPECTS OF EPIDEMIOLOGY OF FOOT ROT OF BLACK PEPPER

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ABSTRACT

Foot rot caused by *Phytophthora palmivora* is the most important disease of black pepper, *Piper nigrum*, and causes severe losses to the crop in many countries where pepper is grown. Studies on the disease incidence showed that the disease spread in a centrifugal fashion. The incidence was more in years of heavy rainfall during South West monsoon period. During July the temperature especially soil temperature used to be around 20-24°C for a few days when R. H. also used to be above 90%. The fungus was isolated in greater frequency during July-August mainly from stem, root and soil. The fungus could not be isolated in summer when the soil moisture was less and soil temperature high. The fungus was present at higher intensity at the basal soil and increased with increasing soil moisture. On the vines the fungus could be isolated in higher frequency from soil particles adhering to vines at 25-50 cm height than at higher points.

INTRODUCTION

Among the diseases affecting black pepper, *Piper nigrum* L., foot rot (quick wilt) is by far the most important one inflicting severe losses to pepper gardens in India (Samraj and Jose, 1966, Nambiar and Sarma, 1977), in Indonesia (Muller, 1936), in Sarawak (Robertson, 1955, Holliday and Mowat, 1963) etc. The disease usually occurs during the South West monsoon period (June to September) in India. In Sarawak, maximum disease incidence was observed during October-March, the wettest months of a year (Holliday and Mowat, 1963). Holliday (1961) and Holliday and Mowat (1963) made some preliminary studies on epidemiology of foot rot disease of black pepper. In this paper disease spread in a garden over a period of 6 years, the effect of rainfall, temperature and humidity on disease development and the seasonal effects on fungus are presented.

MATERIALS AND METHODS

The study was carried out in a pepper garden at Bandadka, about 60 km away from Kasaragod, on the Western Ghats. The pepper vines are 25 years old. The cultivar of pepper is locally called "Wynadan Valli" which is very much similar to Balancotta. The disease incidence was judged from the number of vines dead after showing the typical symptoms of the disease.

Air temperature and relative humidity were continuously recorded in a two-point automatic thermohygrograph kept in the garden under a shelter. Soil temperature was

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recorded at 5, 15 and 30 cm depths in the garden. In the absence of any rainfall data available for the garden or in the locality, rainfall measurements made at CPCRI Kasaragod were made use of in this study.

Soil cores were taken at monthly intervals from April onwards and soil moisture content determined. A portion of the soil sample (50 g) was used for determining the presence of fungus using castor seed and leaf disc bait techniques at 24°C. The soil samples were taken at 0, 5, 15 and 30 cm depths at 0 and 50 cm distances. Soil samples were also taken from the surface at 100 and 150 cm distances from the vines.

Soil particles were caught at 25, 50 and 75 cm heights on the undersurface of microscopic slides covered in the centre longitudinally with cello tape strips of 10 mm × 70 mm with the adhering surface facing downwards. The slides were recovered after the rains and examined directly under microscope and the number of soil particles adhering to the entire length of the cello tape strip was counted. Soil particles adhering to the basal portions of five vines of more or less uniform thickness were also collected at 25, 50, 75 and 100 cm heights and using castor baits, the presence of the fungus was assessed.

RESULTS

Spread of the disease : The data on the incidence of the disease observed from 1974 onwards in the pepper garden at Bandadka are presented in Table 5.1 and Fig. 5.1.

Table 5.1. Incidence of foot rot disease of pepper during 1974-79 period in Bandadka (Total No. of vines in the garden is 1060).

Year	Vines killed by foot rot		Rainfall in mm*	
	Number	Percentage	From May-September	Total
Earlier to 1974	368	34.71	—	—
1974	25	2.36	3292 A	3550
1975	12	1.13	3978 B	4332
1976	18	1.69	2708 C	3052
1977	27	2.55	3167 A	3705
1978	51	4.81	4356 A	4656
1979	14	1.34	2899 C	3095

*In the absence of rainfall data at Bandadka, data from Kasaragod are used here.

- A. Received good premonsoon showers in May followed by heavy rains in June-July.
- B. Comparatively less rainfall in July.
- C. Practically no premonsoon rains.

From the data it is seen that in some years (1974, 1977 and 1978) the percentage incidence was more than that in other years. It is also seen from FIG. 1. that the disease spreads in centrifugal fashion from a single infection point. Scattered incidence of disease also is seen from which the disease again spreads outwards.

The maximum disease incidence (4.8%) in 1978 coincides with the highest rainfall received during the year (4656 mm) with 93.5% of rains precipitated during S. W. monsoon period. However, in 1975 recording, the second highest rainfall during the period of study, the disease incidence was only 1.13%. It is observed that during July 1975, the rainfall was comparatively less (950 mm) than in other years (1128-1587 mm). Comparatively no rain was received during summer of 1976 and 1979 and the S.W. monsoon rainfall (2700-2900 mm during May-September) was the lowest in those years.

During July-August months the relative humidity was maximum (mean 87-90%) and ambient temperature ranged between 22.5-28.0°C (Table 5.2). The mean soil temperature ranged between 22-28°C. It was also noticed that the soil temperature ranged between 22-24°C for 6 days in July in 1977, 20-24°C for 8 days in July in 1978 and above 23°C only in 1979. In 1978 July, RH was above 90% for most of the days.

Table 5.2. Rainfall, temperature and relative humidity during different months in a pepper garden at Bandadka during 1977-79 (Mean of 3 years)

	Rainfall		R.H. %	Ambient temp.		Soil temp. range A (°C)
	mm	No. of rainy days		Max. °C	Min. °C	
January	—	—	71	32.0	21.0	26.5—28.5
February	—	—	73	32.0	22.0	28.0—30.5
March	6.4	1	74	33.0	23.0	30.5—33.0
April	27.4	2	73	33.0	24.0	29.0—32.5
May	267.2	12	75	33.0	24.5	28.5—33.0
June	1040.3	26	86	33.0	24.0	24.0—31.0
July	1335.3	29	90	28.0	22.5	22.0—26.0
August	603.7	21	87	28.0	23.0	23.5—28.0
September	224.1	12	84	29.0	25.0	24.0—27.5
October	135.4	9	81	29.0	24.5	24.5—28.5
November	155.1	10	80	32.0	23.5	23.0—27.0
December	20.2	2	70	31.0	22.0	23.5—28.5

A The soil temperatures at 5 cm, 15 cm and 30 cm depths were recorded in the forenoon generally between 8.30-10.30 am.

Isolation of the Pathogen

The pathogen, *Phytophthora palmivora* was frequently isolated from soil/tissue samples collected from different pepper gardens from June to December during 1972-79. Positive isolation, though at a lower frequency, was obtained also in January—February from

soil in a garden provided with irrigation (Table 5.3). It is also seen from Table 5.4 that from all parts of the vine showing various symptoms of the disease, the fungus could be isolated. Majority of the isolation was, however, made from soil, stem and root.

Table 5.3. Frequency of isolation of *P. palmivora* from soil/tissues of black pepper from Infected gardens (1972-79).

Month	No. of samples made from soil/tissues	Positive isolation:	Percentage
January	15	1	6.6
February	12	1	8.3
March	10	—	—
April	10	—	—
May	10	—	—
June	29	7	24.1
July	28	14	50.0
August	30	16	53.3
September	22	10	45.4
October	18	7	38.8
November	15	5	33.3
December	15	4	26.6
Total	214	65	30.4

Table 5.4. Different sources from which *P. palmivora* was isolated during 1972-1979 (June-December period)

Locality	Different sources*						Total
	Root	Stem	Twig	Leaf	Spikes	Soil	
Bandadka	5/10	12/20	2/3	4/8	1/4	11/25	35/72
Kuttikol	2/3	—	—	—	—	1/3	3/6
Malom	1/4	2/4	1/3	1/2	2/15	—	7/28
Mercara	—	1/3	—	—	—	—	1/3
Siddapur	—	1/3	—	—	2/6	—	3/9
Thaliparamba	—	3/7	1/2	1/5	—	—	5/14
Thamarasseri	3/6	3/8	—	—	—	1/3	7/17
Wynad	—	2/8	—	—	—	—	2/8
Total	11/23	24/33	4/10	6/15	5/25	13/31	63/157
Percentage	47.8	45.3	40.0	40.0	20.0	41.9	40.1

*Nominator stands for the number of samples from which isolation of the fungus was positive, and denominator stands for the total number of samples tested.

Inoculum at various distances, depths and heights

The amount of inoculum present in 50 g soil sample at various distances from the vines and at various depths of soil was assessed indirectly by the number of castor seeds

infected on incubation. Samples were drawn during April-July. No fungus could be recovered during April, when the soil moisture at various depths ranged from 3.5 to 10.5%. In May, from the soil sample collected at 5 cm depth from the base of the vine, only 5% of the baits were infected. The soil moisture ranged from 11.6-14.5% at various depths. In June, July and August samples, with moisture percentage ranging from 20.6-28.7, the percentages of positive isolation of the fungus at 0 to 5 cm depths from the base of vine ranged from 10-20.

The data presented in Table 5.5 show that the slides kept at 25 cm heights caught maximum number of particles (58) as against a mean of one particle at 75 cm height. The weight of soil particle also was maximum (0.143 g) on the vines at 25 cm height and gradually reduced as the height increased. The percentages of baits infected were 27.5 and 12.5 at 25 cm and 50 cm heights respectively. No infection occurred in soil particles collected at 75 and 100 cm.

Table 5.5. Isolation of *P. palmivora* from soil particles on the vines at different heights

Height (cm.)	No. of soil particles per slide (Mean of 4 replications)	Weight of soil/vine(g) (Mean of 5 replications)	Isolation of <i>Phytophthora</i> (% of baits infected)
25	58	0.143	27.5
50	26	0.139	12.5
75	1	0.062	Nil
100	NR	0.007	Nil

*The soil particles were collected from 5 cm area around the vine, 2.5 cm on either side of the desired height. NR = Not recorded.

DISCUSSION

From the occurrence of the disease, it is observed that the disease spreads from a central point in a centrifugal fashion initially with scattered infections occurring later. From these points again the disease spreads radially. Similar pattern of disease spread was recorded by Harvey (1944) in avocado decline and by Holliday (1961) and Holliday and Mowat (1963) in foot rot disease of pepper. It is also noted that the northern side of the plot (FIG. 5.1) is at the bottom of a hill slope and according to the owner of the garden it was here that the disease outbreak occurred first. Incidentally a foot path passes through the plot where higher concentration of the dead vines exists. Holliday and Mowat (1963) also recorded similar results in Sarawak. Perhaps the labourers moving along the path for various operations in the garden or cattle grazing in the garden might be aiding in the spread of the disease in addition to other natural means of spread.

The pathogen could be isolated in greater frequency from soil, root and stem. The fungus was either undetectable or at its lowest level in summer months when soil moisture was very low and soil temperature high (28.5-32.5°C). The fungus population increased

during rainy months, as judged from the higher frequency of isolation. Holliday and Mowat (1963) reported that during the wettest months of October-March, the incidence of foot rot was more and isolation was more frequent during those months. Marks *et al.* (1975) working on dieback of Jarrah (*Eucalyptus marginata*) caused by *P. cinnamomi* found that low soil temperature limited the fungal population in winter months and when soil temperatures were favourable during summer months, population levels were dependent on soil moisture, increasing with increasing soil moisture. Ecological factors like temperature and soil moisture were found to be limiting factors in the development of disease caused by *P. cinnamomi* (Hine *et al.*, 1964; Chee and Newhook, 1965), *P. palmivora* on 'cacao' (Lellis, 1952, Tarjot, 1972, Dakwa, 1974) and *P. parasitica* var. *piperina* in betelvine (Venkata Rao *et al.*, 1969; Selvaraj *et al.*, 1973). Holliday and Mowat (1963) and Dawka (1974) also found that the incidence of foot rot or black pod respectively occurred more during wettest months and in areas of heavy rainfall. The data presented in Tables 5.1 and 5.2 are in conformity with the above findings. During July-August, the soil temperature dropped to 20-24°C with high relative humidity (>90%) in most of the days. The intensity of premonsoon showers also seems to play an important role in deciding the disease incidence. In years of heavy premonsoon and monsoon showers, with high R. H. and low soil temperature in July-August, the disease occurred in higher intensity.

The general trend was that during wettest months of a year, the fungus could be isolated in higher frequency, and the percentage of successful isolation reduced gradually towards November-December. During summer months, the isolation made was either negligible or nil. Kliejunas and Nagata (1979) reported that population of *P. cinnamomi* was the lowest in winter months when minimum soil temperature was 10°C and increased as soil temperature increased till October up to about 20°C. From an irrigated arecanut garden having pepper as a mixed crop, we isolated the fungus during January-February also, though the percentage of isolation was very meagre. In irrigated arecanut garden, high soil moisture was prevalent and the minimum temperature used to be around 20°C (Nambiar and Sarma 1977). In such gardens root rot symptoms occurred generally, rather than collar rot symptoms (Nambiar and Sarma, 1977).

The fungus could be isolated in higher frequency from soil, basal stem or root. In the case of soil samples, the fungus was present more at the base of the vines than at farther points. Medeiros (1977) found in black pod of cacao that the highest concentration of *P. palmivora* propagules occurred at 20 cm from cacao trunk than at 40 cm or 60 cm distances. At the same site the amount of propagule at 10-12 cm depths was about one-third of the soil surface. In our studies we found that the soil moisture was the highest at 5 cm depths where the infection of baits was also higher. Similar findings have been reported by Flowers and Hendrix (1972) and Gray and Hine (1975) working on *P. parasitica* var. *nicotianae* on tobacco and *P. megasperma* on alfalfa respectively.

Studies on fungal isolation from soil particles sticking on to the vines at different heights above ground indicated that the infection of the bush at the lower region might be due to inoculum carried through rain splash, as suggested by Muller (1936), and Holliday and Mowat (1963). Spread of *Phytophthora* spp. by rain splash was also reported by Pereis (1956) in rubber and Newhall *et al.* (1966) and Okaisabor (1971) in cacao. It is

also possible that the soil particles are deposited by termites. Sarma (unpublished) could get isolation of *P. palmivora* from the soil deposited by termites or ants on standards having disease-affected vines. Turner (1972) could isolate *P. palmivora* from soil deposited on standard by *Crematogaster* ants.

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DISCUSSIONS

P. H. Tsao : The slide showing effect of height on splashing of soil pathogen is very interesting and this type of work is needed for various crops, because the inoculum at different heights play an important role in infection of aerial parts.

R. S. Mehrotra : What do you mean by soil particles?

Answer : Soil fractions of minute size deposited on the slide as a result of splash.

MORPHOLOGY OF BLACK PEPPER PHYTOPHTHORA ISOLATES FROM INDIA

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ABSTRACT

Based on the morphology, *Phytophthora* isolates from foot rot and root rot affected black pepper (*Piper nigrum* L.) are grouped as '*P. palmivora*' 'MF 4' (Morphological form 4). Sporangioophores are sympodial to umbelloid type. Sporangia are caducous and their shape ranges from ellipsoidal, obovoid to pyriform, with long pedicels. About 58.5% of sporangia showed L : R ratio of 2.0—2.5 and 41.4% showed 1.5—1.8. Oospore formation was noticed when *Phytophthora* isolates from rubber and cacao were paired.

INTRODUCTION

Although *Phytophthora* sp. was reported from black pepper from South India as early as in 1926 (Venkata Rao, 1929), a conclusive proof that *Phytophthora* sp. causes pepper wilt in Kerala was reported much later by Samraj and Jose (1966) who adopted Muller's (Muller, 1936) identification for the fungus as *P. palmivora* var *piperis*. Distribution of foot rot of black pepper caused by *P. palmivora* in different pepper growing tracts of the world was reviewed by Nambiar and Sarma (1977). Non-caducous and non-pedicellate sporangial form of *P. palmivora* from black pepper and betel vine (*Piper betle*) was reported from Sarawak (Holliday and Mowat, 1966 and Turner, 1969). Waterhouse (1974) termed *Phytophthora* isolates from black pepper as 'atypical' strains of *P. palmivora*, while Tsao and Tummakate (1977) kept it under MF4 group of '*P. palmivora*'. The taxonomic position of *Phytophthora* isolates from black pepper has been reviewed (Tsao, 1980, Sarma and Nambiar 1980). In view of the lack of information on the morphology of *Phytophthora* isolates from black pepper in India, a preliminary study on this is reported in this paper.

MATERIALS AND METHODS

Phytophthora isolates from black pepper (BP isolates) were raised on carrot agar medium (Griffin, 1977) kept in darkness for 72 hr. at 35°C. Discs (1 cm) cut out from advancing margins of the colony were incubated in Petri's solution taken in petri plates. The plates were exposed to fluorescent tube light for 72 hr. Floating sporulating growth from the margins of the discs was observed directly under the microscope and was also mounted on slide to examine for the sporangiophore morphology. Measurements of

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freely floating sporangia were recorded for 185 sporangia (Isolate PE 1). Colony morphology of the fungus on potato dextrose agar medium was observed. For induction of sex organ formation five BP isolates were raised on oatmeal agar medium in all possible combinations and the paired culture plates were incubated for 20 days in darkness at 25°C. BP isolates were also paired with *Phytophthora* isolates from cocoa and rubber. *Trichoderma* sp. was raised along with BP isolates on oats agar medium to induce sex organ formation.

RESULTS AND DISCUSSION

The colony of the fungus was petalloid with abundant aerial mycelium. Cultures grew very well and sporulated abundantly on carrot agar medium. Sporangiohores were of umbelloid type although sympodial types were also noticed. Sporangia were ellipsoidal with a tapering base, obovoid or even fusiform. Ovoid sporangia were also noticed. The papilla was shallow but clear. Occasionally sporangia with two papillae were observed. The length \times breadth of the sporangia was $24-60 \times 16-34\mu$. About 58.5% of sporangia showed L:B ratio of 2.0-2.5 and 41.4% showed 1.5-1.8. L:B ratios of 2.5 and 1.8 were reported for the sporangia with tapered base and with rounded base respectively for Thailand isolates of black pepper (Tsao and Tummakate, 1977). The isolates in the present study differed from Thailand isolate in that double septate sporangia were not observed so far.

A gentle agitation of the incubation medium containing sporulating discs released pedicellate sporangia (FIG. 5.2) with a pedicel length ranging from 16-180 μ . Isolates differed in the amount of sporangial release on agitation of the medium. Highly caducous nature of sporangia was also reported for the Thailand isolates (Tsao and Tummakate, 1977) as in the present study. In contrast to this, earlier reports of Holliday and Mowat (1963) and Turner (1969) described sporangia of BP isolates from Sarawak as noncaducous without any pedicels. The importance of sporangial caducity and length of the sporangial stalk as taxonomic criteria has been stressed (Tsao, 1977; Kaosiri, Zentmyer and Erwin, 1978). The identification of morphological forms of *P. palmivora* (viz., MF1, MF2, MF3 & MF4) isolates from cacao was reviewed (Griffin, 1977; Brasier and Griffin, 1979). The black pepper isolates from India fit in well into the 'MF4' of *P. palmivora* and this was further confirmed (Tsao, 1980).

The black pepper isolates from Indonesia were referred to *P. capsici* (Kasim, 1978). From a comparative study of BP isolates from different geographical regions, Tsao (1980) opined that the BP isolates differed from *P. capsici* in spite of small similarities and they cannot be keyed to this species. Further, he opined that the correct nomenclature of the black pepper *Phytophthora* isolates might not be possible until the redescription of *P. palmivora* is given the status of a separate species. Sex organ formation in BP isolates was reported in single spore cultures that were stored for 2-3 months (Turner, 1962; Tsao, 1979). However, the present authors could not notice sex organ formation in cultures on prolonged storage or when BP isolates paired in different combinations. But sex organ formation with amphigynous antheridia and oospores were noticed when BP isolates were paired with isolates from either cacao or rubber. No sex organ

formation in *P. palmivora* has been reported (Brasier, 1978). Further studies are in progress to identify the sexual mating types (A1 & A2) among the BP isolates, from different regions in India. Oospore formation was reported when two compatible types of BP isolates inoculated on to pepper leaves and incubated in darkness at 20°C (Brasier, 1969a and 1969b). The authors could not notice oospore formation in the infected leaves, stems or roots of black pepper.

From the present study the BP isolates are identified as 'MF4' of *P. palmivora*.

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DISCUSSIONS

T. N. Sreenivasan : How do you purify the *Phytophthora* culture for pairing?

Answer : By single zoospore isolation.

P. H. Tsao : I will send the culture of MF. 4 type, for comparison if necessary.

R. S. Mehrotra : Since some scientists raised an objection for pairing of isolates from different countries, getting culture from outside the country may be more dangerous.

T. N. Sreenivasan : Dr. Sarma can send his isolates to California, rather than getting the cultures to India for comparison.

THE DISTRIBUTION OF *PHYTOPHTHORA PALMIVORA* (BUTLER) BUTLER IN NORTH KANARA SOILS AND ITS ROLE IN WILT OF PEPPER

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ABSTRACT

A survey was undertaken during the year 1979-80 to find out the distribution of *Phytophthora* in the main pepper growing regions of North Kanara district of Karnataka viz., Sirsi, Siddapur and Yellapur taluks. The soils collected from different parts were evaluated for the population density of *Phytophthora* using castor seeds as baits. The quantitative estimation was done based on the number of infected castor baits per known quantity of the soil. The data on such examinations revealed a direct correlation between the number of baits that trapped *Phytophthora* and the disease incidence in the field. The population density was directly proportional to the intensity of the rainfall. In some gardens there was a direct correlation between incidence of 'Koleroga' of Arecanut and wilt of black pepper. Drainage conditions of the gardens also had a bearing on the incidence of the wilt.

INTRODUCTION

Wilt of black pepper (*Piper nigrum* L.) caused by *Phytophthora palmivora* (Butler) Butler was first reported in India from pepper growing areas of Kerala by Sam Raj and Jose (1966). During July-September of 1978 and 1979 the disease was observed in an epiphytotic form leading to total destruction of plantations in certain instances, in the pepper belt of North Kanara. The disease was characterised by rotting of leaves and wilting of the vines. It was suggested that the spread of infection was mainly through soil and water (Nambiar and Sarma, 1977) and the soil-borne inoculum was of fundamental importance in initiating epiphytotics of the *Phytophthora* diseases (Thorold, 1955; Holliday and Mowat, 1963; Okaisabor, 1971; and Onesirosan, 1971). Further, the relationship of inoculum levels of several soil-borne species of *Phytophthora* and *Pythium* to infection of several hosts has been very well correlated (Mitchell, 1978). Baits like apple (Holliday and Mowat, 1963) and castor seeds (Narasimhan, 1964) were used to detect *P. palmivora* in soils.

The present investigation was undertaken (i) to find out the efficiency of castor baits in quantitative estimation of soil inoculum and (ii) to determine the relationship between the inoculum levels of *P. palmivora* in soils and the incidence of pepper wilt.

MATERIALS AND METHODS

Baiting of *P. palmivora* from soil

Castor seeds (cv. RC-8) free from *Phytophthora* were used in all baiting tests.

Inoculum density and percentage colonization of castor seed

P. palmivora isolate from pepper was grown on sand maize-meal medium for 21 days at $25 \pm 1^\circ\text{C}$. Sterilized soil was mixed with required quantities of inoculum separately, in order to obtain 0.5, 1, 2, 3, 4, 5, 6 and 7 per cent inoculum in soil, on weight basis. The infested soils were taken in Petri plates at the rate of 100 g each. Ten surface sterilized castor seeds were buried in each plate. Ten replications were maintained for each inoculum level. The soil was moistened to 80 per cent of its moisture holding capacity and the plate were incubated for 72 hr at $25 \pm 1^\circ\text{C}$. Baits were collected after the incubation period, washed with sterile water to remove the soil particles, surface sterilized and plated on two per cent agar plates. Observations were taken for the number of baits colonized by *P. palmivora* at each inoculum level after three days and the percentage colonization was calculated.

Analysis of *P. palmivora* in pepper garden soils by castor seed bait colonization

Soil samples of pepper gardens from 40 villages of North Kanara were collected during September-October, 1979. In each village, three gardens were sampled and in each garden, soil samples (500 g each) of top 10 cm layer were collected around ten randomly selected pepper vines. Thirty such soil samples were pooled, powdered and sieved through 0.2 cm sieve. Castor seeds were baited in each soil sample so obtained and the percentage colonization was calculated as mentioned above.

Disease incidence in the gardens

The number of pepper vines wilted, in the gardens, from where the soil samples were collected for the analysis of *Phytophthora*, was recorded and the percentage disease incidence was calculated for each village. In large holdings, 100 vines in randomly selected rows were taken into consideration for the purpose of calculating the percentage disease incidence.

RESULTS AND DISCUSSION

In order to assess the inoculum levels in the field, the percentage colonization of baits obtained at a known inoculum level in the laboratory was made use of, with the object of building suitable model for the purpose. Scatter diagram (FIG. 5.3) of the percentage of baits colonized against inoculum levels, revealed that the relation obtained was linear. Logically, when the inoculum level is zero, no bait should be colonized by *Phytophthora*. Hence, the model in the form $Y = \beta x$ was suitable for the purpose and the linear regression equation for sample was in the form $Y = a + bx$. The analysis has revealed that the value of 'a' is not significantly different from zero. To estimate the parametric value β , various values were given for 'b' within its range of nonsignificance and error variance was calculated for each value. It was found that the error variance was minimum at $b = 15$ with $\alpha = 0$. Hence, the model was constructed in the form of $Y = 15x$. The correlation coefficient of sample values with a linear regression equation $Y = a + bx$ was 0.991%. The population correlation coefficient was found to be 0.9888 with model $Y = 15x$. Further, it was found that the sample regression coefficient was also not significantly different from population regression coefficient. Thus, the models constructed

Table 5.6. Percentages of baits colonized, expected inoculum levels in soils, observed and calculated disease incidence

Sl. No.	Villages	Baits colonized (%)	Expected inoculum levels(x) ($X = .062y$)	Disease incidence in the gardens (%)	Calculated disease incidence(Y) ($Y = 0.61x$)
1	2	3	4	5	6
1.	Chipigi	66	4.30	40	40.26
2.	Bommanahalli	58	3.78	35	35.38
3.	Bhairumbe	16	1.04	10	9.76
4.	Ashisara	50	3.26	30	30.50
5.	Hulgol	83	5.41	50	50.65
6.	Sonda	66	4.30	40	40.26
7.	Kalve	50	3.26	30	30.50
8.	Hulekal	65	4.24	40	39.6
9.	Puttanamane	50	3.26	30	30.50
10.	Salkani	100	6.52	60	61.00
11.	Mundigesara	33	2.15	20	20.13
12.	Golikoppa	41	2.67	25	25.01
13.	Boppanahalli	83	5.41	50	50.63
14.	Kodigebisu	80	5.22	50	48.80
15.	Yellapur	16	1.04	10	9.76
16.	Kangod	83	5.41	50	50.63
17.	Unachigi	33	2.15	20	20.13
18.	Malkop	24	1.56	15	15.61
19.	Alwad	25	1.63	15	15.25
20.	Tatagar	32	2.09	20	19.52
21.	Chikoti	40	2.61	25	24.40
22.	Chavras	25	1.63	15	15.25
23.	Kalache	16	1.04	10	9.76
24.	Sankadagundi	41	2.67	25	25.01
25.	Kampli	24	1.56	15	14.64
26.	Belsi	25	1.63	15	15.25
27.	Kansur	50	3.26	30	30.50
28.	Tyagali	58	3.78	35	35.38
29.	Kodsara	25	1.63	15	15.25
30.	Hutgar	33	2.15	20	20.13
31.	Hegnur	32	2.09	20	19.52
32.	Heggarani	42	2.74	25	25.62
33.	Surkuli	100	6.52	65	61.00
34.	Nelmov	52	3.39	30	31.72
35.	Koppesara	41	2.67	25	25.01
36.	Siddapur	8	0.52	5	4.88
37.	Birgar	8	0.52	5	4.88
38.	Harsikatta	25	1.63	15	15.25
39.	Nilkunda	25	1.63	15	15.25
40.	Dasanakoppa	18	1.17	10	10.98

were: $Y = 15x$ (1) and $X = 0.0652y$ (2) with population correlation coefficient 0.9888.

Using the equation (2), the inoculum levels in the soil samples collected from 40 villages were calculated and presented in Table 5.6. It can be seen from the Table that the percentage colonization of castor seed baits was directly proportional to the inoculum levels and indicated the population levels in the soil. Further, the observations in the Table also indicated a direct relationship between the percentage colonization of baits and the percentage disease incidence. Scatter diagram (FIG. 5.4) of the percentage of baits colonized against disease incidence also revealed a linear relationship. The models constructed after carrying out the statistical analysis were: $Y = 0.61x$ (3) and $X = 1.63y$ (4) with population correlation coefficient 0.9984 (where, Y = percentage of disease incidence and X = percentage of baits colonized). Using the equation (3), the expected disease incidence in the field was calculated based on the percentage of baits colonized and presented in column (6) of Table 5.6. The data indicate that the calculated values for disease incidence are very close to the values for disease incidence observed in the field.

When the inoculum level was below 6 per cent, the baiting technique found its limitation in detecting it. Again, the assessment of disease incidence beyond 60 per cent was also not possible because of total colonization at this level. With these limitations, the castor seed baiting technique could be successfully followed for the quantification of inoculum level in the soil and the disease incidence in the field using the models prepared.

High rate of recovery of *Phytophthora* in soils indicates that the soil is an important source of primary inoculum for the pepper wilt. The infection and subsequent disease development are influenced by many factors, viz. host tolerance or resistance at different growth stages to infection and/or disease, soil moisture and temperature, hydrogen-ion concentration, cation composition, aeration and biotic composition and the inoculum density and form, as well as the virulence of the pathogen (Mitchell, 1978). Under such circumstances, it is necessary to understand the influence of inoculum density on plant infection before attempting to elucidate quantitatively the overall interactions of host, pathogen and environment.

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The authors sincerely acknowledge the help rendered by Mr. M. R. Advani, Assistant Professor of Statistics, College of Agriculture, Dharwad in fitting suitable models and for statistical interpretations.

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DISCUSSIONS

S. Y. Padmanabhan : When was the sample taken? Was the disease incidence recorded simultaneously along with sample, or separately?

Answer : Soil samples were collected in September and disease incidence was observed in October.

Y. R. Sarma : When you did baiting for *Phytophthora*, what was the difference you could observe between diseased and normal vines?

Answer : In a garden with 5% disease, 8% of baits were successful in isolation.

Y. R. Sarma : Could you isolate the pathogen from necrotic areas of affected roots?

Answer : Yes, on specific medium, I could isolate the fungus.

SCREENING OF BLACK PEPPER (*PIPER NIGRUM* L) AND *PIPER* SPP. AGAINST *PHYTOPHTHORA PALMIVORA*

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ABSTRACT

Fortyone cultivars of black pepper and 73 wild *Piper* spp. were screened against *P. palmivora* adopting root dip inoculation technique. Narayakodi, Kalluvalli, Uthirankotta and Balankotta showed low percentage of infection as compared to others. None of the wild *Piper* spp showed resistance.

INTRODUCTION

Foot rot disease of black pepper is one of the limiting factors of black pepper production in all pepper growing tracts of the world. The disease takes a heavy toll when high soil moisture and low temperature prevail and is destructive where heavy rainfall continues over a period of 2-3 months as in Kerala. The nature and distribution of the disease has been reviewed (Nambiar and Sarma, 1977). Identifying the source of resistance becomes imperative for the effective and long term control of the disease. Earlier workers on foot rot of black pepper identified some resistant types. Muller (1936) found an Indonesian cultivar Belantung as resistant. In Malaysia Belantung and Djambi (Indonesian cultivars), and Uthirankotta (Indian cultivar) were found to be resistant (Holliday and Mowat, 1963). *Piper colubrinum* (Albuquerque, 1968 a, b) and *P. guineense* (Anonymous, 1977) were found to be resistant in Brazil and Ghana respectively. *P. aduncum*, *P. scabrum* and *P. trelesanum* were found to be partially resistant (Ruppel and Almeyda, 1965). Pepper cultivar Balankotta, *P. colubrinum* and *P. obliquum* var. *eximum* were found to be resistant (Turner, 1971 and 1973).

Although successful grafting of cultivars on root stocks of wild *Piper* sp. mentioned above was reported (Garner and Beakbane, 1968; Albuquerque, 1968 a, Gaskins and Almeyda, 1968) their field establishment was a failure. Development of longitudinal cracks at the graft union point involving *P. colubrinum* as the root stock and cultivars like Balankotta as scion was reported by Alconero *et al.* (1972). However, successful grafting and establishment of 'Kuching' cultivar on pink form of *P. colubrinum* has been reported (de Waard, 1979).

No work has been done in India on the screening of pepper cultivars or *Piper* spp. for resistance to *P. palmivora* earlier. The present work was taken up to fill in this lacuna.

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MATERIALS AND METHODS

Screening of the cultivars/wild pepper types was done according to the technique of Sarma and Nambiar(1979). Inoculum was prepared as follows. Two discs (15 mm from the fast growing edges of the colony) were inoculated in light at 25°C. Each sporulating mat free from medium was blended in 100 ml of distilled water and made up to 250 ml. About 15 ml of inoculum per cutting was used. The cuttings were raised in polythene bags (16 × 25 cm) filled with potting mixture.

Four-month-old rooted cuttings were removed from the bags, the root system washed and kept in the inoculum for 48 hours. Later they were transplanted back into the soil and the inoculated cuttings were kept at 25°C. Speed of death and percentage of infection based on the number of cuttings wilted and number of cuttings inoculated were recorded 20 days after inoculation. Where the cuttings did not wilt and show partial infection they were rated as 0.5 infection. A minimum of 10 cuttings were maintained for each type and the number was increased depending upon the availability of the cuttings.

RESULTS AND DISCUSSION

Results presented in Table 5.7 indicate that none of the cultivars and wild *Piper* spp. tested in the present study showed any reasonable degree of tolerance. As compared to others, percentage of infection was minimum in Narayakodi (40%), Kalluvally (45%),

Table 5.7. Reaction of cultivars of *Piper nigrum* L. and *Piper* spp. to *Phytophthora palmivora*

S. No.	Cultivar	No. of inoculated plants	No. of infected plants	Percentage of infection
<i>A. Piper nigrum</i>				
1.	Narayakodi	20	8	40.0
2.	Kalluvally	20	9	45.0
3.	Kalluvally PTB	20	18	90.0
4.	Kalluvally I	15	10	66.6
5.	Kalluvally II	15	12	80.0
6.	Kalluvally III	18	15	72.2
7.	Karimunda II	20	18	90.0
8.	Karimunda	15	13	86.6
9.	Uthirankotta	20	11	55.0
10.	Balankotta	20	13.5	67.5
11.	Arasinamoratta	18	16.5	93.8
12.	Arikottanadan	15	13	86.6
13.	Ceylon	20	17	85.0
14.	Karivally	15	13	86.6
15.	Kuthiravally	14	11	78.5
16.	Malligesara	15	13	86.6
17.	Munda	17	14	82.3
18.	Karivilanchi	15	13	86.6
19.	Kaniakadan	13	10	76.9
20.	Kottanadan	16	13	81.2

1	2	3	4	5
21.	Talipparamba II	20	18	90.0
22.	Talipparamba III	20	16	80.0
23.	Talipparamba IV	20	16	80.0
24.	Talipparamba V	20	17	85.0
25.	Nilgiris	15	13	86.6
26.	Shimoga	20	17	85.0
27.	Panniyur I	20	15	75.0
28.	Kumbhakodi	20	18	90.0
29.	Chumala	20	16	80.0
30.	Cheriakaniakadar	15	14	9.33
31.	Perumkodi	15	13	86.6
32.	Perumunda	15	13	86.6
33.	Munai	20	18	90.0
34.	Vally	15	14	93.3
35.	Sullia	20	20	100.0
36.	Munda I	20	18	90.0
37.	Doddigya	20	16	80.0
38.	Cheriakodi	20	18	90.0
39.	Chola	20	17	85.0
40.	Kuthiravally ARS	20	15	75.0
41.	Veluthanamban	20	16	80.0

B. Piper spp. (Accession numbers)

1.	13	10	7	76.9
2.	37	15	13	86.6
3.	42	10	8	80.0
4.	139	15	13	86.6
5.	142	15	13	86.6
6.	145	10	9	90.0
7.	146	10	10	100.0
8.	151	15	12	80.0
9.	156	14	10	71.4
10.	157	18	16	88.8
11.	161	15	13	86.6
12.	168	14	11	78.5
13.	169	15	13	86.6
14.	170	10	8	80.0
15.	172	10	7	70.0
16.	173	15	9	60.0
17.	176	10	8	80.0
18.	177	10	7	70.0
19.	178	14	9	64.2
20.	179	13	9	69.2
21.	180	14	9	64.2
22.	184	20	17	85.0
23.	191	10	9	90.0
24.	193	10	10	100.0
25.	194	13	10	76.9
26.	196	8	8	100.0
27.	197	10	7	70.0

1	2	3	4	5
28.	198	10	7	70.0
29.	199	10	8	80.0
30.	202	15	11	73.3
31.	206	14	12	85.7
32.	109	11	9	81.8
33.	213	12	10	83.3
34.	216	15	11	73.3
35.	219	14	10	71.4
36.	220	10	8	80.0
37.	228	10	8	80.0
38.	232	10	7	70.0
39.	238	10	6	60.0
40.	241	10	8	80.0
41.	246	10	9	90.0
42.	248	15	13	86.6
43.	252	15	11	91.6
44.	264	13	10	76.9
45.	270	10	8	80.0
46.	275	10	7	70.0
47.	281	10	6	30.0
48.	287	15	13	86.6
49.	294	13	10	76.9
50.	325	14	11	78.5
51.	356	13	10	76.9
52.	362	15	11	73.3
53.	367	10	8	80.0
54.	391	10	7	70.0
55.	475	10	6	60.0
56.	431	10	7	70.0
57.	443	10	6	60.0
58.	539	10	8	80.0
59.	663	10	7	70.0
60.	665	14	12	85.7
61.	671	15	13	86.6
62.	679	10	7	70.0
63.	435	10	7	70.0
64.	440	10	8	80.0
65.	453	15	14	93.3
66.	455	15	14	93.3
67.	455	14	13	86.6
68.	495	14	12	85.7
69.	537	10	7	70.0
70.	703	10	6	60.0
71.	735	10	8	80.0
72.	780	10	7	70.0

Uthirankotta (55%) and Balankotta (67.5%). Cultivar Sullia was highly susceptible and succumbed in about a week. Out of the four Indian cultivars tested at Sarawak, Uthirankotta showed high resistance (wilt rating 1.00) followed by Kalluvally and Balankotta (1.13) and Cheriakaniakadan (1.38) (Holliday and Mowat, 1963). However, Alconero *et al.* (1972) found Uthirankotta to be susceptible in Puerto Rico. Turner

(1973) screened four Indian cultivars and found that Balankotta was highly tolerant with a disease rating of 2.7 followed by Cheriakaniakadan (3.3), Kalluvally (3.5) and Uthirankotta (3.6). Kuching, a Malaysian type, recorded maximum incidence with disease rating 4.5. The results of the present study are in general agreement with the findings of Turner (1973) and Holliday and Mowat (1963) in that the cultivars mentioned above gave minimum percentage of infection in the present study also. Variation in the reaction of cultivars of black pepper to *P. palmivora* in different countries might be due to geographical variation in virulence and also due to climatic factors that might alter the host physiology to react differently.

Further screening of open pollinated seedling progenies of cultivars is under progress.

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DISCUSSIONS

S. Y. Padmanabhan : Are you recording the temperature at the time of inoculation? What was the temperature?

Answer : Inoculation was done in an air conditioned room at 20-25°C.

J. Subbaiah : In screening technique you have tried 3 methods, and even in uninoculated control some plants were affected. How do you explain this?

Answer : It was found to be due to *Pythium* infection.

Kuch Tiong Kheng : May I know which method is good for screening of pepper seedlings?

Answer : I have presented all the three methods. Among these stem inoculation appears to be a better method.

EFFECT OF TWO SYSTEMIC FUNGICIDES, ALIETTE AND RIDOMIL ON *PHYTOPHTHORA PALMIVORA* ISOLATE OF *PIPER NIGRUM*

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ABSTRACT

The sensitivity of two systemic fungicides, Ridomil and Aliette to *Phytophthora palmivora* was tested by poison bait technique. Ridomil at 100 and 200 ppm showed 80% and 86% inhibition respectively, in the radial growth of the fungus and complete inhibition at 300 ppm. Aliette showed 22% and 50% inhibition of the fungus at 500 and 1000 ppm respectively and growth was absent at 1500 ppm. The systemic activity of the fungicides was evaluated by measuring the lesion diameter on the inoculated leaves of black pepper pretreated with fungicides. Ridomil-treated leaves showed no lesion at 500 and 1000 ppm whereas Aliette-treated leaves showed inhibition of the lesion development to the extent of 57% and 71% at 500 and 1000 ppm respectively.

INTRODUCTION

Systemic fungicides are becoming increasingly prominent in the field of plant disease control. Though a large number of chemicals were reported to have shown systemic activity against many fungi, chemicals with systemic activity against pythiaceous fungi have been identified only in recent times. (Schwinn *et al.*, 1977; Frossard, 1978; Gullino *et al.*, 1979; Papavizas *et al.*; 1979, Benson, 1980).

Systemic activity of DL methyl N (2, 6 dimethyl phenyl) N (2-methoxy acetyl alaninate) (Ridomil) and Aluminium tris (3-ethyl phosphonate) (Aliette) against *Phytophthora* diseases has been reported recently (Frossard, 1978; Benson, 1980). The primary object of the present study is to study the efficiency of these compounds in controlling foot rot and leaf rot of black pepper caused by *P. palmivora*. Detached leaf technique has been employed for studying systemic activity of these chemicals.

MATERIALS AND METHODS

For the poison bait technique, required amounts of fungicides were dissolved/suspended in 2 ml of sterile water and this was later added to 98 ml molten carrot agar. The medium was poured @ 20 ml per plate. The poured plates were seeded with 4 mm discs of 72-hour-old culture of *Papalmivora* and incubated at laboratory temperature (23-25°C). Five replicates were maintained for each treatment. Colony diameters were recorded after 72 hours of incubation. Ridomil was used at 100-300 ppm concentrations (weight/

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volume) at 100 ppm intervals and Aliette at 500-1500 ppm (weight/volume) with an interval of 500 ppm. Inhibition percentage was calculated over the control.

In another experiment, mature leaves of Panniyur-I black pepper were detached, and the petioles were dipped in water and fungicidal solution taken in 250 ml beakers for the control and treatments respectively. After keeping the leaves for 24 hours, they were washed, blotted and transferred to humid petri plates. These leaves were inoculated by placing a 4 mm culture disc in the centre of the lower surface after making a gentle pin-prick injury. After 48 hours of incubation at 25°C, the lesion diameters were measured and inhibition percentage calculated in comparison with control.

The fungicide-treated and control leaves were extracted separately with 80% ethyl alcohol and the extracts evaporated to dryness. The respective residues were extracted with fresh 80% ethyl alcohol. One ml of the extract (equivalent to 5 g fresh leaf material) was added to 99 ml of carrot agar. Alcohol alone added to carrot agar similarly served as absolute control. The radial growth of the fungus was recorded as mentioned earlier.

The effect of the fungicides on sporulation was tested by placing 72-hour-old culture discs in the respective fungicide solutions for 5 days.

RESULTS AND DISCUSSION

The results given in Table 5.8 indicate that both Ridomil and Aliette are capable of inhibiting the growth of *P. palmivora in vitro*. Ridomil showed 80% and 86% inhibition at 100 and 200 ppm respectively. Aliette showed 22 and 50% inhibition at 500 and 1000 ppm respectively. Thus, Ridomil was more efficient in inhibiting the growth of the fungus than Aliette. Sporulation of the fungus was also inhibited at the above concentrations tested. Lack of *in vitro* inhibitory effect of Ridomil on *P. infestans* was reported although it reduced lesion development and sporulation in treated leaves (Bruck *et al.*, 1980). Ridomil was reported to inhibit the growth and sporulation of *P. cinnamomi* (Benson, 1979) and *P. parasitica* (Staub and Young, 1980).

Table 5.8. Effect of Ridomil and Aliette on growth of *P. palmivora in vitro* and lesion development in inoculated leaves

	Concentration in ppm. (wt./vol.)	Colony diameter mm	Growth inhibition %	Sporulation	Effect on lesion Lesion diameter mm	development Inhibition %
Ridomil	100	10	80	0	0	100
	200	7	86	0	0	100
	300	0	100	0	—	—
Aliette	500	39	22	0	13.6	57.5
	1000	25	50	0	9.3	71.5
	1500	0	100	0	—	—
Control		50		+	32.0	

0 = Absent + = Present.

There was a complete inhibition of lesion development in the leaves treated with Ridomil at 100 and 200 ppm. In Aliette-treated leaves, the lesion development was inhibited by 57% and 71% at 500 and 1000 ppm respectively. Die back of Rhododendron caused by *P. heveae* was effectively controlled when Ridomil and Aliette of 0.96 g/l(a.i.) were used as soil drenches prior to inoculation (Benson, 1980). However Aliette was ineffective in controlling soybean infection caused by *P. megasperma* var. *sojae* race 6. (Lazarovits *et al.*, 1980). In the present study although there was a lesion development in the Aliette treated leaves, further development was arrested after 72 hours indicating the slow action of the fungicide. Lack of growth inhibition of the fungus on the medium incorporated with alcoholic extracts of Aliette-treated leaves in contrast to the total growth inhibition with extracts of Ridomil-treated leaves indicates the efficacy of the latter (Table 5.9). However the time lag in the case of Aliette before it becomes effective in plant

Table 5.9. Effect of fungicide treated and untreated leaf extracts of black pepper on growth of *P. pmlivora*.

Medium	Colony diameter (mm)	
1. Carrot Agar	49	
2. Carrot Agar + Alcohol	46	
3. Carrot Agar + alcoholic extracts of untreated leaves	24	
4. Carrot Agar + alcoholic extracts of leaves treated with	Ridomil at 500 ppm	0
	1000 ppm	0
	Aliette at 500 ppm	24
	1000 ppm	23

systems is yet to be studied in detail. Further studies both in pot culture and field control trials are in progress to test the efficacy of these compounds in controlling foot and root rot of black pepper.

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DISCUSSIONS

F. J. Newhook : Screening of the fungicides under laboratory condition by using detached leaf could be good if the fungicides are translocated upwards. Seedlings should have been used for this study.

Answer : Our idea was to test the systemic effect of the fungicide. The studies with rooted cuttings in pot culture are in progress.

S. K. Bhattacharyya : In the poison bait techniques it has been reported that Ridomil 300 ppm and Aliette 1500 ppm inhibited the growth; but in the detached leaf technique why Aliette was not tested at 1500 ppm ?

Answer : We tested only arbitray levels to start with. Different doses even below 300 ppm in the case of Ridomil and above 1500 ppm in the case of Aliette are being tried and studies are in progress in pot-culture experiments.

Tuesday, 23 September, 1980
2.00 P.M. – 3.15 P.M.

SESSION 6

PLENARY SESSION

Chairman : **Dr. N. M. NAYAR**

Rapporteur : **Dr. K. K. N. NAMBIAR**

PLENARY SESSION

Dr. N. M. Nayar welcomed the delegates and requested the Chairmen of various technical sessions to present their reports on the respective sessions. The reports and recommendations of different sessions are given elsewhere. The reports of sessions I and V were presented by the respective chairmen, Drs. S. Y. Padmanabhan and R. S. Mehrotra, those of sessions II and IV by the respective rapporteurs, Dr. Y. R. Sarma and Mr. Thomas Joseph, and that of session III by Mr. N. Ramachandran, in the absence of both the chairman and rapporteur of that session.

After the presentation of the reports, (*vide* pp 258–264) the Chairman invited the delegates to give their impressions on the conducting of the Workshop, whether the objectives of the Workshop have been fulfilled, how far the delegates have been benefitted by the Workshop, and also to suggest whether future WOPDs, if they are decided to be held, should meet separately or under the auspices of the Committee on Phytophthora of ISPP.

Dr. S. Y. Padmanabhan: The Workshop was very beneficial. With regard to the periodicity of the meeting, it would be better that Phytophthora workers on all crops in India meet under the auspices of Indian Phytopathological Society (IPS) as the Phytophthora Workshop on Tropical Crops alone would be restrictive in nature.

Dr. N. M. Nayar: IPS had declined our suggestions to cosponsor this Workshop.

Dr. R. S. Mehrotra: We should have periodic meetings. In the academic universities one is interested more in research in academic areas, but here we should concentrate more on control aspects.

Dr. B. L. Dutt: The objectives with which the Workshop was organised were achieved. All the papers presented in different sessions rather converge ultimately on control aspects. The younger scientists had the opportunity to meet the senior scientists from overseas, and also from other parts of the country and interact with them.

Prof. F. J. Newhook: The notice given for the Workshop was rather short. However, participating it was the most rewarding experience, though we had a hectic exercise. He was rather a learner than a teacher here. He complimented the CPCRI for the very good organisation of the Workshop.

Dr. P. H. Tsao: He very much appreciated the organisation of the Workshop. He complimented Drs. N. M. Nayar and K. K. N. Nambiar for the best way it was organised. This has given a good opportunity for the interaction between Indian and overseas scientists. In this Workshop we had a crop-oriented approach as opposed to discipline-oriented approach in a symposium. He suggested that all *Phytophthora* species except that from black pepper may be sent to Dr. D. J. Stamps (CMI) for identification: *He himself would*

be only glad to obtain black pepper isolates for identification. This would reduce the confusion also. He also suggested that the next Workshop could be organised in Melbourne in 1983 along with ISPP.

Dr. D. J. Stamps: She agreed with Prof. Tsao's suggestion regarding sending cultures for identification. She requested the workers to give as much information as possible while sending the cultures.

Dr. T.S. Sreenivasan: He observed that there were many younger pathology scientists now, which is a most welcome change. The objectives of the Workshop have been fulfilled. The visitors appreciated the conditions here in which plantation crops are grown and the Indian counterpart also were benefitted by their interaction with overseas scientists. He was happy that there was agreement on the modus of sending cultures. The suggestion of holding the centenary of Bordeaux mixture in 1982 was a good idea. Despite all the advances made in the development of plant protection chemicals, Bordeaux mixture still holds a sway in the control of many fungal diseases especially of plantation crops.

Dr. B. L. Dutt: He agreed with the other delegates about the good conduct of the Workshop. This was possible because it was a small group. He immensely enjoyed the participation in this Workshop. It would be a good idea to hold such workshops once in five years. The field trip was much rewarding and refreshing too. Regarding the centenary celebration of Bordeaux mixture in 1982, the time is too short. This should be taken up on an international basis.

Mr. L. N. Saklani: He agreed with the suggestions given by Dr. deWaard in the morning session on foot rot of black pepper to have a five pronged attack on the problem (*vide* p. 257). He also offered to obtain international assistance for intensifying foot rot research.

Dr. N. M. Nayar: The offer of international help is welcome. To avoid overlapping of research work in the international collaboration programme provision for exchange of visiting scientists might also be included.

In the Chairman's concluding remarks Dr. Nayar thanked all his colleagues in the Institute for the total help given in organizing the Workshop and all the delegates for their participation and making the Workshop a success. He realized that there were limitations on part of the organisers in meeting all the demands of the participants. He wondered whether the criteria used for classification of the genus were as definitive as in angiosperms taxonomy. He felt that studies on epidemiology should be intensified for developing an effective forecasting system.

The following resolution proposed by Prof. F. J. Newhook, Dr. P. W. F. deWaard, Prof. P. H. Tsao and Mr. L. N. Saklani was approved:

"The Workshop recommends that an action plan should be formulated aiming at field control of *Phytophthora* taking into account the following areas of investigations:

1. Pathogen biology and epidemiology;
2. Plant breeding, grafting and selection;
3. Cultural practices;
4. Plant nutrition, and
5. Chemical control.

The Workshop further recommends that national funds, international aids and bilateral assistance for implementing the above programme may be sought.

Dr. M. K. Nair, Joint Director, CPCRI Regional Station, Calicut proposed a vote of thanks.

The plenary session came to a close at 3.15 P.M.

REPORTS AND RECOMMENDATIONS OF DIFFERENT SESSIONS

SESSION I

Chairman : Dr. S. Y. PADMANABHAN

Rapporteur : DR. ROHINI IYER

A comprehensive review of the work done in India on bud rot of coconut palms was presented by Dr. K. Radha and the gaps in knowledge of the disease were highlighted by her. Information is lacking on the dispersal of the pathogen. There is need for further investigation on the microclimate favouring build up of inoculum, infection and progress of the disease. The role of bacteria and the organisms involved in causing the soft rot symptoms requires detailed study. The exact part played by insects in dispersal is not clearly understood.

The second status paper, "Abnormal leaf fall disease of rubber caused by *Phytophthora* spp." by Dr. Radhakrishna Pillai, which was presented by Shri M.K. George, discussed the history of the disease, distribution of the pathogen, symptomatology, loss, biology of the pathogen, epidemiology, varietal reactions and control measures. A new organism viz. *Phytophthora botryosa* has been found associated with the disease in Andaman islands. Extreme care is to be taken to prevent the introduction of this pathogen to other places. Information on the strainal variation within the different species of *Phytophthora* infecting rubber is lacking. Inflorescence pruning followed by plant protection sprays could be tried, but how far the practice is economical, has to be decided. Tree injection methods are to be devised for administering systemic chemicals.

The next paper "Azhukul disease of cardamom" by Drs. R. R. Nair and M. Ramana Menon was presented by Dr. R. R. Nair. Many aspects of this recently identified disease were covered: geographic distribution, symptomatology, incitant, problems encountered in isolating the organisms in pure culture, epidemiology and control measures. The possible involvement of *P. palmivora* also in causing the disease is suggested. The role of nematodes in causing the symptoms requires attention. The mechanism of fungicidal or fungistatic action of neem cake is worth investigating. Liming is a normal practice in cardamom plantations, which might bring about pH changes in the soil that may effect infection by the pathogen. The fact that even seeds harbour the fungus offers a potential threat to the nurseries.

"Late blight of potato" by Dr. B. L. Dutt covered various aspects of the disease occurring in India. The variable nature of the pathogen was highlighted. The plant breeding work in progress to incorporate field resistance to evolve suitable varieties for the plains and hills of India was rational. The difficulty of saturating the entire cultivated area with disease resistant/tolerant varieties was stressed. The occurrence of late blight

in the plains was attributed to infected, cold-stored potato being used as seed material. However, the precise role played by the seed-borne inoculum needs to be critically examined. Though tin(Sn)-containing sprays are effective, their cost is at present prohibitive.

The next paper "*Phytophthora* diseases of citrus" was presented by Prof. H.S. Sohi. The paper dealt with the various symptoms produced by *Phytophthora* on citrus, host range of the fungus and control measures. Morphology of the sporangia particularly caducity and its significance in dispersal of the fungus needed further investigation. Importance of rain splash dispersal in spreading the inoculum from soil to the lower branches of the plant which triggers the epidemic was stressed. This suggested the possibility of preventing epidemics by removing the lower branches of the trees. Influence of scion on root stocks and *vice versa* on the susceptibility of the plants to the fungus needs to be studied. Investigations on the role of mycorrhizae in preventing/containing the disease is another aspect that might be rewarding. The relationship of the age of plants in relation to their susceptibility to *Phytophthora* infection has to be fully understood.

The paper "*Piper betle* wilt caused by *Phytophthora parasitica* var. *piperina* Dastur" by Dr. R. S. Mehrotra reviewed critically the information available with regard to symptoms, causal organism, epidemiology, effect of season on the natural antagonists and control measures. The importance of incubating the culture in light for production of typical sporangia was emphasised. The role of apparently healthy cuttings in disseminating the disease needs thorough investigation.

Dr. T. N. Sreenivasan presented the paper, "Recent studies of cocoa *Phytophthora*". The review elucidated the various aspects of the disease, the different species of *Phytophthora* associated with cocoa and their geographic distribution. The taxonomical position of pathogens involved was highlighted.

The review on "Koleroga of arecanut" by Dr. M. Koti Reddy and Mr. M. Anandaraj was presented by Dr. Reddy. The review traced the history, geographic distribution, incitant and control measures adopted in India. The need for studying the epidemiology of the disease in greater detail was emphasised. The source of primary inoculum and the environmental factors favouring the diseases are to be studied to evolve forecasting systems which could be linked with effective control measures.

SESSION II

Chairman : DR. D. N. SRIVASTAVA
Rapporteur : DR. Y. R. SARMA

In this session on "Epidemiology and Forecasting" two lead papers and six research papers were presented.

In his introductory remarks the chairman pointed out that in order to get the maximum benefit from such symposia, it is advisable to ensure in depth discussion on specific aspects of a given disease by those who have made substantial contribution on the disease in question. In the so called "nominal group technique" lately adopted for some seminars in the USA and elsewhere, a few experienced pathologists not involved in the specific disease under discussion are also invited in the discussion. This approach has been found to be very effective in precisely identifying the specific and important research gaps for future investigation.

The lead paper by Prof. F. J. Newhook on "Epidemiology in the genus *Phytophthora*" dealt with the soil borne *P. cinnamomi* infecting *Pinus radiata* and *Eucalyptus* in relation to host, pathogen, and environment interaction. The paper highlighted the soil moisture and temperature as the major factors that governed the disease development. The water economy of the plant depended on the ratio of root loss to root regeneration. The response of the infected host plant to phosphorus was of considerable interest in that the latter boosted up the growth of the tree by ensuring a greater root regeneration and an abundant mycorrhizal development.

The lead paper by Dr. D. H. Lapwood on the "Field studies on the late blight of potato" highlighted the critical meteorological factors and the sources of inoculum that governed disease development and suggested that a critical study of these factors in a given locality could help in efficient forecasting of the epidemics of potato late blight.

Dr. S. K. Bhattacharyya in his paper on forecasting of potato blight in Simla, Shillong and Ootacamund in India brought out the fact that 7 days precipitation with mean temperature of 23°C or less for 7 continuous days would result in blight appearance in about 3 weeks. If temperature remains between 10-20°C, associated with RH of 80% or more for continuously 18 hrs. for 2 consecutive days, the blight would appear in a week. This information is relayed on to farmers through the All India Radio.

"Occurrence of betelvine wilt in Andhra Pradesh" was presented by Prof. J. Subbaiyya highlighting the enormous losses incurred by the farmers. The source of inoculum in such an endemic area was not understood. However the possibilities of *Phytophthora* of castor infecting betelvine cannot be ruled out. It was suggested that sandy soil be re-

commended for fresh plantation and in clayey soils better drainage be provided. An action programme based on previous experience was presented and the same was approved.

Mr. Narula's paper showed the presence of A¹ mating type in *P. colocasiae*. Mr. Jain in his paper on the nutritional aspects of *P. nicotianae* var. *nicotianae* pointed out that the thiamine and biotin boosted growth and reproduction of the fungus. Prof. Newhook's paper on *P. colocasiae* in Solomon Islands showed that increasing the density of the plant/ha in high leaf blight areas still gave increased yield. His paper on the sources of *P. palmivora* inoculum in cocoa in Solomon Islands showed that stem cankers associated with flower cushion act as continuing source of inoculum and infection of maiden plantations can result from inoculum brought in by rats and harvesting tools. Interception of falling leaflets of the shade tree *Leucaena leucocephala* increased inoculum holding frequency of cocoa canopy leaves.

The Chairman in his concluding remarks emphasised the relative importance of the environmental factors which can be highly valuable in development of disease epidemics. He stressed the need for critical studies on the pest and pathogenic responses to the environmental factors for evolving practical and effective management programmes to reduce disease losses.

SESSION III

Chairman : PROF. H. S. SOHI
Rapporteur : DR. P. CHIDAMBARAM

In this session four lead papers and two research papers were presented. Dr. D. J. Stamps spoke on the "Taxonomy of *Phytophthora*" highlighting the important taxonomic characters of both asexual and sexual phases of the fungus in identification of species followed in CMI. Prof. P. H. Tsao presented a lead paper on "The morphology and ontogeny of the black pepper identity of black pepper *Phytophthora* isolates" based on the sporangial morphology and isolates from different parts of the world. From the data gathered so far he felt that black pepper isolates are entirely different from *P. palmivora* and until further investigation he opined to keep them in MF4 of *P. palmivora*.

Dr. B. Boccas presented a paper on "Genetics of the genus *Phytophthora*" outlining the homothallism and heterothallism of the genus. He said that very little is known on the genetical regulation of the mating types but it seems that this could be determined by a complex system of genes based on his studies. Dr. B. L. Dutt in his paper on "Physiological specialisation in the genus *Phytophthora*" explained the criteria in race determination in *P. infestans*. He emphasised the need for further studies on the mechanisms involved in the differentiation and also the germination of oospores resulted from the crosses of different mating types.

Dr. J. T. Dakwa presented a paper on "Effect of source of isolates on the virulence of *P. palmivora*" Dr. S. K. Bhattacharyya in the paper on "Races of *Phytophthora infestans*" mentioned that 74 races have been recorded and that more and more complex races developed when complex R-gene genotypes are employed. However, in the plains only simple races were noticed and might be due to cultivation of susceptible cultivars.

The following recommendations were made out of the discussion:

1) A number of scientists are at present involved on various *Phytophthora* diseases infecting different crop plants. There is a need to have a collection of all these at one place. Subsequently these may be studied critically for proper identification of the species.

2) All the research workers engaged on *Phytophthora* diseases in India may be required to record data on various taxonomic characters in different species on a standard proforma which can be prepared in consultation with Dr. Stamps and Dr. Tsao so as to remove the confusion in the identification of species.

SESSION IV

Chairman: DR. C. S. VENKATRAM
Rapporteur: MR. THOMAS JOSEPH

At the outset, the chairman emphasised that the growers require useful control measures rather than scientific information. So he exhorted the participants to adopt an attitude in this direction.

Summing up the discussions on the paper "Agronomic problems of Cocoa *Phytophthora* pod rot control" by Dr. A. A. Adebayo, he observed the desirability of having judicious selection of the kind of shade.

Regarding Mr. S. Kularatne's paper, "Screening cocoa cultivars for black pod resistance" it was felt that the practice of wounding before inoculation had to be continued.

A thorough examination of the paper "Chemical control of *Phytophthora* diseases of citrus" by Mr. Sawant revealed the need to have fresh check up on the etiology of the disease as *Phytophthora* appeared to be only one of the incitants. Possibility of the involvement of root stocks in carrying over the pathogen also has to be looked into.

Estimated loss of 11 % reported in the paper "Evaluation of germplasm and chemical control of *P. parasitica* var. *nicotianae* on tobacco" by Drs. K. Nagaraj and T. S. N. Reddy appears low.

"Defoliation to improve yield" was a fascinating new information contained in "Studies on the taro leaf blight fungus in Solomon Islands: control by fungicides and spacing," by Prof. Newhook.

The papers presented in the Session on control contained valuable information and the discussions were useful. It was felt that it was high time to have a symposium on the most popular and reliable fungicide Bordeaux mixture as exhaustive sessions on pathogens and diseases have been on for long.

SESSION V

Chairman : DR. R. S. MEHROTRA
Rapporteur: DR. R. NAIDU

The Session was entirely devoted to 'foot rot' of black pepper. Dr. Y. R. Sarma in his presentation of status paper gave a comprehensive account of the disease history, distribution, crop losses, symptomatology, etiology, taxonomy, biology, toxins involved, epidemiological aspects, and control measures. Dr. K. K. N. Nambiar presented a paper on "Some aspects of epidemiology of foot rot disease." It was shown that the disease spreads in a centrifugal fashion and that disease incidence was more in years of heavy rainfall during south-west monsoon. There was a direct correlation between soil temperature and disease incidence. The paper on morphology of *Phytophthora palmivora* on black pepper from India showed that the fungus can be grouped as *P. palmivora* MF 4 type, with minor variations like absence of double septate sporangia.

Paper on the distribution of *Phytophthora palmivora* in the soils of North Kanara, was presented by Mr. M. N. L. Sastry. The study included investigation on the population density of the pathogen in different localities and its relation to incidence of the disease.

An account of methods employed for screening of black pepper for resistance against foot rot was given by Dr. Sarma. Stem inoculation method could be used which is more convenient than root inoculation method.

Mr. N. Ramachandran presented a paper on evaluation of two systemic fungicides namely, Ridomil and Aliette, against the foot rot pathogen under laboratory conditions. He showed that Ridomil was more effective than Aliette while both fungicides inhibited the growth and sporulation of the pathogen and showed systemic activity.

Subsequent to the presentation of these papers a discussion was held, on future programme of work on foot rot, and the following suggestions were made:

- 1) Studies may be initiated for early detection of the infection.
- 2) More emphasis may be given to the taxonomy of the pathogen to avoid confusion.
- 3) Duplication of the work may be avoided through coordinated approach at international level.
- 4) To make cheap and effective control measures against this disease research on pathological side, breeding and selection of resistant varieties, cultural practices, judicious use of fertilizers and chemical control through systemic fungicides may be initiated.
- 5) Funds for conducting research on this disease may be obtained through international, national and bilateral aids.

Comments by Prof. F. J. NEWHOOK*

As a postscript to the successful Workshop just concluded I would like to offer comments on a few major principles that apply especially to more than one of the disease syndromes that received individual attention at the Workshop. The list is far from exhaustive and other delegates could obviously extend the coverage. Inevitably, because of my personal interests there is a bias towards soil-borne examples.

1. I cannot re-emphasise too strongly the point I made in discussion during the session on black pepper foot rot, illustrated earlier with my slide of an outwardly healthy *Eucalyptus* sapling that was almost devoid of healthy roots. The onset of first visible symptoms may be months or even years later than actual commencement of root or collar rot. Not only are secondary invaders likely to be dominant at the earliest visible symptom stage, but treatment then with Bordeaux paste or other fungicides is futile. Plants can compensate for loss of water uptake by such mechanisms as stomatal closure and leaf abscission and by drawing on reserves. It has been demonstrated that some degree of water uptake through dead roots is possible. Visible stress often with 'sudden death', frequently represents the irreversible crossing of a threshold situation, especially in the case of collar or foot rots and of root rots that have progressed into major rots. One can cite analogous situations with other woody perennials: apple, citrus, avocado, *Pinus*, *Eucalyptus* etc. Attempts to determine by eye the extent and distribution of infection are thus likely to be grossly inadequate. More meaningful would be surveys based on baiting of soil samples collected systematically in a garden. Each sample could be say, a well-mixed composite of five soil auger cores from each tree in an area of interest or on sampling transects.

In many cases I believe that there might be no practicable alternative but to apply preventive treatment to every plant in a plantation that is known to be infested with *Phytophthora*. The cost of regular periodic treatment will be negligible compared with the loss of production that would otherwise result. Growers the world over dislike paying more for cost of production. Thus, an intensive education programme might be needed and perhaps also the application of subsidies or a loan scheme with repayment through marketing organisations.

2. Passive movement of *Phytophthora* propagules in even small quantities of soil occurs only too readily, on human feet, cultivating tools, vehicles, domestic and wild animals etc. Thus, once a garden has an initial infestation there must be an expectation of steady increase in incidence, often on a seemingly random basis. In this context, once again, there may be no practical alternative to treatment of all plants in newly infested gardens.

* This comment was sent to us by Prof. Newhook after the Workshop.

3. There are well-authenticated cases of an 'age effect' with hosts susceptible to *Phytophthora* and other pathogens: symptom expression with apples subjected to *P. cactorum* root and collar rot frequently coincides with onset of crop maturity; *Pinus radiata* tolerates *P. cinnamomi* root rot from the late seedling stage until the transition from conical growth form to rounded crown at age about 20 years-the same stage when physiological changes make it cease to be susceptible to *Dothistroma pini* needle blight. We heard several references at the Workshop to black pepper not displaying symptoms of root and foot rot until age 3-5 years, when cropping becomes substantial. Amongst other implications, this factor should be considered when screening for resistance.

4. Some comments on soil assays for *Phytophthora* populations:

- (a) There will always be room for improvement in sensitivity of methods of detection.
- (b) A second attempt will often transform a negative into a positive recording from a single soil sample.
- (c) Reports of seasonal fluctuations in detectable population levels in soil indicate that caution is needed in interpretation of results. We need to know more, amongst other things, about the nature of perennating propagules, dormancy, soil fungistasis, factors influencing multiplication, decline of the pathogen after disappearance of host roots etc.

5. The balance between rootlet death and rootlet replacement is crucial in determining whether there is tolerance or recovery on the one hand, and decline and death on the other. Attention should, therefore, be paid to agronomic factors that have the potential of encouraging root growth, e.g., fertilizer, irrigation and drainage practices. Advice should be sought on depth of drains needed to provide effective control of the water table right to the midway line between drains. Sloping terrain by itself does not ensure good internal drainage. The inherently good drainage property of sandy soil may be neutralised by an unbroken hard pan, by over-irrigation or by prolonged rainfall.

6. Indirect methods of control should be sought actively, especially where financial and social constraints are important. Here I include biological control and reduction of the impact of disease through modification of crop management practices. The success obtained with close spacing of *Colocasia* is an example of what can be achieved.

7. Use of new systemic fungicides needs a new approach to screening. *In vitro* trials have limited relevance and may even be misleading. There is no alternative to use of actively growing host plants.

STUDY TOUR AT CALICUT

There was a day-long study tour on 22 September, 1980 during which delegates were taken to different estates in the verdant green Wynad area in the Western Ghats in Kerala where they could see crops like cardamom, black pepper, arecanut, cocoa and rubber. This gave them an opportunity to observe the diseases occurring on these crops.

The delegates left Calicut at 8 O'clock in the morning and after riding through the winding ghat road of Wynad, they visited Meenakshi Vilas Estate, Kalladi, 80 Km from Calicut, where they saw capsule rot disease of cardamom. In the afternoon the delegates visited Koyappathodi Rubber Estate, Adivaram, to see the abnormal leaf fall of rubber in blocks left without plant protection measures. Later in the evening they visited the Amalgamated Estates, Pudupady, where they could see *Koleroga* and bud rot of arecanut, foot rot of pepper and pod rot of cocoa. The delegates returned to Calicut at night with the satisfaction of seeing not only one of the most beautiful regions in India, but also of getting an opportunity to study *Phytophthora* diseases in the field on the important plantation crops of Kerala. This had helped the scientist delegates especially from North India and abroad, in no small a measure, in the discussions that ensued on the next day.

ABSTRACTS OF PAPERS NOT PRESENTED

A NEW FRUIT ROT OF POMEGRANATE CAUSED BY *PHYTOPHTHORA NICOTIANAE* B. DE HAAN VAR. *NICOTIANAE* A₂ AND ITS CONTROL

ASHOK MISHRA, ASHA SHIVPURI, AND R. N. MAHLA
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A hitherto unreported fruit rot of pomegranate (*Punica granatum* L.) caused by *Phytophthora nicotianae* B. de Haan var. *nicotianae* (A₂) was observed in August-September 1978 at Udaipur. In natural condition the rotting started from the blossom-end and within 48 hours the entire fruit was covered resulting into soft rot. On inoculation the pathogen infected injured fruits of orange, chilli and brinjal. During 1979, nine fungicides were evaluated by dipping pomegranate fruits in fungicide solution for 10 mts. Dithane M-45 as preinoculation treatment completely checked the disease.

**CONTROL OF FOOT ROT DISEASE OF BETELVINE (*PIPER BETLE* L.)
IN MADHYA PRADESH, INDIA.**

CHAURASIA, S. C.
Government Post-Graduate College, Tikamgarh, India.

Field experiments were conducted by random method at Laundi (Chhatarpur), Madhya Pradesh to control *Phytophthora* foot-rot disease of Pan (*Piper betle* L.). Streptonex was used for dipping the cuttings before planting and Bordeaux mixture was employed for spraying the growing plants and for drenching the soil.

The seven different types of treatments comprised Streptonex and Bordeaux mixture applied either individually or in combination. Although all the treatments proved very effective in reducing the foot-rot disease incidence, the best results were obtained when both Streptonex as well as Bordeaux mixture were used in the treatment. Streptonex treatment not only reduced the foot-rot incidence but also increased the establishment of cuttings and growth of the betel vine plants.

OCCURRENCE OF PHYTOPHTHORA ON CITRUS IN INDIA

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In literature, nine species of *Phytophthora* have been found associated with various disease symptoms on *Citrus* species, in various citrus growing regions of the world. In

the present study, a survey was conducted to find out the identity and relative importance of species involved in India, particularly in the citrus growing regions of central and peninsular India. In the survey it was found that only two species viz. *P. nicotianae* var. *parasitica* and *P. palmivora* infect the *Citrus* spp. in India. The occurrence of *P. citrophthora* which is also reported from India, could not be found. Therefore, it appears that the previous records of this species from India need further confirmation. *P. nicotianae* var. *parasitica* was found in relatively warmer and drier areas of Maharashtra and Andhra Pradesh and chiefly associated with leaf fall symptoms and also responsible for foot rot, whereas *P. palmivora* is prevalent in relatively more humid areas of Maharashtra and Karnataka. It has been determined to be responsible for foot rot and gummosis and is particularly severe under conditions of high soil moisture. Taxonomic characters, distribution, etc. of all the species of *Phytophthora* have been presented for proper appraisal.

A FORECASTING SYSTEM FOR USE IN THE CHEMICAL CONTROL OF PHYTOPHTHORA LEAF FALL ON PLANTATION RUBBER IN MALAYSIA

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In Malaysia, *Phytophthora* leaf fall and pod rot caused by *Phytophthora botryosa* first occurred on an epidemic scale in 1966. Since then, annual outbreaks of the disease regularly occurred, its virulence increasing on susceptible clones planted in the North-western and North-eastern regions of the country. Severity of the leaf fall, however, varied markedly from year to year, this being largely determined by the prevailing local weather.

In a detailed study on weather conditions associated with *Phytophthora* outbreaks on three different plantations from 1977-80, a method for short-range forecasting of the annual outbreak of the leaf fall was successfully developed.

Prediction of a first outbreak of the disease is based on the occurrence of a continuous 4-day period during which the daily rainfall is 2.5 mm or more, accompanied by RH > 90% persisting for 14 hours and the maximum temperature falling to 32.2°C or less. Its reliability was established in fungicide controlled spraying/fogging trials when a better disease control was achieved, with less fungicide being required in the annual pre-monsoon prophylactic treatment timed in accordance with the newly formulated simple, weather rule.

SAPROPHYTIC SURVIVAL AND MATING TYPES OF *PHYTOPHTHORA*
NICOTIANAE VAR. *PARASITICA* CAUSING FRUIT ROT OF GUAVA

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Phytophthora nicotianae var. *parasitica* causes severe fruit rot of guava (*Psidium guajava*) in Punjab, Haryana and adjoining areas in Uttar Pradesh. Saprophytic survival of the pathogen was studied in naturally infected guava orchard soil, artificially infested soil and in dead host tissues. Results revealed that in all cases the pathogen was able to survive for more than a year. It survives for fourteen months in naturally infected guava orchard soil and in artificially infested soil and for 16 months in dead host tissues. Studies on mating types of the pathogen was also made. Both A₁ and A₂ compatibility types of the pathogen are found in Kurukshetra and adjoining areas. However, oospores of the pathogen have not been found in nature so far.

AGRONOMICAL PRACTICES THAT MAY REDUCE THE DAMAGE OF FOOT
ROT DISEASE ON BLACK PEPPER

PASRIL WAHID AND R. ZAUBIN

Lembaga Penelitian Tanaman Industri, Bogor, Indonesia

The development of *Phytophthora* foot rot disease on pepper is closely related to environmental conditions. Physical environmental conditions that promote the growth of the vine also stimulate the spread of the disease. However, the latter could be reduced by the application of cultural practices.

A field observation was carried out to study the effect of different management and cultural practices on the incidence of the disease. It included planting vine on mound or hole, time and methods of weeding, provision of isolation canal including blind canal, composition and method of fertilizer application, pruning of live post and pepper vine. The results showed that some of these combinations reduced the incidence of foot rot disease on pepper.

STUDIES ON FOOT ROT DISEASE OF PEPPER IN INDONESIA

RUSLI KASIM

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The most serious disease of pepper in Indonesia is foot rot. Although, this disease has been known to exist for more than fifty years, it has become serious only recently. Loss in crop due to the disease in Lampung Province has been estimated to be about 50%.

Muller (1936) was the first to observe the foot rot disease in Indonesia. After that no serious effort has been made to carry out a research programme until the Pepper Research Stations in Lampung and Bangka were established in 1973.

According to Muller, foot rot was caused by *Phytophthora palmivora* var. *piperis*. This disease spread through soil and water. Inoculation on mature vines resulted in wilt and death after 4-8 weeks. Muller also reported that pepper varieties Belantung was more tolerant to the disease.

Recent studies indicated that the fungus grows rapidly on Oat Meal Agar medium at 27.5-30°C. There was growth at 37.5°C; maximum sporulation was at pH 6-7; no sporulation at pH 3 and 9.

Inoculation of soil with soil-meal-inoculum and mycelial suspension produced foot rot symptoms on single cuttings after 4 and 6 days respectively.

Pathogenicity test by using infected leaves as source of disease for 20 varieties of pepper showed that none of the varieties were affected. Fungicide screening trials carried out in Lampung showed that fungicide Aliette gave promising results.

SURVIVAL OF *PHYTOPHTHORA PALMIVORA* CAUSING LEAF BLIGHT OF *COLOCASIA ESCULENTA* IN SOIL

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Umabala and Ramarao (1972) reported a non-oosporic isolate of *Phytophthora palmivora* as the causal agent of leaf blight of *Colocasia esculenta* from Hyderabad.

From the soil survival tests, it was observed that the present isolate of *P. palmivora* survived for a longer duration (60 days) in natural soil than in autoclaved soil (30 days).

It is known that soil-borne pathogens can survive presumably as chlamydo-spores for longer periods in soils in the absence of host plants. In the present work, chlamydo-spore germination of *P. palmivora* has been studied in two different soils, employing agar slide method (Chinn, 1953). No germination was observed in autoclaved soils; germination was fairly good in natural soils. Significant improvement was observed when 1% glucose in water was added to the soils. Germination occurred even in autoclaved soils after glucose amendment.

ROLE OF PHENOLICS IN DISEASE RESISTANCE WITH SPECIAL REFERENCE TO BETEL VINE PHYTOPHTHORA

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Phenolics are known to offer a varying degree of resistance in host plants against various fungal diseases. With a view to ascertaining the role of phenolics in *Phytophthora* incidence in betel vine, phenol analysis of healthy and *Phytophthora* infected pan leaves was carried out under various experimental conditions. In general the least susceptible 'Madrasi variety' was found to possess maximum amount of total phenols, whereas the most susceptible 'Kapoori' pan leaves contained the least amount of phenolics. Comparatively the least virulent *Phytophthora* isolates from Saugar caused maximum induction of phenolics after infection in the least susceptible 'Madrasi' variety. However, on the whole there appeared a gradual reduction in phenol contents of pan leaves in almost all the cases following infection by all the three differentially pathogenic isolates. Interestingly, compatible system showed a more rapid decline in phenol contents as compared to incompatible systems. As such, rate of phenol metabolism appeared to be actively involved in the betel vine *Phytophthora* pathogenesis.

FIGURES

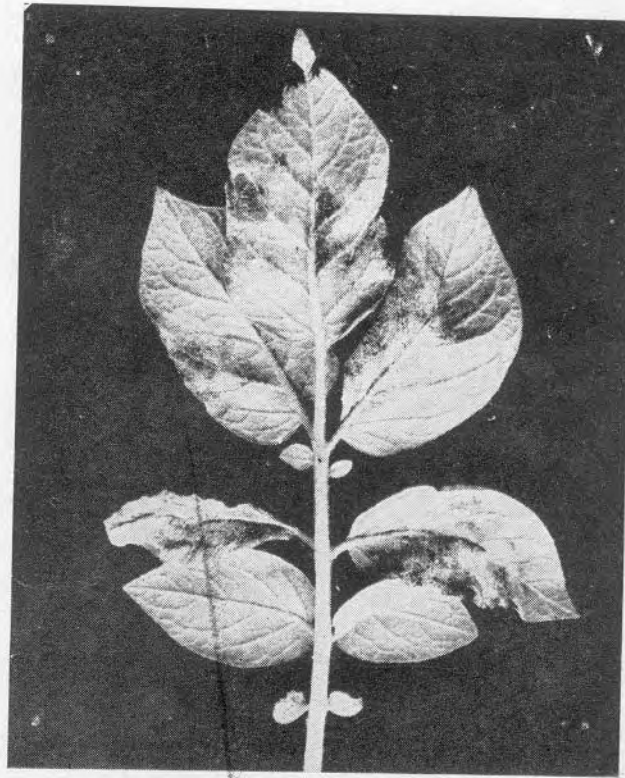


Fig. 1.2 Late blight of potato—foliar symptoms

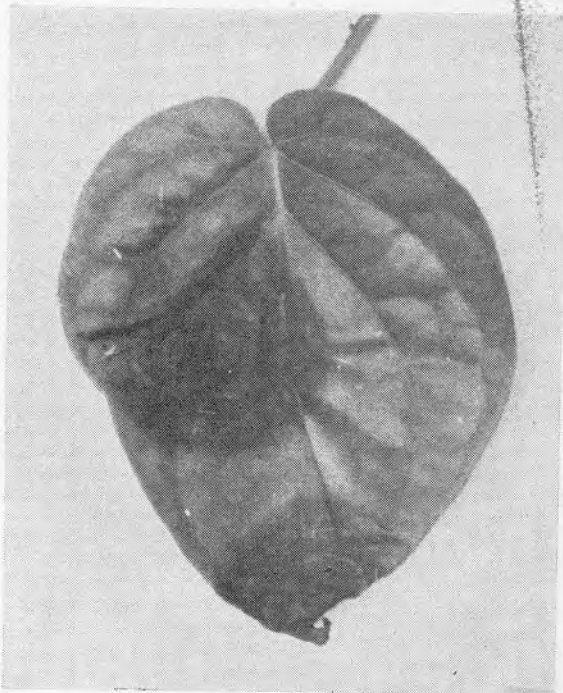


Fig. 1.3 Leaf rot symptoms of *Piper betle* wilt

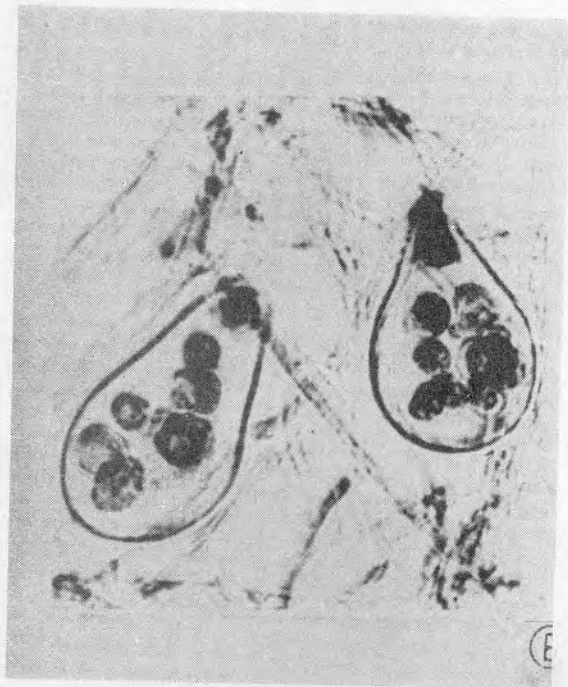


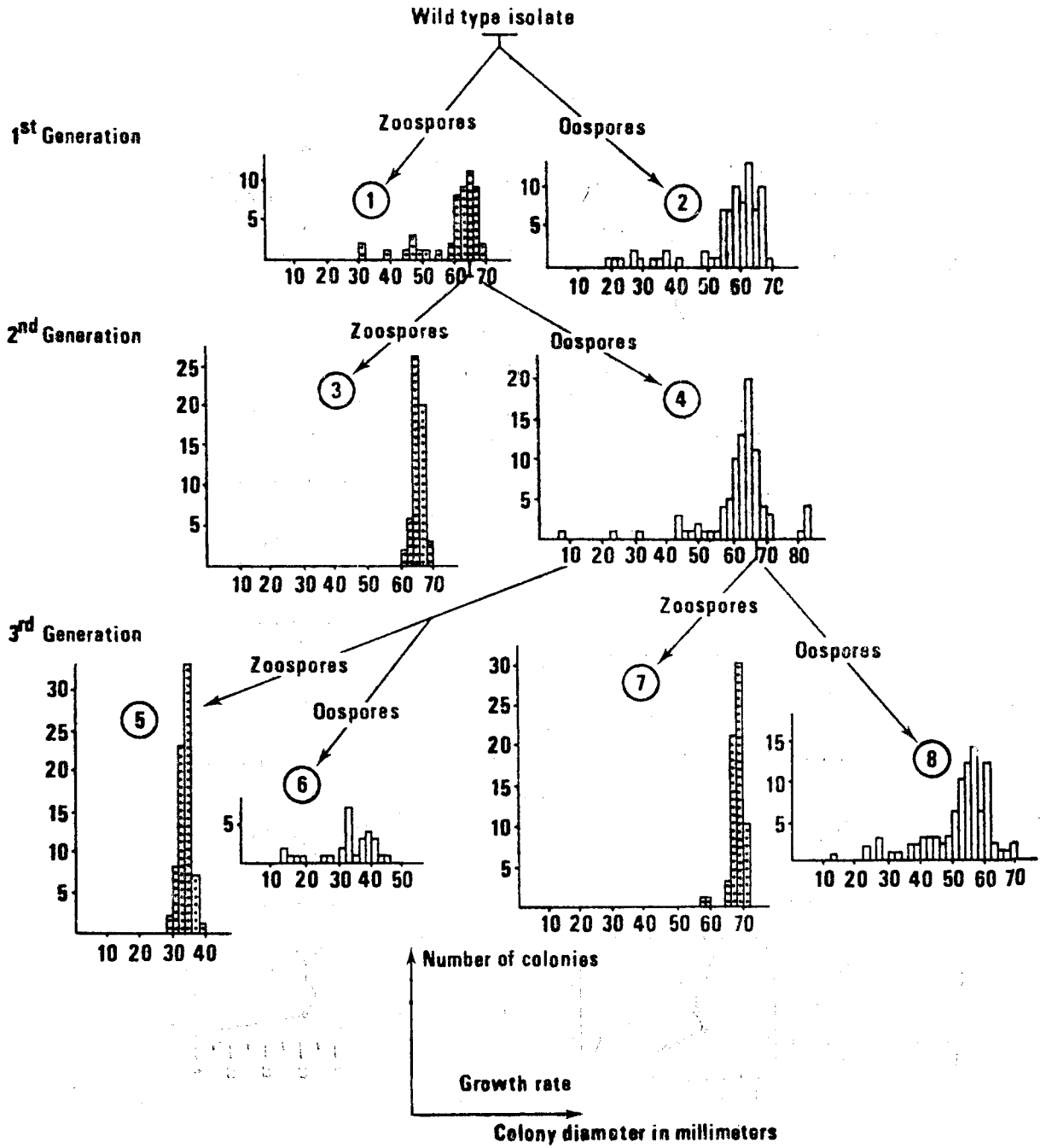
Fig. 1.3a Sporangia with zoospores of *P. parasitica*

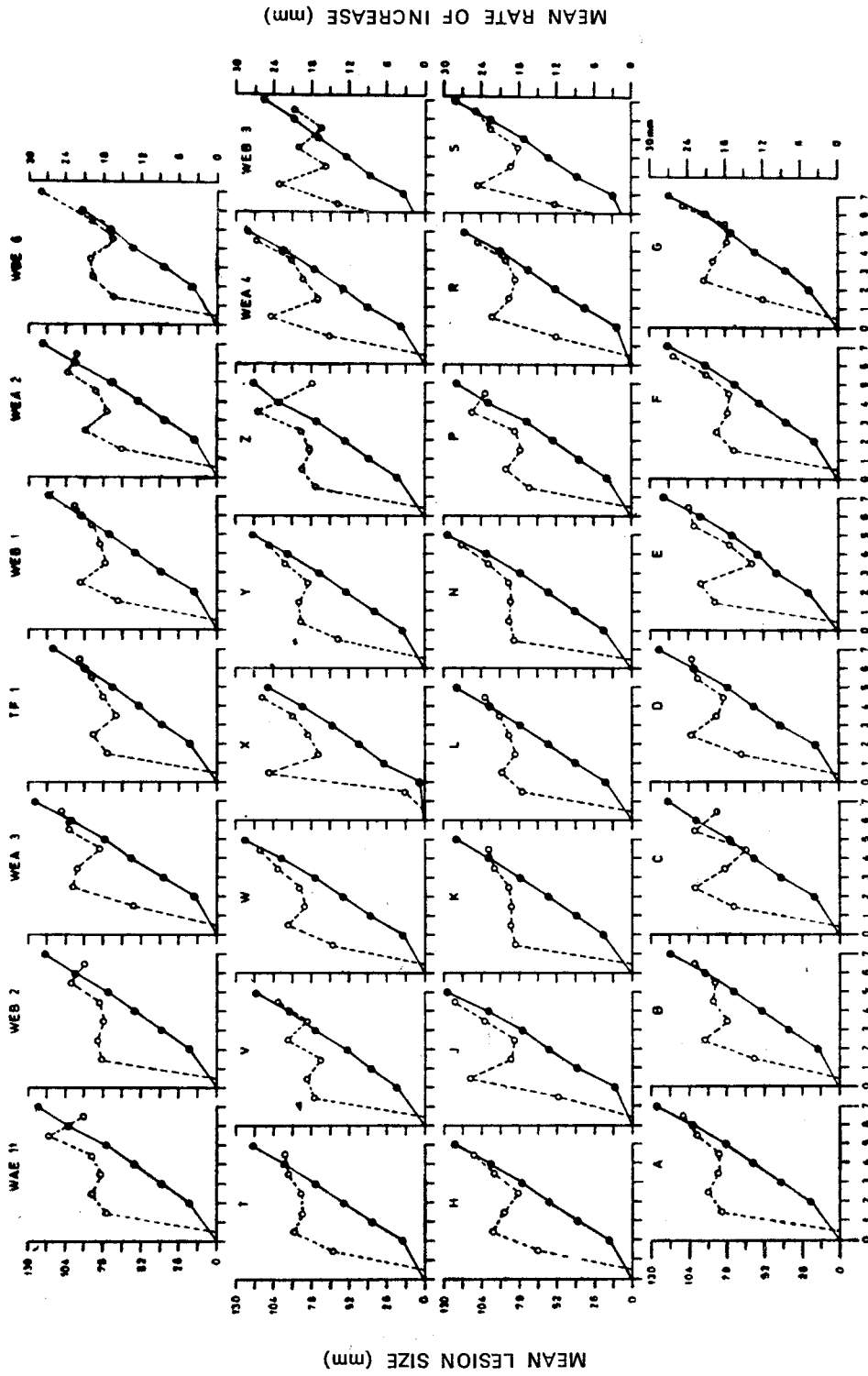


Fig. 1.1 Bud rot of coconut

Fig. 3.1 PHYTOPHTHORA SYRINGAE

Variation in growth rate among sexual and asexual progeny





DAYS AFTER INOCULATION

Fig. 3.2 Variations in the pattern of black pod lesion development by *P. palmivora* isolates collected from 30 cocoa progenies
 (●—● cumulative, ○---○ rate of increase)

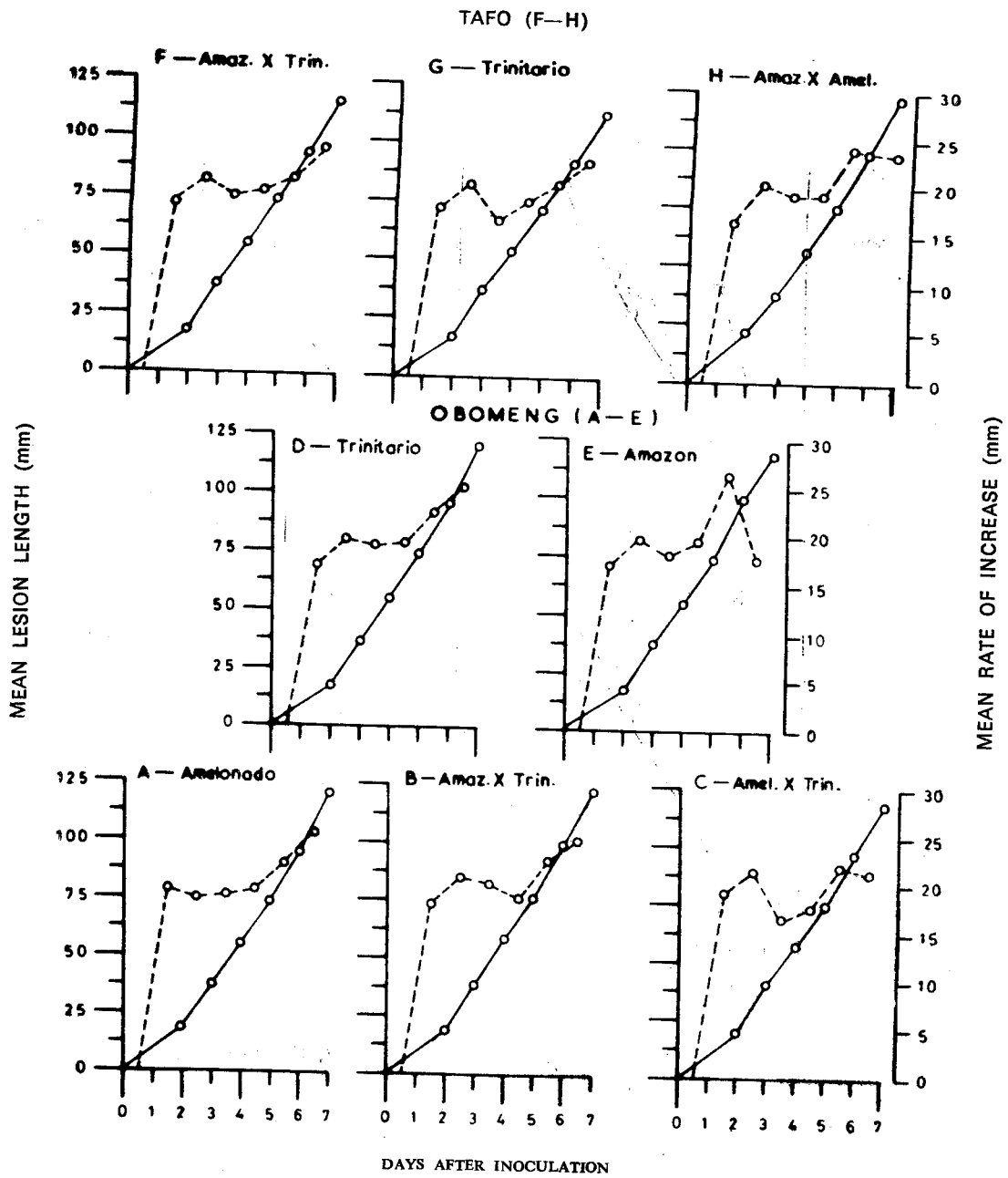


Fig. 3.3 Pattern of black pod lesion development by *P. palmivora* isolates collected from different cocoa types at Obomeng (A—E) and Tafo (F—H) ○—○ Mean lesion length, ○—○ Mean rate of increase

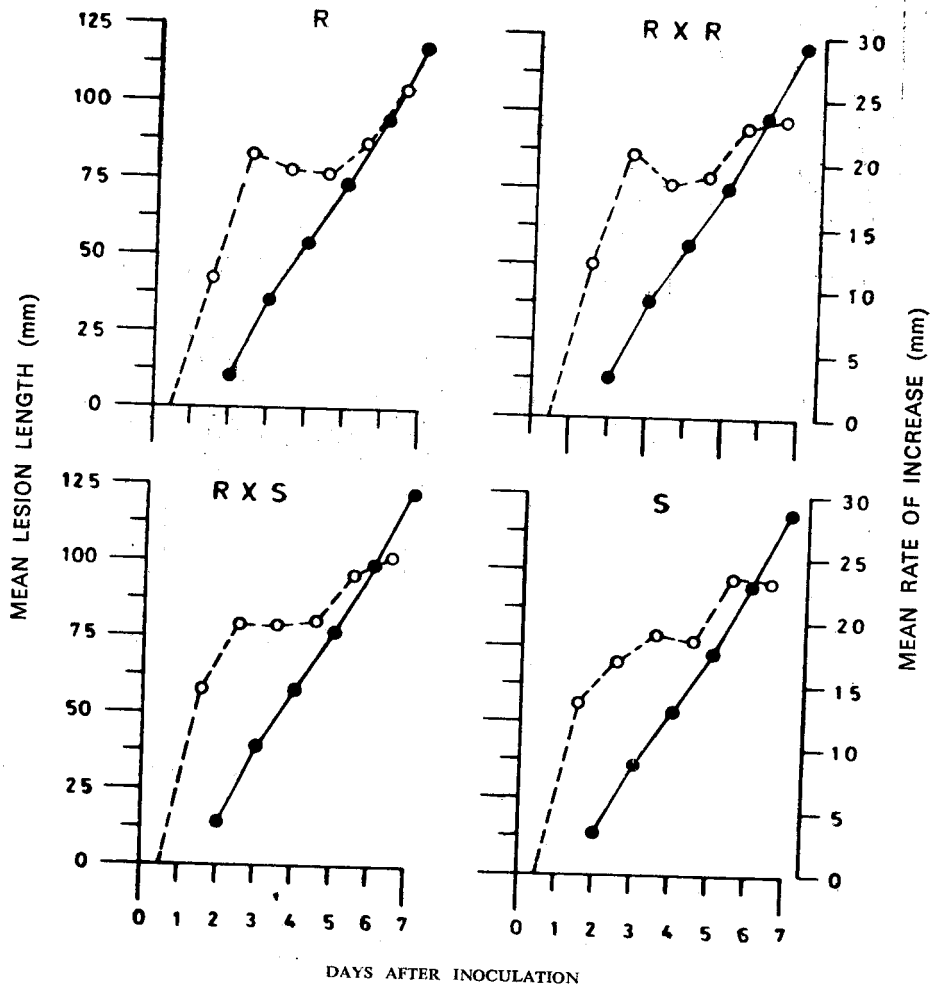


Fig. 3.4 Effect of host susceptibility rating (R—resistant, S—susceptible) on the pattern of black pod lesion development ●—● Mean lesion length, ○—○ Mean rate of increase

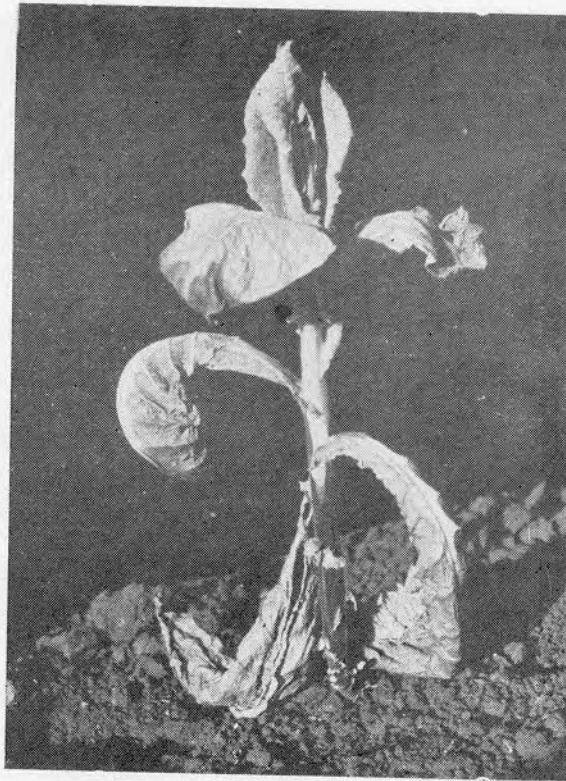


Fig. 4.1 Tobacco blight

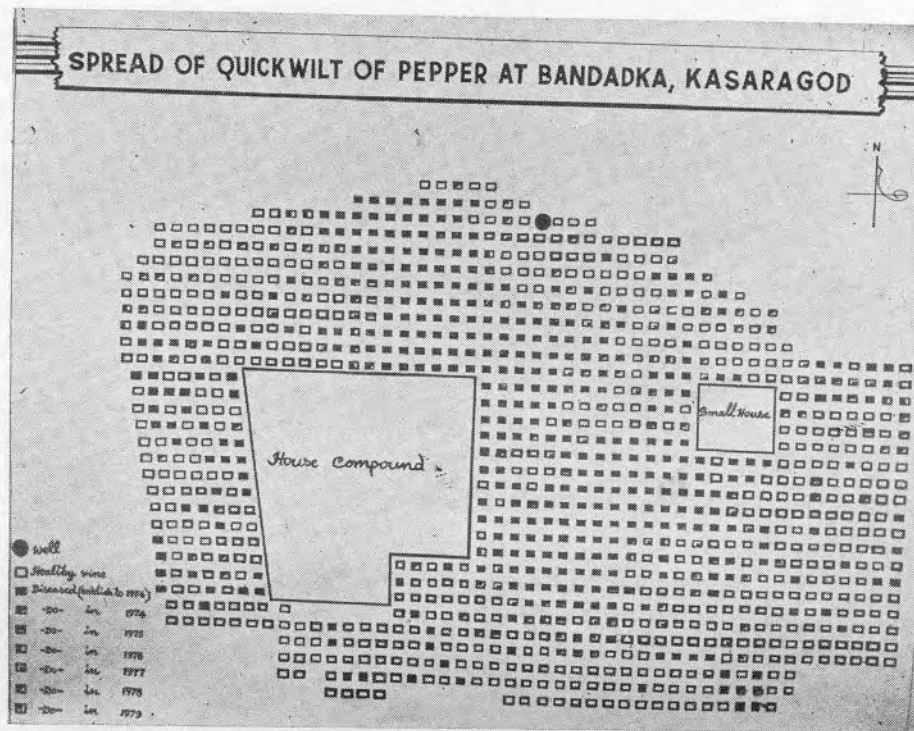




Fig. 5.3 Sporangia of pepper *Phytophthora*

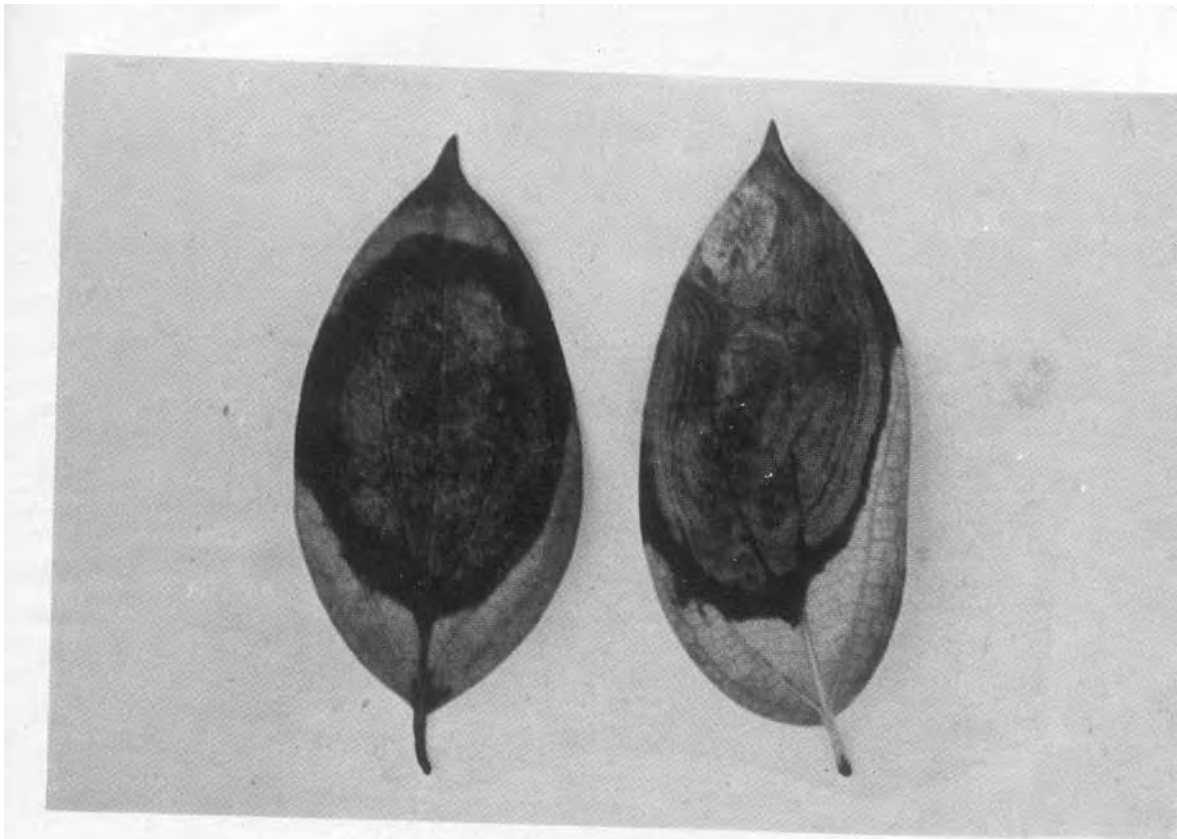


Fig. 5.1 Foliar symptoms of pepper foot rot



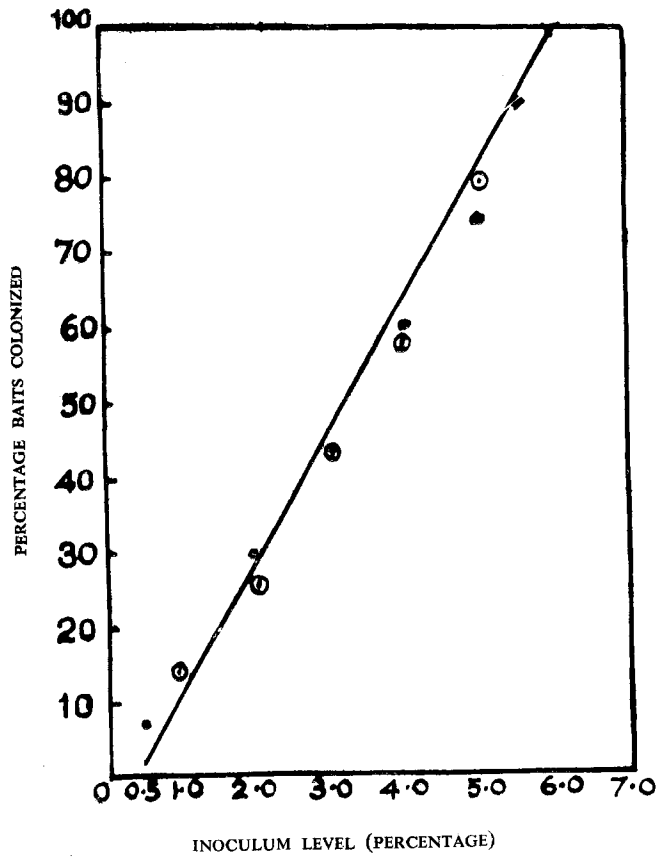


Fig. 5.4 The relationship of percentage of baits colonized to inoculum level

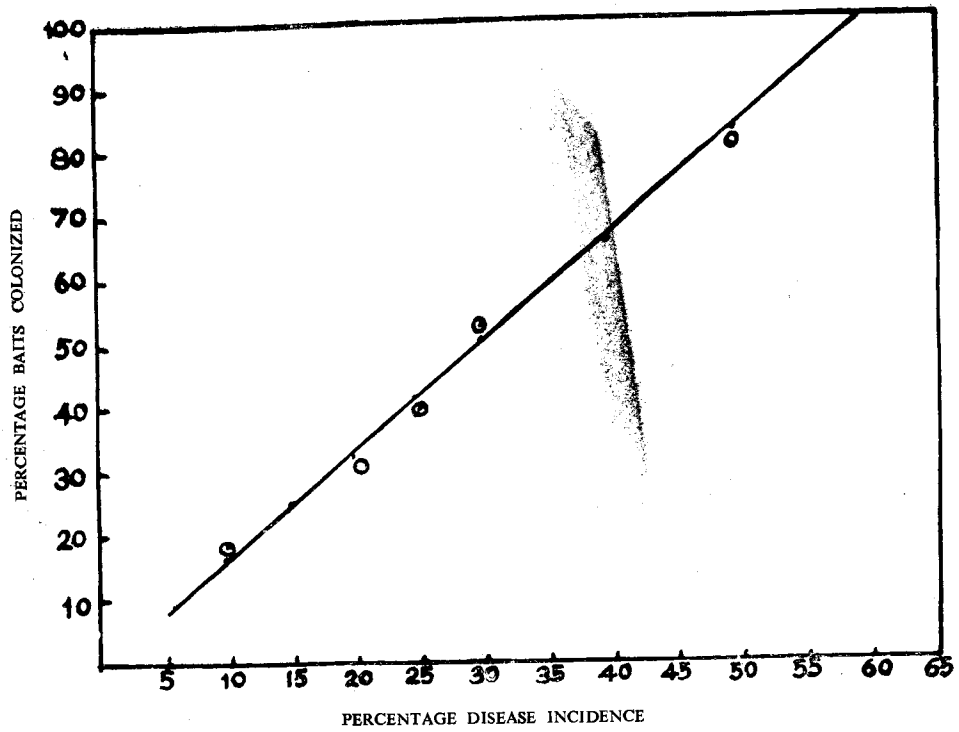


Fig. 5.5 The relationship of percentage of disease incidence to the baits colonised

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